

Review

Biomolecule-Nanomaterial Hybrid Systems: Chemical Design, Assembly, and Function

Sandhya Savita¹, Sanmatee Bai¹, Archana Markam², Ashish Singh¹, Pallab Bhandari¹, Sunil Kumar Singh¹, Suryabhan Singh¹, Ashish Kumar Singh¹, Uday Pratap Azad^{1,*}, Bijnaneswar Mondal^{1,*} and Bharat Lal Sahu^{1,*}

¹ Department of Chemistry, Guru Ghasidas Vishwavidyalaya, Bilaspur 495009, Chhattisgarh, India

² Department of Botany, Veerangana Rani Durgavati, Govt. Girls College, Takhatpur, Bilaspur 495330, Chhattisgarh, India

* Correspondence: azadchembhu@yahoo.co.in (U.P.A.); bijnaneswarm@gmail.com (B.M.); bharatred007@gmail.com (B.L.S.)

How To Cite: Savita, S.; Bai, S.; Markam, A.; et al. Biomolecule-Nanomaterial Hybrid Systems: Chemical Design, Assembly, and Function. *Advances in Applied Biomaterials and Biocomposites* **2026**, *1*(1), 6.

Received: 15 April 2026

Revised: 3 August 2026

Accepted: 11 August 2026

Published: 20 August 2026

Abstract: The interface of biomolecular and nanomaterials has emerged as one of the most appealing areas of focus of research in nanochemistry, materials science and biotechnology. The review gives a detailed overview of the key principles that can regulate the form and behaviour of interfaces between biomolecules and nanomaterials with particular attention to chemical design, assembly, and structure-function relations. The major interaction mechanisms, such as covalent bonding, electrostatic interaction, π - π stacking, hydrogen bonding, and metal ligand coordination, are discussed, where they can play a role in the stability as well as functionality of the interface. Different types of biomolecules (proteins, nucleic acids, peptides and polysaccharides) are studied, and the key nanomaterial platforms are met (metal nanoparticles, carbon-based nanomaterials, semiconductor quantum dots and metal-organic frameworks). The review also addresses interface engineering strategies based on functionalizing surfaces, bio-templating, self-assembling, and stimulus-responsive design. Such complex methods of characterization to investigate nano-bio interfaces and explain their structural and electronic characteristics are also mentioned. Notably, the interface structure/functional performance spectrum of the application is studied in biosensing and nanozyme catalysis, as well as targeted drug delivery and environmental or energy technologies. Lastly, the issues of the complexity of interface, stability, reproducibility, and scalability are considered, and an outlook of the issues in the future is provided with a focus on the role of computational modelling and artificial intelligence to support rational design of the next generation biomolecule-nanomaterial systems.

Keywords: biomolecule; nanomaterials; biosensing; biointerfaces; self-assembly; interfaces

1. Introduction

Nanotechnology, chemistry, and molecular biology have now joined forces and established biomolecule-nanomaterial interfaces an exciting new field of research with numerous emerging applications in biotechnology, nanomedicine, catalysis, environmental science and advanced materials [1,2]. The hybrid combination of the molecular recognition, catalytic activity and structural specificity of biomolecules and the exceptional optical, electronic, catalytic and physicochemical properties of nanomaterials lead to systems with properties that are not possible with the biomolecules themselves or with the nanomaterials alone [3–5]. These interfaces have led to the



Copyright: © 2026 by the authors. This is an open access article under the terms and conditions of the Creative Commons Attribution (CC BY) license (<https://creativecommons.org/licenses/by/4.0/>).

Publisher's Note: Scilight stays neutral with regard to jurisdictional claims in published maps and institutional affiliations.

creation of highly sensitive biosensors, efficient nanozymes, targeted drug delivery systems, bioelectronic devices, and multifunctional nanoplatforms for environmental and energy applications [6–8].

The chemical interactions between biomolecules and nanomaterials at the interphase are crucial for the performance of biomolecule–nanomaterial hybrid systems [9–11]. Covalent conjugation, electrostatic interactions, hydrogen bonding, π – π stacking, hydrophobic interactions, and metal–ligand coordination are all critical for the biomolecular adsorption, orientation, conformational stability, electron transfer, and biological recognition at the nano–bio interface [12–17]. Therefore, rational control of these interactions is essential to develop high stability, selectivity, reproducibility and functional performance interfaces.

The ability to design well-defined and programmable nano–bio interfaces has been greatly enhanced by the recent developments in nanomaterial synthesis, surface engineering, supramolecular chemistry and biomolecular self-assembly [18–20]. Meanwhile, new characterization methods, computational modelling, and the use of artificial intelligence (AI) in materials design have provided unprecedented insights into interfacial interactions and enabled the prediction and optimization of hybrid systems [21–24]. While these accomplishments are remarkable, there are issues like heterogeneity at the interface, dynamic formation of protein corona, preservation of biomolecular activity, reproducibility in biological systems, scalability, and translation of laboratory studies into real-world applications [5,25–28].

There have been numerous reviews on each component of the protein corona, biosensing, nanozymes, or drug delivery, but few reviews have integrated the fundamental chemistry of interfacing formation, assembly mechanisms, structure–function relationships, advanced characterization and emerging computational strategies in an integrated way [18,21,28]. The limited understanding restricts the ability to elucidate the effect of the interface engineering on the molecular level in different application domains on the performance of biomolecule/nanomaterial systems [5,24].

In this context, the present review aims to provide a comprehensive and critical overview of the chemical principles of biomolecule–nanomaterial interfaces. It includes the fundamental interaction mechanisms, the classes of biomolecules, the most important nanomaterial platforms, approaches to interface engineering, and advanced characterization methods, before examining the various applications of nanomaterials in biosensing, catalysis, nanomedicine, environmental remediation and energy devices. Finally, the current challenges and future directions are discussed with a focus on the rational development of next-generation biomolecule–nanomaterial hybrid systems by predictive computational modelling, machine learning and AI-assisted interface engineering.

1.1. Significance of Biomolecule–Nanomaterial Interfaces in Nanochemistry

Interfaces between biomolecules and nanomaterials occupy a central role in nanochemistry because they enable the integration of biological functionality and engineered nanostructures. Biomolecules are identified as recognition components, catalytic components or structural templates in this interface and physical support and enhanced physicochemical properties are present in nanomaterials. These hybrid systems exhibit properties and functions that neither component can achieve independently.

The significance of these interfaces is associated with the special size adaptation between the nanomaterials and the biological macromolecules. Nanomaterials with dimensions comparable to those of enzymes, strands of DNA or protein complexes exhibit highly selective interactions, and provide the opportunity to develop hierarchical assemblies of nanoscale [1,29–31]. Such structural compatibility enables precise control over biological functions, and also influencing cellular pathways, and facilitating the targeted delivery of therapeutic molecules.

In addition to this, the incorporation of biomolecules into nanostructured materials has led to the development of bioinspired nanomaterials that replicate the functionality of the biological system and also provide system strength, as in the synthetic systems. Nanozyme system nanomaterials with an enzyme-like catalytic activity are, as an example, illustrations of how inorganic nanostructures can mimic the action of biological catalysts, as well as while being more stable and easier to manipulate [32–37]. These technologies demonstrate how nano–bio interfaces are becoming a revolution in the advancement of modern nanochemistry.

1.2. From Bioinspiration to Chemical Engineering of Interfaces

Since the development of such materials began, biological systems have been used to inspire advanced materials, molecular self-assembly, phase separation, and biomolecular recognition. For examples: Molecular self-assembly, phase separation, and biomolecular recognition are natural processes that provide blueprints for the fabrication of functional nanomaterials with a precise structural organization. An example is that non-covalent (e.g., hydrogen bonding, hydrophobic interactions, electrostatic forces) biomolecular self-assembly can be employed to form biologically highly ordered structures like lipid bilayers or protein complexes spontaneously [38–41].

Researchers have developed chemical engineering strategies to precisely control the formation and function of biomolecule–nanomaterial interfaces. A surface property of nanomaterials is readily manipulable and therefore, selective surface biomolecule binding and orienting is possible via surface functionalization, ligand engineering and molecular templating techniques. In addition, more innovative concepts such as liquid-liquid phase separation (LLPS) provide new insights into how biomolecules assemble to form dynamic condensates that can be employed in conjunction with nanoparticles to generate responsive nanomaterials [42–47].

Such bioinspired engineering processes have significantly expanded the scope of nanotechnology in such a way that adaptable materials that can respond to environmental stimuli may be prepared. These strategies have enabled the development of drug delivery systems, responsive hydrogel, catalytic nanostructures and programmable nanodevices.

1.3. Scope and Structure of the Review

The research in the field of nano-bio interfaces has accelerated, creating need for an integrated design framework to connect the chemical design, assembly mechanisms and functional performance [9]. This review is centred on this need, covering the basics of the chemistry of biomolecule–nanomaterial interfaces, the covalent and noncovalent bonding, coordination chemistry, as well as the functionalization of surfaces and biomolecule-directed assembly strategies [5,24,38]. The review does not aim to provide a comprehensive overview of just one application field, but rather, provides a common basis around which to tie together the principles of interfacial design with the behaviour and performance of hybrid systems in biosensing, catalysis, nanomedicine, and environmental technologies.

It is important to note that in comparison with the previous reviews, which concentrated only on specific aspects of interface engineering—such as the formation of protein corona, the development of nanozymes, biosensors or drug delivery—this work is relevant to a wide range of areas related to the control of catalytic activity, charge transfer, optical properties, biorecognition and structural stability [9,11,38]. The review also critically overviews the current limitations such as interface heterogeneity, reproducibility, toxicity and challenges related to transfer of laboratory-based discoveries to the practical application. Furthermore, future directions of the rational design of new hybrid materials like predictive computational modelling, interface engineering with adaptive nano–bio architectures and the use of artificial intelligence (AI) are highlighted [17,31,47].

The review is organized so as to provide a logical flow beginning with the fundamental chemistry of biomolecule–nanomaterial interactions, then moving from biomolecular building blocks to nanomaterial platforms, interface engineering, advanced characterization methods and to the illustrative applications. The final sections of the review look forward to the current issues and future directions, provide a meaningful connection between the molecular-level design of interfaces and functional performance, and lead the reader to the design of stable, selective, and application-specific biomolecule–nanomaterial systems.

1.4. Review Methodology

This review is based on a structured literature survey of peer-reviewed publications on biomolecule–nanomaterial interfaces. Major scientific databases such as Web of Science, Scopus, PubMed and Google Scholar were mainly used for literature search. Common search terms included nanomaterial–biomolecule interfaces, nanomaterial–biomolecule interactions, surface functionalization, protein corona, nanomaterial catalysis by enzymes, biosensing, drug delivery, MOFs (Metal-organic frameworks)/COFs (Covalent organic frameworks) biointerfaces, and the use of AI in nanomaterial design. The focus was on influential foundational studies and recent publications, primarily from the past 10–15 years, that made significant advances in understanding the mechanics, and strategies for interface engineering, characterization techniques, and novel uses. Articles were selected based on the following criteria: peer review, scientific rigor, conceptual relevance, and citation impact, within the field of nanochemistry, materials science and biointerface engineering. This is a critical narrative review not a systematic review, so the goal was to represent key developments to key challenges and future directions, rather than to provide an exhaustive literature listing [13,31,38].

2. Fundamental Chemistry of Biomolecule–Nanomaterial Interactions

The nature of chemical interactions between biomolecules and nanomaterials at their interface is the core determinants of the functionality as well as the stability of hybrids. These responses characterize the process of biomolecular binding to surfaces of nanomaterials, structural orientation, and eventually the control of physicochemical and biological properties of the assemblies formed. At the nano-scale interfaces are governed by a mixture of covalent bonding, coordination chemistry, electrostatic attraction, hydrogen bonding, π – π stacking,

hydrophobic interactions and van der Waals forces, all of which are jointly involved in the formation of stable nano-bio conjugates [48].

The high surface-to-volume ratio and tunable surface chemistry of nanomaterials enable precise engineering of nano-bio interfaces through interactions with biomolecules. The nano-bio interfaces resulting are not only able to regulate the biomolecular conformation and functionality of the nano-material but also control the nanomaterial properties such as catalytic efficiency, optical response and biocompatibility [9–12,49]. The basic chemistry of such interactions is therefore essential in the rational design of nanomaterials that can be used in biosensing, nanomedicine, catalysis, and environmental technology.

2.1. Covalent and Noncovalent Binding Mechanisms

Biomolecules can react with nanomaterial surfaces through covalent or noncovalent bonding mechanism, and the advantages of each approach differ depending on the intended use. Covalent conjugation involves the formation of chemical bonds between functional groups on biomolecules and the reactive moieties on the surfaces of nanomaterials, resulting in stable biomolecule–nanomaterial conjugates. They include these common methods, which are amide coupling, thiol-gold bonding, click chemistry and carbodiimide-mediated reactions. The techniques enable the stable immobilization of biomolecules while preserving their structural orientation and biological function. In particular, covalent modification may be particularly useful in biosensors, catalysts, and nanomedicine systems in cases when long-term stability and resistance to changes in the environment are required [50].

In comparison, noncovalent modification gives reversible and dynamic binding of biomolecules with nanomaterials. Such interactions are mediated by weaker intermolecular forces that include electrostatic attraction, hydrophobic, hydrogen bonding and π - π stacking. Noncovalent assembly is common in nature and is commonly used to make biomolecule-nanomaterial hybrids by using self-assembly. These kinds of interactions allow the creation of responsive systems that are able to adjust to the changes in the environment, at the same time, without functional breakdown [38–40,51]. The appropriateness of a particular interaction mode depends greatly on the goal of the function and the environment it's used in. In biosensing platforms, catalytic systems, and therapeutic formulations with long operational stability, resistance to desorption and molecular orientation, covalent conjugation is generally preferred. Irreversible attachment can, however, hinder the biomolecular flexibility, or even biological activity. The π - π assembly, on the other hand, is beneficial for reversible, stimulus-responsive, and self-assembled systems, but its efficiency may decrease under changing ionic strength or pH values that disrupt the π - π interactions. Electrostatic and hydrogen-bond-mediated assemblies are even more useful for systems that are reversible, stimulus-responsive, and self-assembled, but in which the performance may suffer under changing ionic strength or pH conditions that break the electrostatic or hydrogen bond interactions. The differences illustrate how interface chemistry can affect binding ability, responsiveness, stability, electronic communication and even functionality [1,5,31,48].

Notably, the noncovalent interactions in biological systems tend to prevail in the development of biomolecular coronas around nanoparticles, and noncovalent interactions are common in this environment [52]. The presence of adsorbed proteins, nucleic acids, and other biomolecules that dynamically bind to the nanomaterial surfaces constitute the biomolecular corona, and they change their biological identity as well as cellular interactions and biodistribution [32,53,54].

While both covalent and noncovalent strategies have been extensively exploited in biomolecule–nanomaterial interface engineering, the former or the latter perform better in different applications depending on the nanomaterials involved. This is because covalent conjugation offers better interfacial stability, controlled immobilization and resistance to environmental changes, which is beneficial for the applications of long-term biosensing, catalysis, and therapeutic systems. But irreversible coupling can cause changes in biomolecules conformation or can decrease biological activity. Noncovalent assembly, on the other hand, can maintain native biomolecular structures more faithfully and can achieve stimuli responsive behavior in the system, although its binding may be weaker, making it more susceptible to desorption, and is less reproducible under physiological conditions. This trade-off is one of the key challenges in interface engineering and hybrid approaches based on reversible molecular recognition seem to be attractive solutions [5,9,42,53].

The major interaction mechanisms governing biomolecule–nanomaterial interfaces are schematically illustrated in Figure 1.

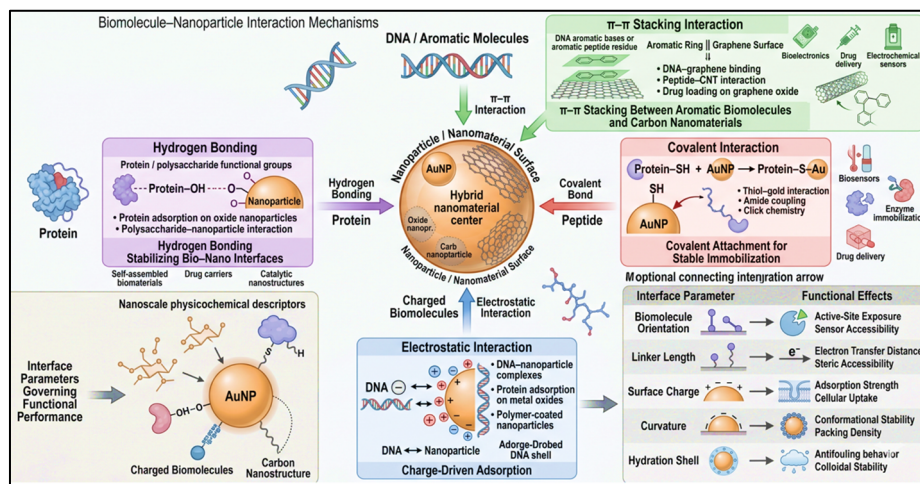


Figure 1. Mechanisms of biomolecule–nanomaterial interactions with covalent bonding, electrostatic attractions, hydrogen bonding, π - π stacking and coordination chemistry. The scheme also emphasizes parameters of interface design, such as biomolecule orientation, linker length, surface charge, nanomaterial curvature, and organization of the hydration layer, as well as their influence on the stability of the binding, electron transfer, catalytic accessibility, response to biosensing, and therapeutic efficacy.

2.2. Coordination Chemistry and Metal–Ligand Interactions

In several biomolecule–nanomaterial interfaces, coordination chemistry is a significant factor, especially when metal–metallic nanomaterials, such as gold, silver, platinum, or metal oxides, are used. The systems allow metal to have coordination bonds with metal atoms on the nanomaterial surface and functional groups on the biomolecules, such as thiols, amines, imidazoles, and carboxylates.

The notable example is that of a strong affinity between thiols and the nanoparticles of the noble metals, used particularly on gold nanoparticles. It is possible to use Au–S bonding to immobilize proteins, peptides and DNA strands onto gold nanostructures to generate biosensors, imaging probes and drug delivery systems. The nanomaterials based on noble metals are particularly useful because the surface of the materials can be created with a high level of precision and yet remain chemically stable and have either a great optical or a great catalytic property [9,55–59]. Enzyme–nanoparticle hybrids and the nanozyme systems, where the metal ion or metal surface is a ligand of biomolecules, also play an important role in controlling catalysis. These coordination interactions determine electron transfer pathways, the surface reactivity and accessibility to substrates, which enhances the functionality of hybrid catalytic systems.

2.3. Electrostatic, π - π , and Hydrogen-Bonding Interactions

Besides covalent and coordination interactions, there are several noncovalent forces that play an important role in the formation and stability of biomolecule–nanomaterial interfaces. Electrostatic interactions are the result of attraction between oppositely charged species, like the positively charged surface of nanoparticles and the negatively charged biomolecules, e.g., proteins or DNA. These forces play significant roles, especially in aqueous biological systems where ionic strength and pH have a role in modifying the surface charge of both biomolecules and nanomaterials. Electrostatic adsorption is commonly the first force behind biomolecule binding to the nanomaterial surfaces.

π - π stacking interactions take place when aromatic biomolecules react with nanomaterials that contain conjugated carbon frameworks, e.g., graphene, carbon nanotubes and other carbon-based nanomaterials. The aromatic rings of the nucleic acids, peptides and small biomolecules can planarize with the delocalized systems of the π -electron of the materials, thus providing strong adsorption as well as efficient electronic coupling. These are the interactions that are prolifically used in biosensing, electronic nanodevices, and carbon nanomaterial [60–66].

Biomolecule–nanomaterial assemblies' stability is also due to hydrogen bonding. The biomolecules and functional groups on the surfaces of nanomaterials are hydrogen-bonded between the donor and acceptor groups and take part in the molecular recognition and structuring. With Hydrogen bonding interactions being weaker on an individual basis than covalent bonds, they provide a significant degree of stabilization and specificity to many nano–bio systems. The following tables summarize not only representative systems and applications, but also comparative performance data such as interaction reversibility, environmental sensitivity, stability, characterization approaches, limitations and translational relevance. With these various interface chemistries having different strengths, reversibility, sensitivity to environment, and functional consequences, an overview is provided in Table 1.

Table 1. Comparative analysis of chemical interactions governing biomolecule–nanomaterial interfaces.

Interaction Type	Chemical Principle	Binding Strength	Stability/Reversibility	pH/Ionic Sensitivity	Typical Biomolecules	Typical Nanomaterials	Typical Characterization Methods	Major Advantages	Main Limitations/Denaturation Risk	Suitability for Enzyme Immobilization	Key Applications	Application Readiness	Ref.
Covalent bonding	Stable chemical linkage formation (Au–S, amide, click chemistry)	High	High/Often irreversible	Generally low	Proteins, DNA, peptides	AuNPs, polymers, MOFs	FTIR, XPS, NMR, SPR	Strong attachment, excellent reproducibility, long-term stability	Possible loss of biomolecular flexibility or active-site blockage; moderate denaturation risk	Excellent	Biosensors, drug delivery, enzyme immobilization	High (widely used)	[5,50]
Coordination chemistry	Metal–ligand complexation (thiol, amine, carboxyl coordination)	Moderate–High	Moderate–High/Partially reversible	Moderate	Amino acids, peptides, proteins	Metal nanoparticles, metal oxides, MOFs	XPS, UV–Vis, ITC, Raman	Tunable interactions, catalytic compatibility, controllable binding	Metal-dependent stability; potential conformational perturbation	Good	Catalysis, nanozymes, biofunctional materials	High	[55]
Electrostatic interactions	Coulombic attraction between oppositely charged surfaces	Moderate	Moderate/Reversible	High sensitivity to pH and ionic strength	DNA, proteins, polysaccharides	Metal oxides, polymer-coated nanoparticles	Zeta potential, DLS, SPR	Simple assembly, reversible binding, mild conditions	Weak under physiological ionic environments; nonspecific adsorption; possible aggregation	Moderate	Gene delivery, biosensing, nanoparticle assembly	High	[1,11]
π – π stacking	Interaction between aromatic π -electron systems	Moderate	Moderate/Reversible	Low–Moderate	DNA bases, aromatic peptides, biomolecules	Graphene, CNTs, carbon dots	Raman/SERS, UV–Vis, AFM	Strong compatibility with carbon materials; efficient charge transfer	Limited applicability to aromatic systems; may alter biomolecular conformation	Moderate	Bioelectronics, sensing, drug delivery	Moderate–High	[60]
Hydrogen bonding	Directional donor–acceptor interactions between functional groups	Low–Moderate	Moderate/Reversible	High sensitivity to solvent and pH	Proteins, polysaccharides, peptides	Metal oxides, polymer nanomaterials	FTIR, NMR, ITC	High selectivity, biomimetic recognition, self-assembly capability	Relatively weak interactions; environmental instability; lower structural robustness	Moderate	Self-assembly, stabilization, drug carriers	Moderate	[38]
Hydrophobic interactions	Association of nonpolar domains in aqueous media	Moderate	Moderate/Reversible	Moderate	Proteins, peptides, lipids	Carbon nanomaterials, polymer nanoparticles	Fluorescence, CD, DLS	Efficient adsorption, useful for carrier loading and self-organization	Can induce unfolding, aggregation, or biomolecule denaturation; lower specificity	Limited–Moderate	Drug delivery, protein adsorption	High	[1,40]

All electrostatic interactions, π - π interactions and hydrogen bonding interactions are widely used in nano-bio interfaces, but are not functionally equivalent. Electrostatic interactions are often used for fast biomolecule adsorption and gene-delivery systems due to their tight dependence on surface charge; however, ionic screening effects and environmental conditions make them sensitive and cause destabilisation [5,24–26]. Hydrogen bonding gives molecular specificity and plays a significant role in self-assembly and organization of molecules, but is not usually strong enough for use in extreme operational environments. Conversely, π - π interactions can be very beneficial in graphene and carbon nanotube interfaces where the strong aromatic coupling can facilitate charge transfer and signal amplification in bioelectronic interfaces. Thus, the choice of interaction strategy depends on the sort of stability, reversibility, specificity and electronic functionality desired for a particular application [9,48,55].

2.4. Thermodynamics and Kinetics of Interface Formation

The interfaces between biomolecules and nanomaterials are predetermined, that is, they occur by thermodynamic as well as kinetic reasons. Minimization of the free energy is the thermodynamic basis of an assembly process because of the positive enthalpic terms of the chemical bonding of the molecules and intermolecular interactions versus entropic penalties of the molecular order. Surface properties, including particle dimension, shape, surface charge and chemical functionalization, are major factors leading to adsorption and assembly on the nanoscale. The minor variations in the size of the nanomaterials or the surface chemistry lead to the interaction energy that could differ significantly and therefore lead to different biological reactions [55,57,59,67]. Thermodynamics of biomolecule-nanomaterial association can be assessed based on changes in Gibbs free energy, which indicates the spontaneity of the formation of the interface:

$$\Delta G = \Delta H - T\Delta S$$

where ΔG is Gibbs free energy change, ΔH is enthalpic contribution due to bonding, electrostatic interactions, or the process of surface adsorption, T is temperature and ΔS is the entropy change due to the organization of solvent molecules, conformational change, or the process of adsorption. Usually, ΔG values are negative for thermodynamically favourable interface formations [1,5,31]. Binding affinity between biomolecules and nanomaterials is frequently related to the equilibrium constant K through:

$$\Delta G = -RT \ln K$$

where R is the gas constant and T is absolute temperature. Larger binding constants correspond to stronger and more stable nano-bio interactions. Surface adsorption of biomolecules on the interface of nanomaterials is frequently modeled by Langmuir adsorption model in which the biomolecules are assumed to be adsorbed as a monolayer onto a finite number of binding sites:

$$q_e = \frac{q_{max} K_L C_e}{1 + K_L C_e}$$

The terms q_e (equilibrium adsorption capacity), q_{max} (maximum adsorption capacity), K_L (Langmuir affinity constant), and C_e (equilibrium concentration) are used. These models can be employed to study protein adsorption, enzyme immobilization and ligand-binding processes at nano-bio interfaces. Pseudo-first-order kinetic models are often used to describe adsorption and immobilization kinetics especially for the initial stages of adsorption [4,14,31].

$$\ln(q_e - q_t) = \ln q_e - k_1 t$$

where: q_e = equilibrium adsorption capacity (mg g⁻¹ or equivalent units), q_t = adsorption capacity at time t , k_1 = pseudo-first-order rate constant (min⁻¹), t = adsorption/contact time

The pseudo-second-order kinetic is often used to describe chemisorption-dominated nano-bio interactions, such as electron sharing, covalent coupling or coordination processes, involving 75%.

$$\frac{t}{q_t} = \frac{1}{k_2 q_e^2} + \frac{t}{q_e}$$

where: q_t = adsorption capacity at time t , q_e = equilibrium adsorption capacity, k_2 = pseudo-second-order rate constant (g mg⁻¹ min⁻¹ or corresponding units), t = adsorption time

Mechanistic problems which have not yet been solved are related to dynamic evolution of biomolecular coronas in complex biological environment. Though early studies focused mainly on protein corona formation as a passive adsorption process, recent findings indicate that protein corona formation is a very dynamic and

competitive exchange between biomolecules, surface ligands and environmental elements. This time-dependent evolution can significantly impact on the identity, biodistribution, immune recognition and catalytic properties of nanoparticles. Importantly, however, certain differences between these simplified in vitro experiments and biologically complex in vivo systems remain that make understanding the interactions between nanoparticles and living cells difficult and therefore make it important that the experiments are all done using the same design and in vivo characterization methods are available in real time [15–17,31].

The mechanism of the nano-bio interface formation may be subdivided into a sequence of kinetically distinct biological diffusion, surface adsorption, conformational rearrangement, and structural stabilization. These are highly dynamic processes in biological systems, often resulting in complex and evolving protein coronas or multilayer biomolecular assemblies. The balance of composition, structure and functionality of the interface ultimately is defined by the stability in thermodynamics and the accessibility in kinetics. Surface-engineering decisions have a strong influence on surface stability and biological identity. For instance, PEGylation is extensively used to create steric stabilization and the formation of hydration-layer for the prevention of nonspecific protein adsorption, thus decreasing the uncontrolled development of protein-corona and increasing the stability of the colloidal system in biological systems. Nevertheless, the target recognition, cellular uptake or catalytic accessibility can be decreased in case of too much surface shielding. The trade-offs demonstrate the direct influence of intentional modification of the surface chemistry on the biological interactions and function [1,9,38,55].

This knowledge of these thermodynamic and kinetic principles allows one to be able to engineer rationally nano-bio interfaces with custom properties. Researchers can rationally engineer biomolecule–nanomaterial interfaces by controlling surface chemistry, assembly pathways, and environmental conditions to find applications in nanomedicine and catalysis, as well as environmental remediation and biosensing. Table 1. Summary of fundamental chemical interactions governing biomolecule–nanomaterial interfaces, including covalent bonding, coordination chemistry, electrostatic interactions, π – π stacking, and hydrogen bonding. The table outlines the underlying chemical principles, typical biomolecule–nanomaterial systems, and their relevance to functional applications. The use of thermodynamic, kinetic, adsorption, catalytic and charge-transfer models is common in the quantitative interpretation of biomolecule–nanomaterial interactions. These equations are complementary to information on the spontaneity of binding, the adsorption behaviour, the catalytic performance, and the electron-transfer reaction processes at the interfaces of interest for nano-bio system design and characterization [5,31,32]. A variety of representative equations are often used in nano-bio interface analysis, which are described in Table 2.

Table 2. Representative equations used in biomolecule–nanomaterial thermodynamic and kinetic analysis.

Equation	Application	Information Obtained	References
$\Delta G = \Delta H - T\Delta S$	Thermodynamics	Spontaneity and energetic driving forces of biomolecule–nanomaterial binding	[1,5,31]
$\Delta G = -RT \ln K$	Binding affinity	Equilibrium binding strength and interaction stability	[1,9,57]
Langmuir isotherm	Adsorption analysis	Surface adsorption capacity, monolayer binding behaviour, and affinity constants	[4,51,59]
Pseudo-second-order kinetic model	Adsorption kinetics	Adsorption rate mechanism and chemisorption behaviour	[4,13,57]
Michaelis–Menten equation ($v = V_{max}[S]/(K_m + [S])$)	Nanozymes	Catalytic efficiency, substrate affinity (K_m), and maximum reaction velocity (V_{max})	[6,48,56]
$k_{ET} \propto e^{-\beta d}$	Bioelectronics	Distance-dependent charge-transfer and electron tunneling behavior at bio–nano interfaces	[18,20]

3. Classes of Biomolecules for Interface Engineering

The development of functional biomolecule nanomaterial interfaces depends on the choice of suitable biomolecular components that can be easily coupled with nanomaterial surfaces. Biomolecules have a variety of chemical functionalities, structures, and recognition of their own that can be utilized as building blocks, templates, stabilizing agents, and functional components in nano-bio hybrid systems. These molecules are able to regulate the assembly of nanomaterials, surface chemistry and incorporate biological functionality, including catalysis, molecular recognition, and signal transduction.

Proteins, nucleic acids, peptides, polysaccharides, and other small biomolecules are some of the most popular biological components to be used as interfaces on different surfaces. Each of these classes has distinct physicochemical features, allowing a certain form of interaction with nanomaterials. Biomolecular functionality coupled to nanomaterial physicochemical properties can be combined to create hybrid materials with increased stability, selectivity and performance in biosensing, catalysis, nanomedicine and environmental technologies

applications [5,6,68–74]. Biomolecule–nanomaterial interfaces can be broadly classified based on the nature of biomolecular components and nanomaterial platforms, as illustrated in Figure 2.

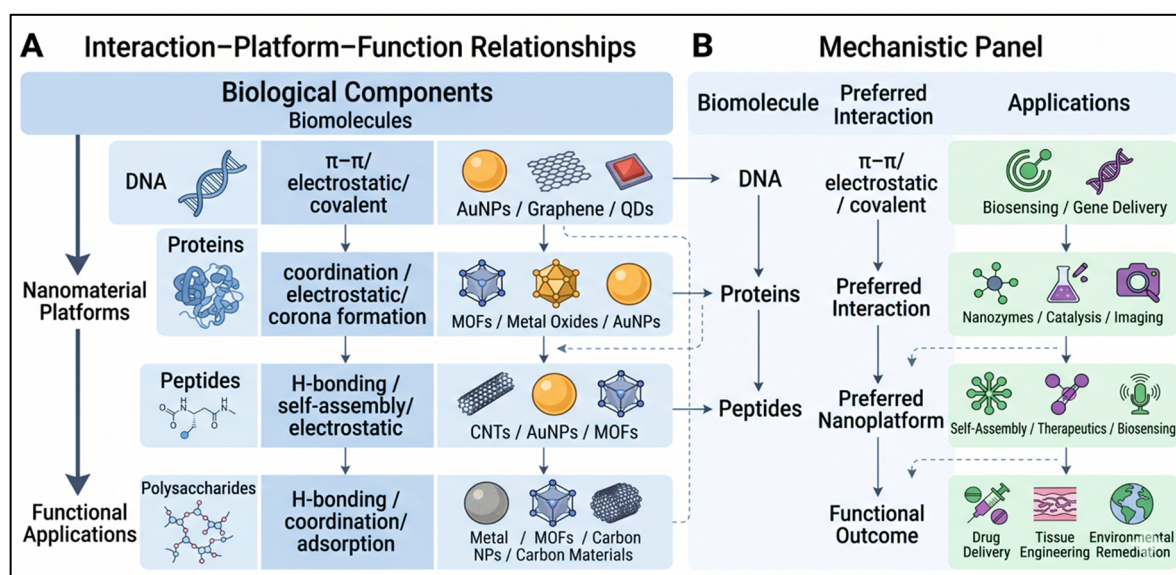


Figure 2. Biomolecule–Nanomaterial interfaces classification framework showing the relationship between biomolecule class, nanomaterial platform, dominant interaction mechanism and representative functional outcomes in biosensing, catalysis, drug delivery, and bioengineering.

3.1. Proteins and Enzymes

Among the most versatile biomolecules in nanomaterial interface engineering are proteins and enzymes, which have complex three-dimensional structures, catalytic activity, and molecular recognition properties. Proteins have diverse functional groups such as amines, carboxylates, thiols and aromatic residues, which enable them to interact with nanomaterial surfaces with a high level of covalent bonding, electrostatic attraction, hydrogen bonding, and hydrophobic forces [75–77].

Enzymes immobilized on nanomaterial surfaces can retain or even improve their catalytic activity and are therefore of much interest in biosensor and, bio fuel cell applications, and also in catalytic nanoreactors [78]. Nanomaterials are commonly used in the form of supports that enhance the stability of enzymes, electron transfer, and increase the effective surface area on which substrates react with enzymes. Especially, metal nanoparticles and carbon-based nanomaterials have been extensively applied for enzyme immobilization in electrochemical sensing and catalytic studies [5–7,71,72].

In addition to adsorption, the orientation, conformational stability, and accessibility of protein active-sites on nanomaterials are significant factors that determine functional performance of the protein–nanomaterial interfaces. When proteins are randomly adsorbed, the surface population is often heterogeneous, with catalytic sites being partially blocked or with structural unfolding that decreases the biological activity of the surface population. Conversely, controlled immobilization approaches like site specific conjugation with engineered cysteine residues, affinity tags or selective functional handles can retain native orientation and enzymatic accessibility. Thus, the compromise between the strong bond and structural maintenance are major design concern in biosensors, nanozymes and therapeutic nanoplatforms. Additionally, protein–nanomaterial interactions are regulated by surface modification strategies including PEGylation and linker engineering, which are also critical for this research work. PEGylation can enhance colloidal stability, decrease non-specific adsorption, and block immune recognition; however, extensive shielding can result in decreased accessibility to the substrate and/or electron transfer efficiency. Similarly, the length of the spacer/linker molecule also affects the steric accessibility, flexibility of the molecule, and communication between interfaces. Short rigid linkers are better for electronic coupling but may be detrimental for biomolecule mobility; the longer the flexible spacer the better the biomolecule is shielded from the surroundings but the lower the transfer efficiency may be. Optimization of the linker architecture is thus still very system specific [5,9,38,55].

Structural changes in protein conformation may also take place, however, during interactions between proteins and nanomaterials. Protein adsorption onto nanoparticle surfaces at the nano-bio interface can cause protein unfolding, reorientation or aggregation. The effects are frequently linked to the creation of protein coronas, which have a significant impact on the stability of nanoparticles, their internalization in cells, and biological

reactions [1,4,29–31]. These interactions thus need to be understood to create functional and stable protein-nanomaterial hybrids.

3.2. Nucleic Acids (DNA and RNA)

Nucleic acids, such as DNA and RNA, are extensively used in interface engineering of nanomaterials due to their programmable structures, sequence-specific recognition and predictable hybridization properties. DNA molecules may be used as molecular templates, structural scaffolds or recognition elements that direct the self-assembly of nanomaterials in highly ordered structures.

Base-pairing is one of the strongest properties of nucleic acids that allows accurate molecular recognition and self-organization. DNA-guided assembly has been widely applied to assemble nanoparticles into ordered nanostructures, make nanoscale circuits, and develop programmable materials [79]. The functionality of nucleic acid-nanomaterial interfaces has also been extended by DNA origami and aptamer-based systems, enabling the generation of extremely complex nanoscale structures. Nucleic acids may also be utilized as biosensing units in addition to the structural functions, which allow them to sense the presence of certain biomolecules, metal ions or pathogens. High specificity and sensitivity of DNA-functionalized nanoparticles have largely been utilized in colorimetric assays, fluorescence sensing, and electrochemical detection platforms because of their excellent molecular recognition capability [5,7,70,72–74]. At interfaces between nucleic acids and nanomaterials, orientation, surface density and the attachment chemistry of the materials are factors that are critical to successful hybridization and signal generation. Favourable conformations, steric crowding or decreased target accessibility may be obtained by random adsorption of DNA strands. Therefore, controlled immobilization strategies, like sequence-directed orientation control, spacer-assisted tethering and thiolated DNA attachment, are often used to ensure high recognition efficiency and reproducibility. Spacer molecules are especially useful for electrochemical and fluorescence biosensors as steric interference can be minimized and the hybridization performance may be maintained by proper separation between the surface of the nanomaterial and the spacer molecule [21,68,72].

3.3. Peptides and Peptidomimetics

Peptides are highly versatile biomolecules for nanomaterial interface engineering in that they provide both the diversity of functionality found in proteins, as well as the simplicity of structure and synthetic flexibility of small molecules. The short peptide sequences can be designed to identify specific surfaces of nanomaterials that enables specific control of nanoparticle synthesis, stabilization and assembly. Peptides have found particular use in biomimetic nanotechnology, where peptides can be used to recapitulate interactions in nature, but provide significantly increased control over chemical composition and structure. Nanomaterial surfaces can interact with specific amino acid residues through an electrostatic interaction, hydrogen bonding, or coordination chemistry. One example is peptides containing cysteine residues that can strongly bind to metal nanoparticles via the thiol-metal interactions. Peptide-nanomaterial systems also illustrate the significance of site-selective binding and conformational control in interface engineering. The spatial arrangement and localization of the amino acid residues have a significant effect on the adsorption geometry, surface affinity and the self-assembly properties. Random peptide adsorption can help to speed up assembly, but can also create poorly-controlled interfaces with varying biological activity. Rational peptide engineering, ranging from sequence modification to terminal functionalization and the use of linkers for immobilization, can be used to create controlled orientation, modified recognition properties, as well as enhanced structural stability in catalytic, sensing, and regenerative applications [41,56,67].

Moreover, it is possible to design peptide-based molecules so that they can self-assemble as nanostructures, nanofibers, nanotubes, or hydrogels and include nanomaterials in their structures. There has been interest in these hybrid materials in the medical fields, especially in tissue engineering, drug delivery, and regenerative medicine. The synthetic molecules, which are peptidomimetics, are created to mimic peptide structures, and also have the added benefit of increased stability, enzymatic degradation resistance and increased tunability. This attributes to their potential to be valuable in designing stable biomolecule-nanomaterial interfaces.

3.4. Polysaccharides and Small Biomolecules

Polysaccharides and other small biomolecules also play important roles in biomolecule-nanomaterial interface engineering. Natural polysaccharides include chitosan, cellulose, alginate, and dextran, which contain abundant hydroxyl, amino, and carboxyl functional groups that interact intensively with the surface of nanomaterials. Through these interactions, polysaccharides can be used as stabilizing agents, surface modifiers or scaffolds of structural assemblies of nanoparticles during synthesis and assembly. Polysaccharides offer certain advantages in biomedical use, including biocompatibility, biodegradability and low toxicity, which makes them

especially appealing. They have been extensively applied in drug delivery to nanoparticles, enhancing nanoparticles' stability in biorealms, and enhancing cell and tissue interactions. Also, nanomaterials polysaccharide-functionalized have been examined in their environmental remediation, catalysis, and biosensing applications. Polysaccharide-functionalized systems are not only chemically controlled by the composition of the functional group, but also by the polymer architecture, hydration properties and surface presentation. The steric stabilization, access to biomolecules and interaction with cell environments can be modulated by the controlled grafting density and linker design. Polysaccharides are, in general, biocompatible and colloidally stable, but if they cover too much surface area, they could either hinder the accessibility of the catalytic sites or decrease the transport of analytes or therapeutic molecules. These considerations highlight the need for an operational performance for the protective functionality in designing nano–bio interfaces [9,62,63].

Nano-biomolecules like amino acids, polyphenols, and metabolites also play a role in the creation of nano–bio interfaces. Such molecules are capable of either coordinating or reducing metal ions or stabilizing surfaces, which allows green synthesis pathways to nanomaterials and improves the performance of hybrid systems. The wide range of functional group natural biomolecules is a good base to design biointerfaces with desired chemical and biological characteristics [80–88]. Table 3. Representative biomolecule–nanomaterial interface systems illustrating the interaction between different classes of biomolecules (proteins, nucleic acids, peptides, and polysaccharides) and nanomaterial platforms. The table summarizes the dominant interaction mechanisms, including covalent and noncovalent binding, and highlights their corresponding functional applications in biosensing, catalysis, drug delivery, and bioengineering.

One of the common problems for all biomolecules is the difference between random adsorption and controlled immobilization in the nano–bio interface engineering. Spontaneous adsorption strategies are experimentally simple and are widely employed, but they tend to produce interfaces with varying orientations, conformational perturbations, and decreased functional reproducibility. In contrast, controlled immobilization approaches yield more predictable interface architectures and improve the retention of activity and operational stability via site-specific conjugations, linker engineering, and selective surface chemistries. The knowledge of these design principles is crucial to convert biomolecule–nanomaterial assemblies from conceptual platforms to functional and stable systems. Regardless of the biomolecule class, a set of common immobilization and interfacial design parameters that directly impact functionality, stability and reproducibility is required for successful nano–bio interface engineering [1,4,9,68–74]. The molecular orientation, conformational preservation, structure of the linker, the surface density and the adsorption strategy are some of the most important aspects compared in Table 4.

Table 3. Comparative representative biomolecule–nanomaterial interface systems and functional characteristics.

Biomolecule	Nanomaterial	Interaction Mechanism	Binding Characteristics/Stability	Typical Characterization Method	Major Functional Advantage	Key Limitation/Risk	Representative Application	Application Readiness	Ref.
Enzymes (e.g., glucose oxidase)	Gold nanoparticles	Covalent (thiol–Au), electrostatic	High stability; covalent binding often irreversible	FTIR, SPR, electrochemistry, XPS	Efficient electron transfer, improved enzyme retention	Possible active-site blocking/orientation effects	Electrochemical biosensing, bioelectrodes	High	[5,18]
Proteins	Gold nanoparticles/metal NPs	Protein corona formation, hydrophobic + electrostatic	Dynamic, partially reversible	DLS, proteomics, zeta potential, CD	Biological targeting, adaptive surface identity	Uncontrolled corona evolution; possible denaturation	Drug delivery, photothermal therapy	High–Moderate	[10,29]
DNA/RNA	Gold nanoparticles	Electrostatic + covalent (thiolated DNA–Au)	Moderate–High stability; tunable reversibility	UV–Vis, SPR, fluorescence, gel electrophoresis	High recognition specificity, programmable assembly	Salt/pH sensitivity for electrostatic assembly	DNA sensing, gene delivery	High	[1,7]
DNA	Graphene/carbon nanomaterials	π – π stacking, hydrophobic interactions	Moderate; generally reversible	Raman, AFM, fluorescence spectroscopy	Excellent charge transfer and surface adsorption	Possible conformational perturbation; desorption risk	Biosensing, nanoelectronics	Moderate–High	[26,60]
Peptides	CNTs/graphene	π – π stacking, hydrogen bonding	Moderate stability; reversible	Raman, FTIR, AFM	Biomimetic assembly, conductive interfacing	Orientation control challenges	Tissue engineering, bioelectronics	Moderate	[38,80]
Enzymes/proteins	Metal oxide nanoparticles	Electrostatic + coordination interactions	Moderate–High; environmentally sensitive	XPS, ITC, DLS, activity assays	Catalytic enhancement, nanozyme compatibility	Ion release/toxicity; possible activity loss	Nanozyme catalysis, biosensing	High	[32,33]
Polysaccharides (chitosan, alginate)	Metal/polymer nanoparticles	Hydrogen bonding, electrostatic interactions	Moderate; pH/ionic-strength sensitive	FTIR, zeta potential, SEM	Excellent biocompatibility, film formation	Lower structural robustness; swelling/degradation	Drug delivery, antimicrobial coatings	High	[62,75]
DNA (aptamers)	Quantum dots	Covalent + electrostatic	Moderate–High stability	Fluorescence spectroscopy, TEM, SPR	Strong fluorescence response and target selectivity	Potential QD toxicity; photobleaching concerns	Fluorescence biosensing, imaging	Moderate–High	[25,55]
Proteins/enzymes	MOFs/COFs	Coordination bonding, encapsulation	High stability, confinement-assisted protection	XRD, BET, FTIR, activity assays	Enhanced enzyme protection and recyclability	Diffusion limitation; scalability challenges	Catalysis, drug delivery	Moderate–High	[86,87]
Peptides/biomolecules	Hybrid nanomaterials	Self-assembly, noncovalent interactions	Variable; often tunable and reversible	AFM, TEM, spectroscopy	Multifunctionality and adaptive behavior	Design complexity; reproducibility issues	Smart materials, nanomedicine	Emerging/Moderate	[39,42]

Table 4. Design considerations for biomolecule immobilization at nano–bio interfaces.

Parameter	Impact on Function	Design Strategy	References
Orientation	Active-site exposure	Site-specific conjugation	[5,31,71,80]
Conformational stability	Activity retention	PEGylation/mild immobilization	[1,9,10,57]
Linker length	Electron transfer vs flexibility	Spacer engineering	[18,22,71]
Surface density	Accessibility/steric crowding	Controlled grafting	[4,17,49]
Adsorption mode	Reproducibility and binding behaviour	Controlled immobilization	[1,13,31,59]

4. Nanomaterial Platforms for Bio-Interfacing

The high surface area, tunable physicochemical properties and nanoscale dimensions of nanomaterials make them versatile platforms on which biomolecule nanomaterial interfaces can be constructed. These groups allow effective immobilization of biomolecules and allow them to effectively interact to modify biological activity, catalytic activity and signal transduction. Different types of nanomaterials have been investigated as scaffolds for bio-interface in the last twenty years, including metal and metal oxide nanoparticles, carbon-based nanomaterials, semiconductor nanocrystals, and porous framework materials.

Each nanomaterial platform has its own advantages and disadvantages when applied to bio-interfacing applications. However, metal nanoparticles exhibit excellent plasmonic, catalytic and electronic characteristics, but can be a source of concerns about the release of ions, long-term toxicity and colloidal instability. Although high surface area and mechanical strength, and generally good conductivity are achieved with carbon-based nanomaterials, the heterogeneity in surface chemistry and the difficulty to obtain reproducible functionalization are not yet solved. MOFs and COFs are structurally highly tunable and modular platforms with controlled pore architectures and biomolecule loading and interface engineering, compared to these conventional platforms. However, they are currently still being studied for their hydrolytic stability, scaling up and biological compatibility in physiological conditions. These differences emphasize the fact that there is no universal nanoplatform available and application-specific requirements must be taken into account when selecting a platform [9,11,38,55,82].

The nanomaterial platform has a great impact on the nano-bio interface structure and functionality. The essential parameters that control the adsorption, orientation and interaction of the biomolecules on the nanomaterial surface are the size, surface charge, morphology, and chemical functionalization of the biomolecules. As a result, nanomaterial platforms need to be carefully picked and engineered in order to maximize the stability and functionality of hybrid bio-nanostructures in the context of biosensing, catalysis, diagnostics and therapeutic applications [9,11,12,49,89,90].

4.1. Metal and Metal Oxide Nanomaterials

One of the most common platforms on which biomolecule interfacing has been done is metal and metal oxide nanomaterials, which possess unique optical, electronic, and catalytic characteristics. Noble metal nanoparticles like gold, silver, and platinum are highly stable in their chemistry and exhibit strong surface plasmon resonance, which makes them suitable in biosensing, imaging and delivery of drugs [91]. Gold nanoparticles, in particular, have found extensive use in immobilization of biomolecules owing to their high affinity for thiol functional groups (including thiolated strands of DNA and cysteine residues). The reason behind this is that the Au-S bonds of the kind that are formed are precisely stable to enable biomolecules to be engendered in the precise manner that they are biologically pertinent. These attributes have enabled the development of a wide range of biosensing systems, which are: colorimetric, fluorescence sensors and surface-enhanced raman spectroscopy (SERS) sensors [55–58,67,92].

Another valuable platform to be used in bio-interfacing is metal oxide nanomaterials, which include iron oxide, titanium dioxide and zinc oxide nanoparticles. Iron oxide nanoparticles have been actively applied in magnetic targeting and biomedical imaging because of their magnetic properties, which are superparamagnetic. On the same note, metal oxide nanostructures are also applicable as catalytic surfaces on which enzymes can be immobilized and used in nanozyme uses. Nonetheless, surface chemistry should be controlled with utmost care to achieve biocompatibility and eliminate possible toxicity due to the release of metal ions. Metal and metal oxide nanomaterials are not universally interchangeable, and their suitability depends strongly on the intended application. Gold nanoparticles (AuNPs) have several desirable characteristics for plasmonic biosensing: They exhibit a high surface plasmon resonance, they can be easily functionalized with surface chemistry, and they have a well-developed thiol–gold conjugation chemistry for controlled immobilization of biomolecules. Silver nanoparticles tend to have higher plasmonic enhancement and better SERS performance; however, chemically they are not as robust and are more prone to oxidation. These have great potential in catalytic systems, magnetic targeting and applications of nanozyme due to their catalytic activity, semiconducting behavior and magnetic

properties, respectively, among which metal oxide nanomaterials are particularly attractive, such as Fe_3O_4 , TiO_2 and ZnO . Ion release, surface instability and biocompatibility are, however, important limitation and care must be taken to surface engineer it [12,16,49,88,89].

4.2. Carbon-Based Nanomaterials

Another significant group of platforms of interfaces of biomolecules and nanomaterials is carbon-based nanomaterials that exhibit high electrical conductivity, chemical stability, and surface area. Representative examples include graphene, graphene oxide, carbon nanotubes, carbon dots and nanoporous carbon materials.

Graphene and carbon nanotubes have π -electron systems that are extended and facilitate the strong π - π stacking interactions with aromatic biomolecules, including nucleic acids and some proteins. The interactions also lead to a successful immobilization of biomolecules without the need a covalent modification. Moreover, carbon nanomaterials are conductive to a high degree of electricity, and they enable the conduction of electrons, which are particularly beneficial in electrochemical biosensors and bioelectronic devices [60–63,65,66,93]. Carbon-based nanomaterials are especially useful when a rapid charge transfer and efficient electronic communication is needed. The electrical conductivity, high porosity, and long π -electron networks of graphene and carbon nanotubes (CNTs) ensure excellent electron transfer properties and high π - π interactions with aromatic biomolecules. Thus, these materials are extensively used in electrochemical biosensors, bioelectronics and catalytic interfaces. However, the challenges of surface heterogeneity, aggregation, reproducible functionalization and long-term biological compatibility have yet to be overcome for wider translation. Carbon nanomaterials have often been found to be superior in conductivity-driven applications, but may offer less convenient biomolecule conjugation chemistry, when compared with noble metal platforms [22,55,68,72,85,88].

Carbon nanomaterials have been found to be of high potential in the medical field in terms of biosensing, bioimaging and drug delivery [94]. Their surface area is large and can accommodate several biomolecules, therefore presenting multifunctional platforms that can simultaneously be used to sense, target and treat. Also, biomimetic engineering has been able to assist in developing carbon-based nanozymes that mimic the natural enzymatic activity without affecting the stability and catalytic activity.

4.3. Semiconductor Nanocrystals and Quantum Dots

Semiconductor nanocrystals, also known as quantum dots (QDs), are optical and electronic nanomaterials whose properties are determined by their size. The materials possess unique fluorescence characteristics, such as narrow emission, broad absorption, and high photostability, and thus are highly popular in biological imaging and sensing. Biomolecules such as antibodies, peptides, or nucleic acids can be conjugated to quantum dots, creating highly sensitive detection platforms. Multiple biomolecules can be detected simultaneously in a single system because the optical properties of particles can be tuned by manipulating particle size. Thus, QDs have found a wide range of applications in the areas of fluorescence biosensing, cellular imaging, and intracellular processes monitoring applications [5,6,8,71,72].

Other uses of semiconductor nanocrystals include photocatalysis and optoelectronic devices in contact with biological systems, although optical applications are also known to exist. Nevertheless, other concerns, like possible toxicity of heavy metal elements and surface instability, are also critical issues to consider when using them in biomedicine. Passivation and encapsulation of the surfaces is thus a common strategy used to improve their biocompatibility and stability. The distinctive features of quantum dots, such as adjustable optical properties, narrow emission spectra and high photostability make them unique among platforms of nanomaterials. The unique properties of quantum dots are especially attractive for fluorescence biosensing, multiplex detection and bioimaging. However, they still have some restrictions in their biomedical applications due to potential toxicity, long-term biocompatibility concerns, and surface degradation. In order to achieve better translational viability, surface passivation, less toxic quantum-dot compositions, and quantum-dot encapsulation continue to be areas of active research [2,55,82,88,89].

4.4. Metal-Organic Frameworks (MOFs) and Covalent Organic Frameworks (COFs)

Metal-organic frameworks (MOFs) and covalent organic frameworks (COFs) constitute a very fast-emerging category of porous nanomaterials, with significant potential for biomolecule interface engineering. The materials are periodic networks of metal nodes or organic linkers and thus have highly porous structures with tuned pore size, surface chemistry and structural functionality. MOFs have attracted considerable interest in biotechnology and medicine because they can capture biomolecules in their porous structures. Enzymes, drugs, nucleic acids and

other biomolecules can be easily loaded through the high surface area and tunable pore architecture. This has resulted in MOF-based drug delivery systems, catalytic nanoreactors and biosensing systems.

Similarly, COFs also offer highly ordered organic frameworks that have outstanding chemical stability and functionality versatility. Their distinct pore structures and large groupings of functional groups are what allow them to interact well with biomolecules and can be assembled at the nano bio interface in a controlled manner. MOFs and COFs are also beginning to be used in the enzyme immobilization, catalytic, and biomedical fields, with their porous structures having the potential to preserve biomolecules and keep them accessible to substrate interactions. All these properties render them as having a bright future as the next generation of hybrid nano-biomaterial platforms. MOFs and COFs have distinctive advantages over conventional nanoparticle platforms due to their well-ordered porous nature and internal chemistry. These materials are especially suited to the application of biomolecule encapsulation, enzyme stabilization, controlled cargo loading, and catalytic confinement in which the biomolecular functionality must be preserved. MOFs are also seen to have great potential in enzyme immobilization and protected nano-reactor systems, as they create porous framework that can protect the biomolecules from denaturation and can allow access to the substrate into the framework. But, problems of hydrolytic stability, framework degradation, scale up, and physiological compatibility pose challenges. However, MOFs/COFs offer functionalities that are very different from metal or carbon platforms, but are not as ready to use [38,55,86,88]. Table 5. Overview of major nanomaterial platforms used for biomolecule interfacing, including metal and metal oxide nanoparticles, carbon-based nanomaterials, semiconductor quantum dots, and porous frameworks such as MOFs and COFs. Key physicochemical properties, biomolecule integration strategies, and representative applications are summarized.

Ultimately, the choice of an appropriate nanomaterial platform will depend on the dominant functional requirement in the targeted application. Gold nanoparticles, graphene and carbon nanotubes, quantum dots, MOFs and COFs, and polymeric nanoparticles each possess distinct properties that make them attractive for specific biomedical applications. The comparative evaluation would illustrate the importance of considering a combination of optical, catalytic, electronic, structural, and biological design requirements rather than a single best material system to be used as the platform for the device. No single nanomaterial platform is appropriate for every application of biomolecules with nanomaterials, so the selection of the platform must consider the surface chemistry, charge-transfer capability, optical properties, catalytic behavior, biocompatibility, scalability and the suitability of the platform for translation into applications [9,11,86–89]. A comparative summary of the major strengths, limitations and preferred application fields of the main classes of nanomaterials is presented in Table 6.

Table 5. Comparative nanomaterial platforms for biomolecule–nanomaterial interfaces.

Nanomaterial Type	Key Properties	Typical Biomolecule Integration	Interaction Stability/Reversibility	Typical Characterization Method	Major Advantages	Main Limitations/Risk	Best-Suited Applications	Application Readiness	Ref.
Metal nanoparticles (Au, Ag, Pt)	Surface plasmon resonance, catalytic activity, high conductivity	Proteins, DNA, peptides (thiol, electrostatic, covalent coupling)	High stability; Au–thiol often irreversible	UV–Vis, TEM, SPR, XPS	Strong plasmonics, facile biofunctionalization, excellent sensing compatibility	Aggregation, cost, possible cytotoxicity (Ag/Pt)	Biosensing, imaging, drug delivery	High	[10,55]
Metal oxide nanoparticles (Fe ₃ O ₄ , TiO ₂ , ZnO)	Magnetic, catalytic, semiconducting behavior	Enzymes, proteins, polysaccharides	Moderate–High; environmentally sensitive	XRD, TEM, zeta potential, activity assays	Magnetic recovery, catalytic versatility, nanozyme compatibility	Ion release, oxidative stress, toxicity concerns	Nanozymes, imaging, biosensors	High–Moderate	[12,33]
Carbon-based nanomaterials (graphene, CNTs, carbon dots)	High conductivity, large surface area, π -electron system	DNA, peptides, proteins (π – π stacking, hydrophobic adsorption)	Moderate; often reversible	Raman, AFM, electrochemistry, fluorescence	Exceptional charge transfer, high loading capacity	Functionalization complexity, dispersion challenges	Bioelectronics, sensing, drug delivery	High–Moderate	[60,88]
Semiconductor quantum dots	Size-tunable fluorescence, photostability	DNA, antibodies, proteins	Moderate–High stability	Fluorescence spectroscopy, TEM, DLS	Bright fluorescence, multiplexed detection capability	Heavy-metal toxicity, photobleaching risk	Bioimaging, fluorescence biosensing	Moderate–High	[25,94]
Metal–organic frameworks (MOFs)	High porosity, tunable structure, large surface area	Enzymes, proteins, drugs (encapsulation, coordination)	High stability; confinement-assisted protection	XRD, BET, FTIR, activity assays	Enzyme protection, cargo loading, catalytic enhancement	Diffusion limitations, moisture/scalability issues	Drug delivery, catalysis, biosensing	Moderate–High	[86,87]
Covalent organic frameworks (COFs)	Ordered porous framework, chemical stability	Biomolecules through adsorption or functional groups	High; tunable reversibility	XRD, FTIR, SEM, adsorption studies	Structural precision, chemical robustness	Limited aqueous processability, synthetic complexity	Catalysis, sensing, energy applications	Moderate/Emerging	[81,86]
Polymeric nanoparticles	Biocompatibility, flexibility, controlled release	Proteins, drugs, nucleic acids	Moderate–High; formulation-dependent	DLS, SEM/TEM, release assays	Excellent biocompatibility, tunable release kinetics	Lower conductivity, batch variability	Drug delivery, therapeutics	High	[2]
Hybrid nanomaterials	Combined multifunctional properties	Proteins, peptides, DNA	Variable; design-dependent	Multimodal characterization (TEM, spectroscopy, electrochemistry)	Multifunctionality, synergistic performance	Design complexity, reproducibility/scaling challenges	Integrated sensing, nanomedicine, theranostics	Moderate/Emerging	[9,61]

Table 6. Comparative suitability of nanomaterial platforms for biomolecule–nanomaterial applications.

Platform	Major Strength	Main Limitation	Best-Suited Applications	References
Gold nanoparticles	Plasmonics, thiol chemistry	Cost, aggregation	Biosensing, imaging	[5,29,30]
Metal oxides	Catalysis, magnetism	Ion release, toxicity	Nanozymes, imaging	[21,33,56]
Graphene/CNTs	Conductivity, charge transfer	Functionalization complexity	Electrochemical sensing	[22,55,88]
Quantum dots	Fluorescence	Toxicity concerns	Bioimaging, fluorescence assays	[1,15,82,94]
MOFs/COFs	Encapsulation, protection	Stability/scalability	Enzyme immobilization, drug delivery	[86,87,95]
Polymeric nanoparticles	Biocompatibility, controlled release	Lower conductivity	Drug delivery	[2,3,66]
Hybrid nanomaterials	Multifunctionality	Design complexity	Integrated sensing/therapy	[6,38,55]

5. Chemical Design Strategies for Interface Formation

The successful development of stable and functional biomolecule–nanomaterial interfaces requires the application of rational chemical design strategies that controls the biomolecular interaction of the nanomaterial surfaces. With these close interdependencies of the nano–bio interactions with the surface chemistry, molecular orientation and the environmental conditions, the rational design of such interfaces is fundamental to the achievement of predictable functionality in the biosensing, catalysis, nanomedicine and environmental application.

The contemporary interface engineering methods are based on various complementary approaches such as surface functionalization, biomolecule-directed synthesis, self-assembly processes, and stimuli-responsive interface design. These methods permit close control of molecular interactions at the nanoscale and permit the construction of hybrid systems with structures and behaviour of interest. Through a hybrid set of chemical modification and biomolecular recognition processes, scientists can fabricate interfaces that represent a biological system with the advantage of exploiting the physicochemical characteristics of nanomaterials [9,11,12,49,90].

5.1. Surface Functionalization and Ligand Chemistry

One of the most commonly used methods of biomolecule–nanomaterial interface engineering is surface functionalization. This approach involves modifying nanomaterial surfaces with organic ligands, polymers, or biomolecules to introduce specific functional groups to facilitate specific interactions with biological molecules. Ligand chemistry is instrumental in regulating the stability and solubility, as well as the connection with biomolecules in nanoparticles. For example, gold nanoparticles have been functionalized with ligands that have thiol groups because of the good Au–S bond that allows proteins, peptides, and nucleic acids to be fixed onto the surface in a stable manner. Equally, carboxyl, amine, and hydroxyl residues created using surface ligands can undergo covalent coupling reactions with biomolecules and thus incorporation of stable nano–bioconjugates.

Surface functionalization allows a finer controllability of surface charge, hydrophobicity and steric properties too, which have a strong effect on biomolecular adsorption and orientation. Nanomaterials can also be utilized to enhance the workings of a nanomaterial-based system and therapeutic apparatus by appropriately modifying the surface to decrease the non-specific adsorption and promote biocompatibility [9,55–59,95]. Furthermore, nanoparticles may be coated with polymers such as polyethylene glycol (PEG) to enhance their stability and reduce undesired biological interactions. These methods have been particularly applied in biomedical applications where the circulation time of nanoparticles, immune responses and cellular uptake are important.

5.2. Bio-Templated and Bio-Directed Synthesis

Bio-templated synthesis is a powerful approach in which biomolecules and biomolecular assemblies serve as directing agents for nanomaterial formation. In this method, the nucleation, growth and organization of nanomaterials are directed by biomolecules, which can be proteins, DNA, peptides or polysaccharides, and thereby enabling the formation of highly ordered structures under mild conditions [96]. As an example, nucleic acids may serve as programmable scaffolds by which nanoparticles can be assembled to form desired spatial structures by controlling hybridizations with sequences. Likewise, proteins and peptides are able to selectively bind to the specific crystal faces of nanomaterials, affecting the morphology and size of nanoparticles in synthesis.

This approach is inspired by biological biomineralization processes that are found in natural systems, in which biomolecules are used by organisms to regulate the development of mineral structures with very high levels of accuracy. Bio-directed synthesis has thus allowed eco-friendly and best controlled pathways to synthesise nanomaterials with complicated structures. These biomolecule-based methods have been extensively utilized in the preparation of metal

nanoparticles, semiconductor nanocrystals and hybrid nanocomposites, and offer useful devices in the development of functional nano-bio interfaces with desired chemical and structural characteristics [5,68–71,73,74].

5.3. Self-Assembly and Hierarchical Organization

Self-assembly is a process in which molecules are directed to form periodic structures by means of thermodynamically favourable interactions. Self-assembly in biomolecule nanomaterials can take place via noncovalent forces including electrostatic, hydrogen bonding, hydrophobic interaction, and π - π stacking. It is these interactions that allow the nanoscale building blocks to self-assemble into complex structures. Self-assembly is particularly important for the organization of biomolecule-functionalized nanoparticles, nanowires and hierarchical nanostructures.

Molecular self-assembly is also common at biomolecular interfaces (like the adsorption of proteins onto nanoparticles surfaces) and in lipid and lipid bilayer formation and in organizing biomolecular condensates. Using these natural processes, scientists are able to develop nanostructures that can replicate biological processes but which can be produced with controllable characteristics. By incorporating several degrees of internal sophistication, hierarchical organization also extends the self-assembly process. In this type of system, nanoscale parts can be combined to create larger structures with emergent properties absent in the individual building blocks. The hierarchical assemblies may be particularly handy as the biosensing, nanomedicine, and catalytic nanoreactor applications [38,39,41,97]. Bio-templated synthesis and self-assembly processes play a crucial role in the formation of ordered nano-bio structures, as illustrated in Figure 3.

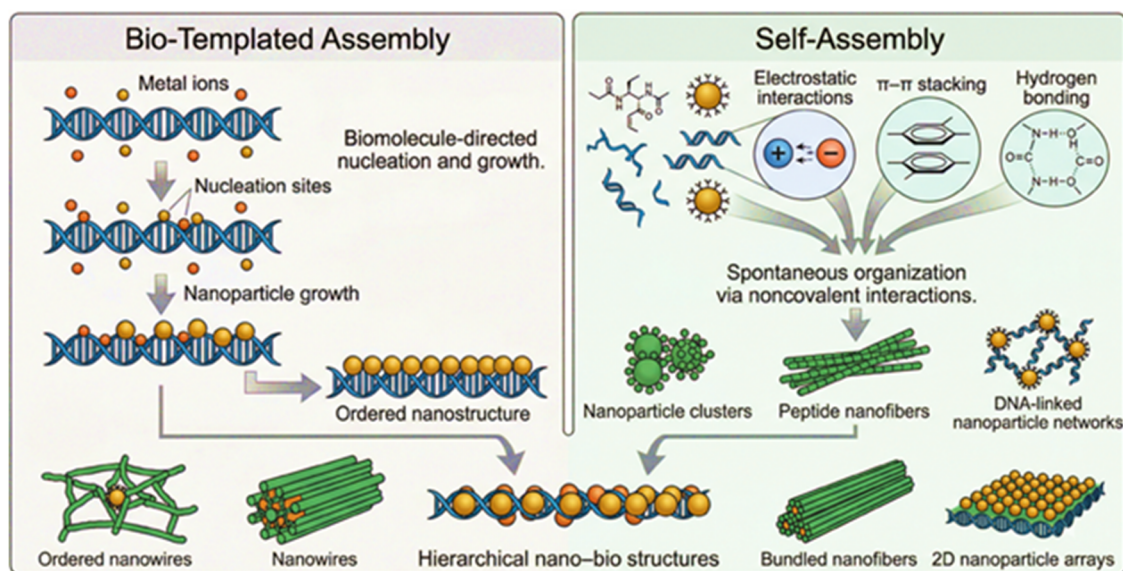


Figure 3. Schematic illustration of bio-templated and self-assembly mechanisms at biomolecule–nanomaterial interfaces, showing biomolecule-directed nucleation and growth alongside spontaneous organization of building blocks into ordered nanostructures.

5.4. Stimuli-Responsive and Dynamic Interfaces

An emerging field of nano-bi interface design involves the fabrication of stimuli-responsive and programmable interfaces that respond to changes in the environmental conditions. These systems are set to react to the outside stimuli like pH, temperature, light, magnetic fields or chemical signals. Stimuli-responsive nanomaterials are capable of changing their structure, surface characteristics, or the affinity of biomolecules for their surroundings. For example, a thermoresponsive polymer attached to nanoparticle surfaces can regulate biomolecule interactions by having a reversible conformational change. Similarly, photo responsive molecules can be used to allow light controlled stimulation of catalytic or sensing activities.

Dynamic nano bio interfaces are also naturally present in biological systems, where biomolecules are constantly interacting with nanomaterials forming dynamic biomolecular structures known as protein coronas. These dynamic interactions may also have a profound effect on the behaviour of nanoparticles within biological systems, including cellular uptake, biodistribution and immune recognition. The responsive interface design therefore has the potential to produce adaptive nanomaterials capable of performing complex functions, i.e., controlled drug delivery, targeted

diagnosis and responsive catalytic reaction. Such systems are expected to advance the development of smart nanomaterials capable of dynamically interacting with biological environments [32,35,37,53].

6. Structural and Spectroscopic Characterization of Interfaces

A comprehensive understanding of biomolecule–nanomaterial interfaces require the presence of advanced analytical tools, which may be employed to investigate structural organization, chemical structure and molecular–interaction processes on a nanoscale. Since such interfaces are frequently complicated and dynamic systems of interactions between inorganic substances and natural biomacromolecules, a combination of microscopic, spectroscopic, and in vivo analytical techniques is often necessary to clarify their characteristics.

One of the most important parameters that can be studied by the researchers using characterization methods is the size of nanoparticles, morphology, surface chemistry, the orientation of biomolecules, and their conformational changes during adsorption. These methods also provide insights into the development of biomolecular coronas, binding, and stability of nano–bio assemblies. Proper characterization is thus critical in learning effects of nano–bio interfaces on biological reaction, catalysis and device functionality [1,4,31,98,99]. Recent progress in nanoscale characterization has greatly enhanced the capability of real time and biologically relevant observation and analysis of nano bio interactions. A combination of complementary methods will result in a better comprehension of the physical and chemical mechanisms that regulate the formation and functioning of these interfaces.

6.1. Microscopic and Surface Analytical Techniques

Microscopy is important in revealing the structure of biomolecule–nanomaterial interfaces. Transmission electron microscopy (TEM) and scanning electron microscopy (SEM) are high-resolution imaging methods that are commonly used to observe the structure of nanoparticles, size distribution, and surface architecture. TEM, specifically, offers atomic level resolution which allows direct examination of the structure of nanoparticles and biomolecular assemblies in the nanoscale. The other highly effective method used to examine the nano– bio interfaces is the atomic force microscopy (AFM) which enables the examination of the surfaces in the near physiological conditions. Other mechanical properties that can be determined by AFM include roughness of surfaces, adhesion forces, and molecular interactions between biomolecules and the surfaces of nanomaterials.

Other than imaging methods, a number of surface analytical methods provide information about the chemical composition and elemental distribution at the interface. It is often necessary to use other physicochemical characterisation methods to gain a fuller picture of the nano–bio interfaces. Dynamic light scattering (DLS) and zeta potential analysis are commonly used to assess the hydrodynamic size and the aggregation behaviour as well as the changes in the surface charge after biomolecule adsorption. Changes in particle diameter or zeta potential although indicative of successful surface modification or corona formation, do not provide any information to distinguish between covalent conjugation, non-specific adsorption or multilayer assembly [4,10,14,59]. Similarly, SEM and TEM can offer insight into nanoscale organization and aggregation state and into particle morphology, but it is not usually possible to make direct conclusions about conformational integrity and biomolecule orientation or binding mechanisms. Under more physiologically relevant conditions, advanced structural approaches such as cryo-TEM and small-angle X-ray/neutron scattering (SAXS/SANS) provide further insight into nano–bio-organization. Cryo-TEM allows the imaging of hydrated systems without drying artifacts that are typical for conventional electron microscopy. SAXS and SANS are especially well suited to investigate the structural organization, shell thickness and the architecture of the interfaces in solution on the nanoscale. These methods, however, generally yield only average structural data and are often combined with spectroscopic or biochemical data for a fuller characterization of molecular organization [1,14,31,47,55]. As an example, XPS can be used to determine the chemical state of surface atoms and identify functional groups involved in biomolecular binding. Equally, elemental mapping of hybrid nanostructures may be done by energy-dispersive X-ray spectroscopy (EDS) and scanning transmission electron microscopy (STEM). Together, these methods provide a comprehensive understanding of nanomaterial surfaces and biomolecular adsorption mechanisms underlying the formation of nano–bio interfaces.

6.2. Spectroscopic Probes of Binding and Conformation

The spectroscopic methods are commonly used to examine chemical events and structural dynamics at the interfaces between biomolecules and nanomaterials. The techniques can give data on molecular bonding, conformational alteration of biomolecules and electronics of nanomaterials with biological components. The determination of functional groups and the modification in the secondary structure of proteins after adsorption on

the surface of nanomaterials are usually performed by Fourier-transform infrared spectroscopy (FTIR). Changes in characteristic vibrational bands can indicate interactions between biomolecules and nanomaterials such as hydrogen bonding or coordination bonding.

The SERS and Raman spectroscopy are potent on investigations of molecular interactions at the nano-bio interfaces. Raman signals can be strongly enhanced by metal nanoparticles with high plasmonic scattering and thus biomolecules can be detected at a very sensitive level and can give specific details on the orientation and chemical bonding of the molecules. The spectroscopic characterization of nano-bio interfaces should be interpreted carefully because none of the existing techniques are able to provide unambiguous information about all aspects of interface structure and function. FTIR spectroscopy may be used to identify characteristic vibrational signatures, and to infer changes in chemical environments and/or changes in secondary structure, but it generally cannot directly confirm site-specific attachment and/or controlled biomolecule orientation. In the same way, Raman spectroscopy and SERS can give extremely sensitive molecular fingerprints and information about nearby bonding environment, but quantification can be challenging because of signal enhancement and spectral complexity. Thus, binding mechanisms and orientation are in general only conclusively demonstrated with complementary analysis techniques [1,14,31,47,55]. Another significant methodology for investigating nano-bio interactions is the fluorescence spectroscopy. The effects of fluorescence quenching or enhancement may be used to give information about the binding events, and the energy transfer of biomolecules with nanomaterials. Semiconductor nanocrystals and quantum dots are also of great benefit in the fluorescence-based detection systems as they have strong and tunable optical characteristics [5–8,71,72]. Circular dichroism (CD) spectroscopy can also be used extensively to study observed alterations in the secondary structure of biomolecules, especially in proteins and peptides that interact with nanomaterial surfaces. These spectroscopic techniques are useful in establishing whether biomolecules retain their functional conformations following adsorption.

6.3. In Situ and Operando Characterization Approaches

The sample preparation procedures of traditional characterization methods usually change the natural structure of nano-bio interfaces. In order to surmount those constraints, in situ and operando characterization techniques have been established to trace nano-bio interactions in natural environmental circumstances. Cooperative biomolecule binding behaviour at nano-bio interfaces can also be modelled as Hill-type models to gain insight into binding cooperativity and multivalent interactions [1,5,31,40,55].

$$\theta = \frac{[L]^n}{K_d + [L]^n}$$

where: θ = fraction of occupied binding sites, $[L]$ = ligand concentration, K_d = dissociation constant describing binding affinity, n = Hill coefficient indicating degree of cooperativity

Among several analytical methods, certain ones are of special significance to quantifying binding behaviour and molecular dynamics at interfaces. Surface plasmon resonance (SPR) and quartz crystal microbalance with dissipation monitoring (QCM-D) allow the real-time monitoring of adsorption kinetics, mass accumulation and viscoelastic changes during interface formation. Isothermal Titration Calorimetry (ITC) delivers thermodynamic parameters such as binding affinity, enthalpic parameters, and stoichiometry, thereby providing insight into the mechanisms of biomolecule–nanomaterial interactions. Nuclear Magnetic Resonance (NMR) may be able to provide information about the molecular structure as well as local chemical environments and conformational changes, but can be experimentally challenging for larger-scale nanoparticle systems. These methods are used in addition to microscopy and spectroscopy to give quantitative information on interaction energetics and the evolution of the interfaces during the interaction [1,14,31,47,55]. In situ methods enable real-time monitoring of the formation of interfaces and structural variations during a chemical reaction, biomolecular adsorption, or the assembly of nanomaterials. To illustrate, dynamic processes, e.g., nanoparticle growth, formation of biomolecular corona, and structural changes of the interface can be observed through in situ spectroscopy and electron microscopy.

Operando characterization methods go a step further and measure structural changes and functional performance of the system as it operates. Such methods are especially useful in catalytic systems, biosensors, and nanozyme systems where interface dynamics has a direct impact on the performance of the device. The characterization of dynamic nano-bio interfaces are increasingly dependent on techniques that are able to follow biomolecule exchange, corona evolution and nanoparticles behaviour in complex environments. Proteomics-based analytical workflows, such as identification of proteins by mass spectrometry (MS), have proven to be powerful tools for understanding the composition of coronas, competitive adsorption processes and temporal evolution of biomolecular layers. Single-particle tracking methods also provide the possibility of following the motion of nanoparticles, cell uptake and heterogeneity in nanoparticle interactions at the single particle level, which can be

hidden in ensemble averaged measurements. These methods are very useful to differentiate between transient adsorption events and stable interface architectures [10,14,52,57,59]. Advances in analytical instrumentation have enabled methods that can probe nano- bio interfaces at high spatial and temporal scales. Indicatively, real-time spectroscopic imaging of nanozyme systems has demonstrated that interactions of bio-molecules influence catalysing activity and stability of nanozyme systems in complicated biological systems [32–36,100]. A combination of in situ and operando characterization methods offers important information on the dynamic character of nano bio interfaces, provides a better understanding of how molecular interactions change with time, and in functional conditions.

One important point in considering nano–bio characterization is the type and the certainty of evidence provided by a technique. The main techniques used to identify the structural organization are morphological methods; chemical environment is revealed by the use of spectroscopic methods; interaction energetics is quantified by calorimetric and kinetic methods; and biological complexity and temporal dynamics are emphasized by proteomic and single particle methods. Therefore, it is generally necessary to perform multimodal analysis of biomolecule–nanomaterial interfaces in order to avoid overinterpretation when using a single technique for adsorption, orientation, conjugation, or functional response. The diversity in the information that can be obtained from different characterization techniques dictates that generally, the interpretation of biomolecule–nanomaterial interfaces is best achieved through multimodal analysis rather than by relying on any single characterization technique [1,14,31,47,55]. Various approaches explore complementary facets like morphology, surface chemistry, adsorption kinetics and thermodynamics, conformational behavior, composition of the corona and dynamics of the evolution of the interface. The principal characterization tools are reviewed here briefly, along with their strengths and interpretational shortcomings, and presented in a tabular format in Table 7.

Table 7. Comparative characterization tools for biomolecule–nanomaterial interfaces.

Technique	What it Measures	Strength	Limitation/Cannot Prove	References
TEM/SEM	Morphology, particle size	High-resolution imaging	Cannot confirm binding chemistry	[4,14,78,91]
AFM	Topography, interaction forces	Surface mapping	Limited chemical specificity	[43,73,96,97]
DLS	Hydrodynamic size	Fast solution analysis	Cannot distinguish adsorption type	[4,14,57,59]
Zeta potential	Surface charge	Detects surface modification	Cannot confirm covalent conjugation	[4,9,17,57]
FTIR	Functional groups	Chemical signatures	Cannot prove molecular orientation	[13,32,56,80]
Raman/SERS	Molecular fingerprints	High sensitivity	Complex quantitative interpretation	[44,98]
SPR	Binding kinetics	Real-time monitoring	Limited structural detail	[1,31,57]
ITC	Thermodynamics	Binding energetics	Requires careful model fitting	[1,5,31,40]
NMR	Structure/conformation	Molecular-level insight	Challenging for complex systems	[1,5,31,38]
Cryo-TEM	Hydrated morphology	Native-state imaging	Limited direct chemistry	[47,78,86,95]
SAXS/SANS	Solution structure	Ensemble architecture	Indirect structural inference	[5,47,52,57]
Proteomics	Protein corona composition	Biological specificity	Requires advanced analysis	[10,14,52,59]
Single-particle tracking	Particle dynamics	Resolves heterogeneity	Technically demanding	[17,52]

7. Structure-Function Relationships at Bio-Nano Interfaces

The structural order and molecular interactions at the nano- bio interface inherently determine the functionality of biomolecule- nanomaterial systems. The geometry, chemical bonding and electronic interactions between biomolecules and nanomaterials that occur at this interface dictate the physicochemical behaviour of the hybrid system. Knowledge of these structure-function relationships is needed to rationally design nanomaterials that can be utilized to effectively interact with biological systems and carry out defined tasks including sensing, catalysis or energy conversion.

An overarching principle that has emerged from biomolecule–nanomaterial engineering is that the chemistry of the interfaces is directly related to the functional output. The catalytic efficiency, the sensitivity towards the target, the drug release rate, and the biological compatibility of the molecule depends significantly on the binding mode, the surface coverage, the orientation and the electronic coupling of the molecule. Thus, the same nanomaterial platform can show quite different properties depending on the prevailing interaction in the interface. Hence, the understanding of these chemistry–function relationships is crucial to rational selection and optimization of bio–nano interface strategies [1,5,9,55,82].

Nanomaterials are materials with size-dependent behaviour that is typically quite different than that of bulk materials, such as enhanced optical absorption, altered electronic structure, or modified catalytic activity. When

nanomaterial surfaces are bound to biomolecules, these properties can be altered through biomolecule control of surface states, charge distribution and interfacial energy landscapes. Therefore, the behaviour of bio-nano hybrid systems is very sensitive to the structural properties of the interface, such as direction of biomolecules on the surface, coverage, and chemical interactions between the biomolecule and the surface [23,25,29–31].

More often than not, biomolecule adsorption can induce conformational changes that will influence biological activity, and nanomaterial surfaces can promote electron- or energy-transfer processes that will increase system performance. Thus, one of the primary goals in designing advanced nano-bio systems are to elucidate the correlation between interface structure and functional performance. The relationship between interface chemistry, structural organization, and functional performance in biomolecule-nanomaterial systems is schematically illustrated in Figure 4.

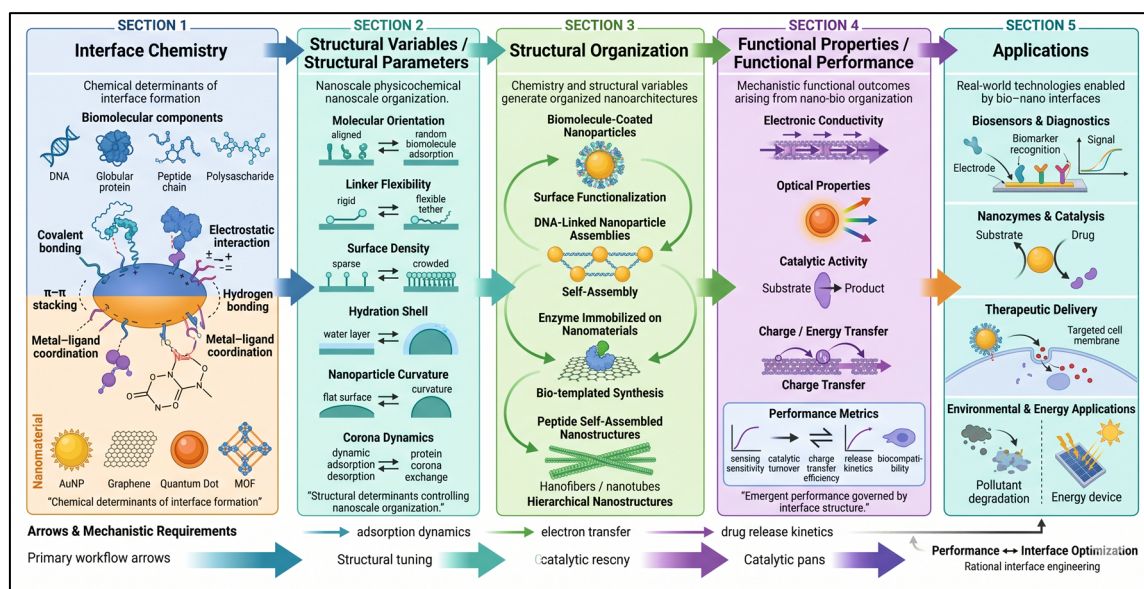


Figure 4. Extended structure–function framework to show how the interfacial chemistry dictates structural organization by means of molecular orientation, linker structure, surface charge, hydration effects, and corona evolution, all of which dictate the efficiency of the sensing, catalytic, charge transport, therapeutic delivery, and biological compatibility.

7.1. Interface-Driven Electronic and Optical Properties

Interaction of biomolecules and nanomaterials has some of the most significant implications because it results in the alteration of both electronic and optical properties at the interface. Biomolecules have the ability to change the electronic environment of the nanomaterial upon contact with the surface, affecting charge distribution, surface plasmon resonance, and optical absorption characteristics [101–103]. Nanoparticles in metals, especially gold and silver have a significant surface plasmon resonance (SPR) that is very sensitive to the local dielectric environment. Compassion of the biomolecules to the nanoparticle surface can cause quantifiable changes in the plasmonic resonance wavelengths, which is the foundation of most biosensing systems. These plasmonic responses allow biomolecules to be detected in a label-free and real-time molecular interactions to be monitored. Subtle changes in the interfaces and their design can significantly impact the sensing performance as demonstrated in practical examples. Sensitization of gold nanoparticles with controlled thiol-mediated immobilization of antibodies often increases the biomolecular orientation and increases accessibility of the targets when compared with random adsorption strategies and yields stronger plasmonic responses and lower detection limits. In a similar fashion, DNA-functionalized quantum dots have shown to become more fluorescently sensitive as a result of the optimum spacer architecture, which ensures that the quantum dots are not undergoing fluorescence quenching yet remain accessible for hybridization. Carbon-based bioelectronic systems can be improved by the incorporation of suitable biomolecular linkers into the graphene structure in order to reduce the steric constraints and facilitate the formation of optimal electronic coupling at the interface [18–22,60,67,85].

Likewise, semiconductor nanocrystals e.g., quantum dots exhibit unusual optical properties, which are a function of particle size and surface chemistry. Biomolecule functionalization can be used to affect fluorescence emission either via energy transfer, fluorescence quenching or via alteration of the surface state. Biosensors and bioimaging systems have greatly utilized these effects in fluorescence-based systems [5–8,71,72]. Interfaces

Selective carbon-based nanomaterials display electronic behaviour at the interface, as well. The approach of aromatic biomolecules to the π -electron systems of graphene or carbon nanotubes can have a substantial impact on electrical conductivity and charge transport and pave the way to highly sensitive electrochemical biosensors and bioelectronic devices [60,61,65,66,93]. Biomolecule–nanomaterial interfaces are well controlled by a variety of interfacial descriptors that include biomolecule orientation, linker architecture, surface charge, nanoscale curvature, organization of the hydration layers, and dynamics of the exchange of the corona. They play a role in different nano-bio applications, such as accessibility to the active site, efficiency of electron transfer, Colloidal stability, Adsorption behavior, and Biological identity of the nano-bio system [1,18–22,52,55,57].

7.2. Catalytic and Enzyme-Like Activities

The nano bio interface is also able to play important catalytic role especially the hybrid system that uses the enzymes or enzyme mimicking nanomaterials. The interface of immobilized enzymes on the surface of nanomaterials can be used to catalyse substances because of better accessibility of substrates, stabilization of enzyme structures and pathways of electron transfer. Another new class of materials that resembles natural enzymatic action is represented by nanomaterials exhibiting inherent catalytic properties, also known as nanozymes. These systems contribute the stability of the inorganic nanomaterials together with the activity of the biocatalysts. Most nanozymes have enzyme-like functions, including peroxidase, oxidase or catalase functionality, and they can be used in biosensing, environmental cleanup, and biomedical diagnostics [32–37,104]. The Michaelis–Menten kinetics approach is often used to assess the catalytic activity of enzyme–nanomaterial and nanozyme systems:

$$v = \frac{V_{max}[S]}{K_m + [S]} \quad (1)$$

The equation is $v = V_{max} [S]/(K_m + [S])$. The equation is: $v = V_{max} [S]/(K_m + [S])$. These parameters allow quantitative estimation of the catalytic efficiency, affinity to the substrate and changes in the functional state after immobilization or nanozyme formation [6,32,58,67,83].

Another challenge in Nanozyme technology is translating its activity from laboratory tests to real-world operating conditions. In many cases, catalytic activity is assessed by simpler chromogenic or fluorogenic model substrates using carefully controlled experimental conditions, which may not necessarily reflect the activity in a complex biological fluid, environmental matrix or multi-component catalytic environment. Furthermore, variations in nanoparticle synthesis and surface modification, as well as dispersion stability, can introduce batch-to-batch variability and reproducibility issues, hindering comparability across studies and wider practical application. Although the nanozymes are very stable and can be synthetically tuned, they have several significant drawbacks compared to natural enzymes [32,65,83]. One of the common issues is the ability to express reduced substrate specificity, where the catalytic activity of many nanozyme systems is broad against model substrates, but it is less selective in recognizing different substrates compared to that of biological enzymes. Moreover, the exact nature of catalytic active sites can sometimes be poorly defined, especially in heterogeneous nanoparticle systems, in which the catalytic activity can vary with particle size, crystallographic facets, oxidation state, surface ligands or defect structure. This lack of mechanism makes the optimization and comparison between studies a bit complicated. The catalytic performance has been shown to be strongly dependent on the interface design, and not just on the nanomaterial composition, in numerous studies. The site-controlled immobilization of enzymes on nanostructured supports, for instance, often results in greater catalytic retention and operational stability than nonspecific adsorption, mainly because the optimum orientation of the enzyme allows to maintain the accessibility of the active site [6,48,55,67,71]. The immobilized enzyme systems attached to gold nanoparticle, carbon nanomaterials, or MOF frameworks have often shown long enzyme half-life, enhanced substrate diffusion, and high resistance to thermal and/or chemical degradation. The catalytic turnovers of nanozyme systems can be strongly affected by surface functionalization, which affects the adsorption of substrates, the local electron density, and exposure of the active site. This highlights that measurable changes in the catalytic activity are obtained when architecture of the interface is controlled. Another example of porous framework materials involves structure–function coupling. Enzymes are typically better protected from pH, protein and heat degradation in MOF based enzyme capsules and retain substrate accessibility as the pores in the MOF are controlled. In comparison to enzyme systems that are free in solution, enzyme systems that are encapsulated have often shown increased operational durability and increased catalytic recyclability. The examples highlight various parameters like confinement environment, linker chemistry, and accessibility at the interface that directly dictate their functional catalytic behaviour [6,9,48,55,67,86].

The interfacial interaction between the nanozyme systems and the biomolecules and the biological environment greatly contribute to the catalytic behaviour of the nanozyme systems. Surface functionalization, particle size, and biomolecular corona formation are factors that can be used to control catalytic activity by changing active sites and reaction pathways. Also, biomolecule-nanomaterial hybrids may be used as artificial catalytic system that will be able to undergo a complicated biochemical reaction. Such systems frequently couple immobilization of enzymes to catalytic sites on nanomaterials to form multifunctional platforms giving biologic specificity with greater chemical stability. The catalytic mechanism of nanozymes at biomolecule-nanomaterial interfaces, including substrate adsorption, electron transfer, and product formation, is illustrated in Figure 5.

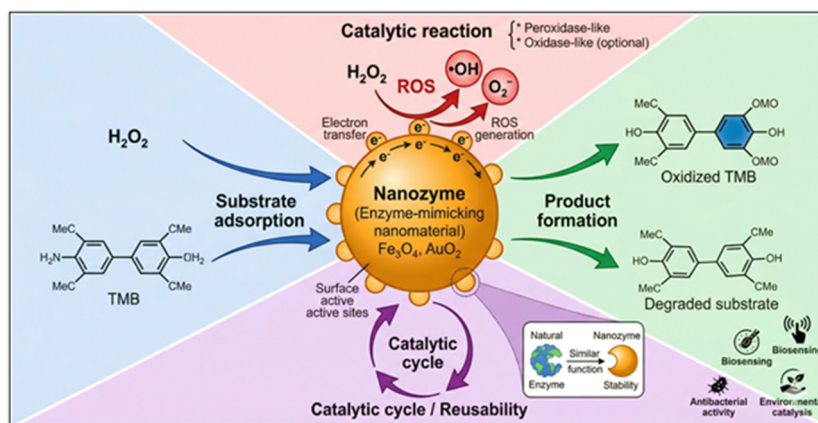


Figure 5. Schematic representation of the nanozyme catalytic mechanism showing substrate adsorption, catalytic transformation via electron transfer, and product formation at active sites of nanomaterials.

7.3. Charge Transfer and Energy Conversion Processes

The charge transfer processes of biomolecule-nanomaterial interfaces are essential in most bioelectronic and energy related industrial applications. Molecular orientation, electronic coupling, and distance between the donor and acceptor species are some of the factors that determine electron transfer between biomolecules and nanomaterials at the molecular level. Nanomaterials may serve as conductive channels that allow straightforward transfer of electrons among a biomolecule and an external electrode. The separation distance between donor and acceptor is often a critical factor in determining electron transfer rates in bioelectronic and catalytic systems:

$$k_{ET} \propto e^{-\beta d}$$

It is the product of electron-transfer rate (k_{ET}), transfer distance (d) and attenuation coefficient (β). This relationship is shown to be a consequence of the important role of linker length, biomolecule orientation, and interface organization in charge-transfer performance [6,22,31,80,81].

The charge transfer is especially dependent on the orientation and the linker architecture at the nano-bio interface. The short conductive linking molecules and enzyme orientation control can significantly enhance electron transfer efficiency in electrochemical biosensors by minimizing donor–acceptor distance between the redox centres and conductive nanomaterial surfaces. Biointerfaces made of graphene- and carbon nanotube materials often rely on π – π and conductive pathways over the surface for fast charge transport and enhanced electrochemical response. On the other hand, if the biomolecule is attached but the electron is not transferred due to excessive polymer coating or improper biomolecule adsorption, the electron transfer will not be true. However, if the biomolecule is attached with the electron transfer not happening due to excess polymer coating or improper adsorption of biomolecule, the electron transfer will not be true. The examples above show that it is not just attachment but also the organization of the interfaces that decides on the bioelectronic functionality [1,9,67,80,81]. Metallic nanoparticles and carbon nanostructures are also exemplary in terms of their application as an electrochemical or electrochemical biosensor and biofuel cell through the electrochemical connection of redox enzymes with electrodes [5–7,71,72].

Though the molecular orientation, distance from biomolecule to nanomaterials and the strength of electronic coupling still play a critical role on the charge transfer efficiency at biomolecule-nanomaterial interfaces. While electron transport pathways can be improved with the use of metallic nanoparticles and carbon nanostructures, the efficiencies have been reported as widely varying as between different studies, likely because of variations in surface chemistry, immobilization strategies and environmental conditions. The discrepancies suggest that the prediction of interfacial electron transfer is still incomplete. More of this computational simulation relying on

spectroscopy might then be crucial to understanding charge transport mechanisms and to rationally design bioelectronic properties [1,5,14,82].

Photoelectrochemical devices and catalytic nanoreactors are also energy conversion systems in which charge transfer occurs. Biomolecules could also be used as light-gathering units, catalysts, or electron-transfer components in such systems. The structural configuration and electronic interface at the nano-bio interface is very critical in determining the efficiency of these processes. However, nanoscale interfaces may also support energy transfer processes, such as Förster resonance energy transfer (FRET) and the plasmon-enhanced energy transfer. The utilization of these mechanisms has been extensively utilized in fluorescence-based biosensing and imaging technologies. In general, understanding charge transfer and energy conversion at biomolecule–nanomaterial interfaces can provide valuable insights for designing advanced nanomaterials to be applied in biosensing, catalysis, bioelectronics, and renewable energy technologies. The structure-function relationships are also vital in the therapeutic nano-bio systems. The drug-loading behaviour, circulation stability and cytocompatibility can be greatly influenced by the surface modification strategies including PEGylation, ligand targeting and biomolecule encapsulation. For instance, the use of PEGylated nanocarriers generally has a lower nonspecific protein adsorption and improved colloidal stability, but a high density of surface shielding can also decrease cellular uptake or targeting efficiency. Similarly, porous nanostructures with designed surface functionality can not only improve drug-loading efficiency but also enable controlled drug release. The examples illustrate the direct impact of intentional interface design on therapeutic efficacy and biological results. It is often true of biomolecule–nanomaterial systems that the functional enhancements are not the result of material composition, but of careful control of the architecture of the interfaces. These factors range from the biomolecule orientation, the length of the linker, the surface density, electronic coupling, the confinement effects to the corona dynamics and can affect measurable performance indicators including the sensing sensitivity, catalytic efficiency, biomolecular stability, charge-transfer kinetics and therapeutic efficacy. Depending upon the material composition and their interface architectures, the performance of biomolecule–nanomaterial systems could be measured. Various functions like charge-transfer behaviour, sensing sensitivity, catalytic efficiency and therapeutic functionality are collectively controlled by linker design, orientation/alignment, hydration behaviour and surface charge of biomolecules. The Figure 6 provides a detailed interface–function relationships as follows.

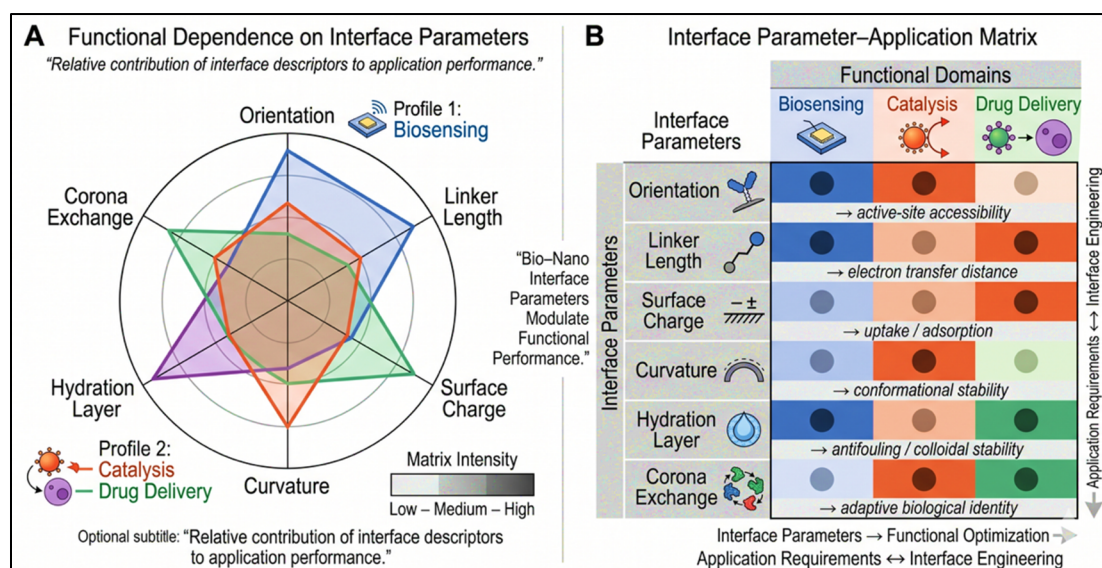


Figure 6. Interface parameters governing functional performance in biomolecule–nanomaterial systems.

Hence, it is still a challenge to develop quantitative correlations between the structure of the interface and the functional output in next generation nano–bio engineering. The applicability of these mechanistic principles is explored in the various application fields examined in next section, with particular attention to the practical realization, performance figures and implications for the translation of these principles. All these examples above illustrate the general idea that observed measurable functional performance of biomolecule–nanomaterial systems is not a result of composition alone, but rather is a consequence of intentional control of the architecture of the interfaces [9–11,38,55,80,81]. Changes in orientation, immobilization, linker design, surface functionalization, and confinement can bring about measurable enhancements in the ability to sense, catalyze, bioelectronics, and

therapeutic systems. Some representative examples of the relationship between interface modification and functional outcome are outlined in Table 8.

Table 8. Representative examples linking interface modification to functional improvement.

Interface Modification	System	Functional Consequence	Representative Metric	References
Controlled antibody orientation	AuNP biosensor	Higher target accessibility	Lower detection limit	[72,98]
Site-specific enzyme immobilization	Enzyme–MOF system	Better catalytic retention	Longer enzyme half-life	[6,34,71,86]
PEGylation	Drug nanocarrier	Reduced protein fouling	Lower cytotoxicity	[2,3,10,57]
Linker optimization	Graphene bioelectrode	Improved electron transport	Faster charge transfer	[18,22]
MOF encapsulation	Nanozyme/enzyme hybrid	Improved protection	Greater recyclability	[34,48,86]

8. Functional Applications Enabled by Biomolecule–Nanomaterial Interfaces

Although, the previous section deals with the fundamental connections between the chemistry of interfaces, structural organization, and functional behavior, the present section explores the translation of these principles to application-oriented biomolecule–nanomaterial technologies with relevance to sensing, catalysis, medicine, environmental remediation, and energy systems.

Interface chemistry plays an important role in the functional performance of biomolecule–nanomaterial systems, in addition to composition. While stable covalent interfaces are often preferred for long-term sensing or therapeutic applications, reversible noncovalent interactions are preferred in adaptive, stimuli-responsive, or self-healing systems. Likewise, coordination chemistry can be very important in catalytic nanozyme systems in controlling the accessibility of the active site and the electron transfer pathways. These chemistry–function relationships create a more predictive framework to understand why certain interface strategies are more effective than others in certain application domains [1,9–11,31,38,53].

Combination of biomolecules with nanomaterials has given way to the consideration of improved hybrid systems that can perform numerous functions in the fields of biotechnology, medicine, environmental science and energy studies. The bio-nano interface is the centre of these systems, and the complementary features of nanomaterials and biomolecules will be used to produce materials of high performance and new functions [105].

Biomolecules offer targeted molecular recognition, catalytic activity and biological compatibility and nanomaterials provide adjustable optical, electronic and catalytic properties in addition to high surface area and architecture [106,107]. The combination of these elements facilitates the creation of multifunctional platforms, which can sense, catalyze, deliver therapeutic agent and remediation of the environment. This has rendered biomolecule nanomaterial to be the focus of many of the new nano scientific and nanomedical innovations [9,11,12,49,90]. One of the merits of the biomolecule–nanomaterial hybrid systems is the incorporation of the physicochemical functionality of the nanomaterials with the selectivity, catalytic activity and adaptability of biomolecules. Hybrid systems often show measurable advantages over traditional material-based systems in terms of sensing sensitivity, catalytic stability, charge transfer efficiency, therapeutic activity and environmental functionality [6,82,84,87,95]. Table 9 shows some representative comparisons between conventional and hybrid systems.

Table 9. Representative performance comparison between conventional and biomolecule–nanomaterial hybrid systems.

Application	Conventional System	Hybrid System	Representative Improvement	Reference(s)
Biosensing	Bare AuNP sensor	Antibody–AuNP sensor	Lower detection limit/higher selectivity	[68,72,98]
Catalysis	Free enzyme	Enzyme–MOF hybrid	Higher stability/recyclability	[6,34,48,86]
Bioelectronics	Graphene electrode	DNA–graphene interface	Enhanced charge transfer	[18,22,26]
Drug delivery	Unmodified nanoparticle	PEGylated targeted nanocarrier	Reduced nonspecific adsorption	[2,3,57,87]
Remediation	Conventional catalyst	Nanozyme–biomolecule hybrid	Improved substrate specificity	[33,56,65]

8.1. Biosensing and Bioanalytical Systems

One of the most developed and investigated applications of biomolecule–nanomaterial interfaces is biosensing. In biosensor devices, biomolecules like enzymes, antibodies, nucleic acids or aptamers are recognition elements whereas nanomaterials are signal transducers or amplification platforms [108]. The biomolecule–nanomaterial hybrid systems often achieve better performance than conventional nanomaterial sensors, thanks to the increased selectivity offered by the biomolecular recognition elements, and the signal amplification and enhanced transduction efficiency of the nanomaterials. For instance, biosensors based on enzyme- or antibody-functionalized gold nanoparticles can have significantly lower detection limits than unmodified nanoparticle-based

biosensors because of the capability to boost and improve the target specificity of the biosensors. Similarly, DNA–graphene hybrid sensors have been shown to have higher electrochemical sensitivity and quicker signal response compared to graphene-based sensors alone due to the fact that they offer increased molecular recognition and charge transfer coupling. The above examples are examples of how Hybrid interface Engineering translates into measurable Analytical gains [1,5,9,38,55,82].

Nanomaterials are effective to improve the biosensor in a number of ways, such as, improved biomolecule immobilization on surfaces, improved efficacy in electron transfer and increased optical response. Indicatively, those noble metal nanoparticles are a strong plasmonic material that can be used in colorimetric identification as well as SERS. These properties facilitate extremely sensitive detection of the biomolecules, the pathogens, and the contaminants of the environment. Graphene and carbon nanotubes are carbon-based nanomaterials that have found specific applications in electrochemical biosensors due to their high electrical conductivity as well as great electron transferability. With the addition of biomolecules, these materials can be used to detect some of the most sensitive biomolecules, including glucose, neurotransmitters, and nucleic acids.

In the same manner semiconductor nanocrystals and quantum dots have been widely used in fluorescence-based biosensing because of their size tunable optical behaviour as well as high photostability. These substances allow concurrent detection of biological targets in addition to real-time observation of cellular activities [5–8,71,72]. The latest development of nano-enabled biosensors has led to development of ultra-sensitive diagnostic tools which are capable of detecting disease biomarkers in minute percentages. They are quite promising technologies in the sphere of early diagnostics of diseases and individual health care [109,110].

8.2. Catalysis and Nanozyme Design

The biomolecule nanomaterial interfaces have supported the expansion of hybrid catalytic systems with a biological selectivity and the power of inorganic nanomaterials. One of the most important types of material that are coming out of this direction is the nanozymes; these are nanomaterials that are able to mimic the catalytic action of the natural enzymes. Nanozymes have quite a few benefits over natural enzymes, which include a greater stability even at extreme conditions, the possibility of controlling their catalytic activity, their price of production. The materials can contain such performances of enzymes as peroxidase, oxidase and catalase and super oxide dismutase performance, which can enable its use in biosensing, environmental remediation and biomedical diagnostics. Even when comparing conventional nanomaterials with biomolecule–nanomaterial hybrid catalysts, how the interface can be engineered is of great importance. Although conventional inorganic catalysts in general offer high catalytic activity, they are less selective towards substrates or less compatible with biological environment. In contrast, enzyme–nanomaterial hybrids and nanozyme platforms can exploit the stability of nanomaterials as well as biomolecular recognition and catalytic specificity. The immobilization of enzymes on porous nanomaterials or conductive nanostructures has often led to greater operational stability, longer catalytic lifespan, and greater access to substrates compared to free enzymes or non-functionalized nanomaterial catalysts. The benefits of using nanozymes over natural enzymes cannot be overlooked, such as thermal stability, storage strength and low production costs, but must be balanced by practical limitations. Nanozymes often, however, have lower catalytic selectivity and weaker substrate discrimination compared to biological enzymes and this could make them less effective in highly specific sensing and/or therapeutic applications. Furthermore, the factors associated with nanoparticles, including their release of ions, persistence over time, surface reactivity, and unfortunate biological interactions, present toxicity and biocompatibility concerns, especially for biomedical application [6,9,48,55,67,86]. To overcome these challenges, further knowledge about structure–activity relationships, standardizing activity assessment procedures, and designing more application-specific and safer nanozyme platforms are necessary. The following representative literature studies demonstrate the importance of hybrid nano–bio engineering. Enzymes attached to conductive nanomaterials, for example, have been investigated with the aim of controlling the immobilization of the enzyme and optimizing the linker architecture in order to achieve catalytic retention, electron transfer efficiency and operational stability simultaneously better than for conventional adsorption approaches. Likewise, investigations using MOF based biomolecule encapsulation have shown better resistance to thermal and chemical stresses than the biomolecules in free solution. The examples highlight the significance of interface architecture to measurable functional outcomes [1,5,9–11,80,81].

Nanozymes also have a high catalytic capacity that is enhanced by the structure and chemistry of the nano–bio interface. Biomolecules adsorbed on the surface of nanomaterial may influence a catalytic activity through the modification of the active site, through the modification of substrate binding or through the transfer of electrons. Catalytic behaviour and reaction kinetics can also be affected in a biological environment when attempts are made to assemble biomolecular coronas about nanoparticles [32–37,100,104]. Nano-based enzyme immobilization

Hybrid systems, in which the enzymes are attached on the supports produced by nanomaterials are also desired. These systems increase the stability of enzymes, the catalytic activity as well as enabling the creation of multifunctional catalytic platforms capable of conducting complex biochemical reactions.

8.3. Drug Delivery and Therapeutic Platforms

The second important area in which biomolecule-nanomaterial interfaces have been applicable is drug delivery and therapeutic systems. Nanomaterials have the advantage of multipurpose carriers, which can be employed to deliver therapeutic agents to biological targets as opposed to biomolecules with their ability to target and be biologically compatible [111]. There are several advantages of the drug delivery systems produced through nanoparticles in comparison to the traditional therapy delivery approaches. These include, enhanced solubility of the drugs, controlled drug release system, and selective delivery of drugs to diseased tissues with minimum side effects. Biomolecules (antibodies, peptides or nucleic acids) may be regarded as being surface functionalized and targeted to the cellular receptors [112,113]. In addition, hybrid biomolecule–nanomaterial drug delivery systems are shown to offer benefits compared to traditional nanocarriers. Ligand targeted nanoparticles and biomolecule coated nanoplateforms may have better targeting efficiency and less non-specific interactions than non-functionalized nanoparticles, and may have greater therapeutic selectivity. The PEGylated or protein-functionalized carriers, for instance, have exhibited extensive colloidal stability and minimal protein fouling in comparison with traditional nanoparticle formulations without biomolecular surface engineering. These differences highlight the critical role of interface chemistry in controlling biological performance and translational potential [1,9–12,87–89,99].

Although considerable advances have been made in the development of nanomedicines for therapeutics, it is crucial to differentiate between proof-of-concept systems developed in the pre-clinical setting, those that have been investigated in patients, and those that have established regulatory approval. Many biomolecule–nanomaterial drug-delivery systems are still in their laboratory or animal-study stages, and their potential targeting and slow-release capabilities, along with their therapeutic effects, have been successfully tested under laboratory conditions. In contrast, the number of nanomedicine platforms that have progressed to clinical trials or regulatory approval is smaller. Thus, functional performance is only a part of the criteria for a successful translation-reproducibility, manufacturability, pharmacokinetics, safety, and regulatory compliance are additional requirements [31,82,87–89]. This translational spectrum is illustrated by representative clinically established nanomedicine platforms. One type of approved nanotherapeutics is PEGylated liposomal formulations, including Doxil[®]/Caelyx[®] (liposomal doxorubicin), which have an increased circulation time and lower systemic toxicity than normal chemotherapy. Biomolecule-assisted delivery is a feature of albumin-bound nanoparticle systems like Abraxane[®] (nab-paclitaxel) where the biomolecule is used to improve the transport and accumulation of the drug in tumors. By contrast, a large number of nanoparticle systems with targeted ligands, protein-functionalized carriers, inorganic nanomaterials and responsive biointerfaces remain under investigation, primarily in preclinical or early clinical trials, where persistent issues of long-term safety, biodistribution control and scalable manufacturing remain challenges. Hence, caution is needed when translating the results of nano–bio therapeutic studies. Beneficial results seen in cell or animal models do not guarantee clinical efficacy as there are many potential differences between biological complexity, immune interaction, pharmacokinetics, and regulatory issues that could significantly impact clinical outcomes. When talking about advanced interface engineered nanocarriers, the distinction is especially relevant since the preclinical results are very promising, while only moderate clinical validation has been achieved [87–89,95,99,112].

Nanomaterials include liposomes, polymeric nanoparticles, and inorganic nanostructures, which have to an extent been utilized in the treatment of diseases. Many of these systems are in or have already been in clinical trials or clinical use. In this instance, nanomedicine platforms can be used to deliver chemoperative drugs to tumor tissues, thereby improving the therapeutic effect and reducing treatment systemic toxicity [9,11,89,90,114,115]. In addition to drug delivery, biomolecule-nanomaterial hybrids are also under consideration in the newer treatment methods such as photothermal therapy, photodynamic therapy, gene delivery and immunotherapy [116]. In such approaches, external stimuli, such as lighting or magnetic fields, can be induced to cause some type of response to nanomaterials to apply localized therapeutic effects to a biological system. Therapeutic nanoplateforms have varying translation characteristics. Currently, lipid and polymer-based carriers have the most clinical maturity due to their favorable biocompatibility, scalability of manufacturing, and proven regulatory experience. In contrast to the optical, magnetic, and catalytic function, inorganic nanomaterials also present other challenges, such as biodegradation, accumulation over time, and toxicity testing. These differences emphasize the need to take into account the translation readiness in addition to the functional performance of nano–bio therapeutic systems. Since there is a broad variation in translational maturity around biomolecule-nanomaterial therapeutic systems, it is

crucial to differentiate the biomolecule-nanomaterial systems approved by regulators, those currently under clinical investigation and those that are mainly based on preclinical platforms [87–89,95,99,112,113]. Table 10 summarizes the key features of typical examples of composition, interface strategy, therapeutic application and state of translation of clinically relevant nano-bio systems.

Table 10. Representative clinically approved or clinically evaluated nano-bio therapeutic systems.

System	Nano-Bio Composition	Therapeutic Area	Interface Strategy	Translational Status	References
Doxil®/Caelyx®	PEGylated liposomal doxorubicin	Cancer therapy	PEGylation/lipid encapsulation	FDA-approved	[2,3,82]
Abraxane®	Albumin-bound paclitaxel nanoparticles	Oncology	Biomolecule-assisted carrier design	FDA-approved	[2,3,11]
Onpattro® (Patisiran)	Lipid nanoparticle-siRNA system	Hereditary ATTR amyloidosis	Nucleic acid encapsulation	FDA-approved	[2,3,53]
mRNA lipid nanoparticle vaccines	LNP-encapsulated nucleic acids	Infectious disease	Nucleic acid delivery	Approved clinical use	[2,3,52,111]
Targeted ligand-functionalized nanoparticles	Protein/peptide-modified carriers	Oncology/precision medicine	Active targeting	Clinical/preclinical evaluation	[11,55,87,114]
Inorganic nanoparticle therapeutics	Gold, silica, magnetic nanomaterials	Imaging/therapy	Surface biofunctionalization	Mostly preclinical/selected clinical evaluation	[3,12,55,99]

8.4. Environmental and Energy-Related Applications

In addition to biomedical and sensing applications, biomolecule-nanomaterial interfaces are now being investigated for environmental remediation, environmental monitoring, catalysis and sustainable energy technologies. In such systems, biomolecules can provide molecular recognition and catalytic selectivity, whereas nanomaterials can enhance catalytic activity and have a tunable electronic structure and high surface area. The detection and degradation of pollutants, development of biofuel cells, photocatalysis, artificial photosynthesis and renewable energy conversion are therefore a growing interest of hybrid Nano-Bio interfaces [82,84,88,108,116]. In addition to biomedical use, it has been seen that biomolecule-nanomaterial interfaces have application in environmental remediation and energy technologies. The catalytic and adsorption characteristics of nanomaterials are unique, which makes them ideal in the elimination of pollutants, breakdown of poisonous chemicals, and detection of contaminants in the environment [117]. Nanozyme-based catalytic systems have been recognized in degrading organic contaminants, detoxification of environmental pollutants and biosensing of toxic substances. Nanomaterials can also become biomolecule-functionalized and become more selective in pollutant detection and removal. Hybrid systems offer significant benefits over traditional materials in environmental and energy applications when combined with catalytic functionality and biomolecular selectivity or adaptive response. The performance of the biofunctionalized photocatalysts, enzyme-electrode assemblies, and nanozyme-based remediation systems have often been improved compared to the same systems without biofunctionalization in terms of efficiency of pollutant transformation, charge-transfer performance, and/or substrate specificity. However, the benefits need to be weighed against the extra challenge of biomolecule stability, catalyst recovery and production price [82,84,88,117].

Biomolecule-nanomaterial systems have demonstrated excellent potential in the field of pollutant degradation and environmental sensing, including contaminants removal and sensing such as heavy metals, dyes, pharmaceuticals, pesticides, and emerging pollutants. Enzyme-nanoparticle hybrids and nanozyme systems are capable of catalysing oxidation, reduction or degradation pathways under relatively mild conditions of operation. Similarly, the use of functionalized carbon nanomaterials and porous frameworks for adsorption enhanced remediation and catalytic transformation of pollutants has been studied [65,82,84,108,116]. Many of the reported systems, however, are tested in simplified laboratory media, and they often are found to exhibit a lower catalytic efficiency and selectivity in actual wastewater media where other ions, organics, pH variations, and complex mixtures of contaminants may compete with the catalyst and diminish its activity. The implementation of the practical nano-bio remediation technologies also needs to be considered, such as catalyst recovery, fouling resistance, long term stability and process reusability. The nanocatalysts that are dispersed in water can have very high activity and are hard to be efficiently recovered after treatment. Immobilization strategies, magnetic supports and porous host architectures have thus been examined to enable the catalyst to be recycled and to minimize the risks of secondary contamination. However, fouling, biomolecular degradation and progressive loss of catalytic activity are still significant challenges to continuous operation [32,48,56,65,117].

Biomolecule-nanomaterial hybrids have found applications in energy-related technologies, including biofuel cells, photocatalytic and renewable energy technologies. Immobilized enzymes on conductive nanomaterials

would support effective transfer of electrons in biofuel cells and therefore biochemical energy can be converted into electrical energy. Biomolecule–nanomaterial interfaces have been broadly studied in the energy sector in biofuel cells, photoelectrochemical systems and platforms for artificial photosynthesis. Bioelectrochemical devices can be improved by using conductive nanomaterials (such as graphene, carbon nanotubes, and metallic nanostructures) functionalized with enzymes and proteins to increase charge transfer, catalytic turnover, and/or electrode stability. Likewise, the use of semiconductor nanomaterials coupled to biomolecular catalysts has been of interest in the context of photo assisted redox reactions and solar-assisted production of fuels from these. Biological selectivity, coupled with the ability to transport electrons with the aid of nanomaterials, opens the door to more effective systems for renewable energy. While there is promising progress, there are still some challenges that impede translation of nano–bio energy technologies [65,82,84,88,116]. Long-term stability, biomolecular deactivation, inefficient electron-transfer coupling and loss of catalytic activity on device operation are common problems that affect device performance. The ability to keep enzymes active and to ensure good electrical communication at the electrode interface is a recurring problem in biofuel cells. Likewise, to make widespread use of artificial photosynthesis systems viable, problems of catalyst durability, quantum efficiency, and scaling up reactor design need to be overcome. Broader evaluation of economic feasibility, lifecycle sustainability and manufacturing scalability is also necessary in environmental and energy applications. A number of laboratories have developed nano–bio systems with good laboratory-scale performance, but various factors such as synthesis complexity, high raw costs, high energy consumption for fabrication, and unpredictable end-of-life behaviour can make large-scale deployment more difficult. Lifecycle assessment approaches are thus an important new tool to help assess the environmental impact, resource consumption, and sustainability impacts of nano-enabled technologies. The integration of green synthesis approaches, catalyst architectures that are easy to recycle and scalable manufacturing processes will probably play significant roles for future practical use [53,82,84,88]. Moreover, nanomaterials that have been templated by biomolecules have been considered in artificial photosynthesis systems and catalytic nanoreactors to generate sustainable energy. These systems act as analogous to natural biological processes to transform solar energy to chemical fuels or electric power. Biological functionality combined with nanoscale material integration thus offers good prospects of coming up with sustainable technologies in both environmental and energy concerns. Table 11 summary of functional applications enabled by biomolecule–nanomaterial interfaces, highlighting representative systems, working principles, and advantages across key areas such as biosensing, nanozyme catalysis, drug delivery, bioelectronics, and environmental remediation.

Environmental and energy-oriented biomolecule-nanomaterial systems have great functional potential, but have significant practical challenges in terms of operational stability, catalyst recovery, fouling resistance, performance in real environments, scalability and economic viability [32,48,108]. Table 12 provides a comparative summary of representative systems, their strengths, weaknesses and translation issues.

Table 11. Comparative functional applications of biomolecule–nanomaterial interfaces.

Application Area	Representative Interface System	Working Principle	Typical Characterization/Validation	Major Advantages	Key Limitation/Translational Challenge	Representative Performance Indicator	Application Readiness	Ref.
Biosensing & Diagnostics	DNA/antibody–AuNPs, graphene bioelectrodes	Optical (SPR, fluorescence) and electrochemical signal transduction	SPR, electrochemistry, fluorescence spectroscopy	High sensitivity, multiplexing, rapid detection	Biofouling, nonspecific adsorption, matrix interference	Lower detection limits, improved selectivity	High/Commercially mature	[20,21,98]
Electrochemical Bioanalysis	Enzyme–nanomaterial electrodes, MXene/graphene interfaces	Enhanced electron transfer and signal amplification	Cyclic voltammetry, impedance spectroscopy	Real-time monitoring, high selectivity	Electrode fouling, enzyme instability	Reduced charge-transfer resistance, higher signal response	High–Moderate	[18,23,109]
Nanozymes & Catalysis	Metal/metal oxide nanozymes, biomolecule-modified catalysts	Enzyme-like catalysis (peroxidase, oxidase, catalase mimicry)	Michaelis–Menten kinetics, activity assays, spectroscopy	High stability, lower cost, tunable activity	Lower substrate specificity, unclear active sites, reproducibility concerns	Enhanced catalytic turnover, recyclability	Moderate–High	[32,35,100]
Drug Delivery & Therapeutics	Polymer/lipid nanoparticles, protein-functionalized nanocarriers	Targeted delivery, controlled release, receptor-mediated uptake	Drug release assays, DLS, cellular uptake studies	Improved bioavailability, reduced systemic toxicity	Scale-up, pharmacokinetics, regulatory challenges	Drug-loading efficiency, reduced cytotoxicity	High (several clinical systems)	[2,111,113]
Cancer Therapy (Photothermal/Photodynamic)	Gold nanostructures, photosensitizer-loaded nanomaterials	Light-triggered heat or ROS generation	Cell viability assays, imaging, photothermal analysis	Target specificity, minimally invasive therapy	Light penetration depth, off-target effects	Tumor suppression efficiency, ROS yield	Moderate–High/Clinical investigation	[29,114,115]
Bioelectronics & Neural Interfaces	Graphene bioelectrodes, flexible nano–bio devices	Charge transfer and electrical signal modulation	Electrochemical impedance, conductivity measurements	High conductivity, flexible interfacing, biocompatibility	Long-term stability, interface degradation	Faster electron transfer, improved signal fidelity	Moderate/Emerging	[22,110]
Environmental Remediation	Enzyme–nanomaterial catalysts, nanozyme systems	Catalytic pollutant degradation and adsorption	Catalytic assays, pollutant analysis, recyclability tests	High efficiency, catalyst reuse	Fouling, catalyst recovery, real wastewater performance	Higher degradation efficiency, recyclability	Moderate	[13,33]
Energy Applications	Biofuel cells, enzyme–electrode interfaces	Biocatalytic energy conversion and electron transfer	Electrochemical testing, power-density analysis	Renewable energy generation, catalytic selectivity	Enzyme instability, scalability limitations	Power density, operational lifetime	Moderate/Experimental	[11,58]

Table 12. Environmental and energy applications of biomolecule–nanomaterial interfaces: opportunities and challenges.

Application	Representative System	Major Strength	Key Limitation	Translational Challenge	References
Pollutant degradation	Nanozyme/enzyme–nanoparticle systems	Catalytic efficiency	Fouling/recovery	Real wastewater performance	[33,48,56]
Environmental sensing	Functionalized biosensors	High sensitivity	Matrix interference	Field deployment	[19,20,56,100]
Biofuel cells	Enzyme–graphene/CNT electrodes	Charge transfer	Enzyme instability	Long-term operation	[18,22,71,110]
Photocatalysis	Semiconductor–bio hybrids	Light harvesting	Catalyst degradation	Scalability	[34,82,84,85]
Artificial photosynthesis	Bio–semiconductor interfaces	Solar fuel generation	Low efficiency	Reactor engineering	[5,38,82,84]

9. Stability, Biocompatibility, and Environmental Considerations

Even though the interface of biomolecules and nanomaterials can be useful in an array of both technological and biomedical applications, their possible use must be supported by the special attention to the aspects of stability, biocompatibility, and environmental. In such systems, nanomaterials tend to exhibit complex reactions with the surrounding biomolecules, ions, and the environment that have an influence on their behaviour. These interactions can alter the physicochemical properties of nanomaterials, their behaviour, functionality and long term behaviour.

Although tremendous progress has been made, there are still some basic challenges that hinder the translation of biomolecule–nanomaterial systems towards real-world applications. Reproducibility is one challenge that is frequently encountered: small differences in the size, morphology, surface chemistry and biomolecule loading of the nanoparticles often result in significant differences in interfacial behavior. The heterogeneity of interfaces makes the mechanistic interpretation and comparison between studies even more intricate. Additionally, there is a lack of a thorough and standardized approach to nanotoxicity assessment, and the results of *in vitro* and *in vivo* studies are still not correlated. These limitations will need to be overcome through standardized characterization procedures, better reporting practices and systematic structure–activity relationships analysis. One of the major themes that arose from the development of nano-bio interface engineering is that the behaviour of a bio and environmental system is greatly influenced by the design of the interface, and not only the material. These parameters include sustainability-oriented synthesis approaches, as well as surface functionalization, charge tuning, adsorption properties, dissolution properties and toxicity, which can all affect performance, toxicity, environmental fate and translational viability [2,3,12,82,84,88,89]. The following Table 13 summarizes representative relationships between the various design decisions made for the interface and the biological/environmental effects.

Table 13. Relationship between interface design parameters and biological/environmental outcomes.

Design Parameter	Intended Benefit	Biological/Environmental Consequence	Potential Trade-Off	References
PEGylation	Reduce fouling, improve circulation	Reduced protein adsorption	Lower cellular uptake	[2,3,10,57]
Positive surface charge	Improve cellular uptake	Increased membrane interaction/toxicity	Reduced biocompatibility	[9,12,16,49]
Hydrophobic surfaces	Strong adsorption	Enhanced protein corona formation	Aggregation risk	[10,29,57,59]
Metal nanoparticle dissolution	Catalytic activity	Ion release/altered toxicity	Stability loss	[12,15]
Controlled biodegradation	Clearance	Reduced persistence	Reduced durability	[2,87,112]
Green synthesis/LCA-informed design	Sustainability	Lower environmental footprint	Possible performance trade-offs	[13,84]

Conformational changes or dynamic exchange processes that effect the stability and biological identity of nanoparticles can also be applied to biomolecules that adsorb onto the surfaces of nanomaterials at nano bio interface. The products of these processes might often involve the formation of biomolecular coronas and have the ability to influence nanoparticle bio-distribution, uptake within whole cells and immune reaction [1–3,29,99,118]. Hence, stability and conversion of nano-bio interfaces can be known to ensure that the use of the interfaces in medicine, environment technologies and industrial processes is safe and efficient. Another important factor for biocompatibility and toxicity is surface charge. The enhanced interaction with negatively charged cellular membranes is often the result of positively charged nanomaterials, as this can lead to amplified uptake but also to higher membrane disruption, oxidative stress, and cytotoxicity. However, neutral or slightly negative surfaces may exhibit less nonspecific biological interactions and better circulation behaviour. Therefore, depending on the

application context, it is necessary to optimize the charge characteristics in order to get a good delivery efficiency, a good colloidal stability and a good safety level [10–12,38,55,89,95]. Dissolution, degradation and environmental transformation processes also influence the behaviour of nanomaterials. Bioactive metal species released from partial dissolution of metallic or metal oxide nanoparticles may change their catalytic activity and toxicity profile, as well as their ionic composition. Similarly, during biological or environmental exposure, the chemistry of interfaces can significantly change as a result of oxidation, aggregation, surface restructuring and exchange of biomolecules. These dynamic changes can alter the bio–nano interactions, catalytic behaviour, biodistribution, and environmental persistence of nanorobots, and thus warrant consideration of the nano–bio interfaces beyond their original synthesized state. Nano-materials can form eco-coronas in environmental systems that consist of natural organic matter, proteins, polysaccharides, humic substances or environmental metabolites. Eco-corona formation, like biological protein coronas, alters the surface chemistry, aggregation behaviour, transport properties, and biological interactions. Most importantly, the composition of the eco-corona depends strongly on the characteristics of the interface (hydrophobicity, surface functionalisation, charge state). These relationships can help inform rational design of nanomaterials that are more environmentally compatible and have fewer unintended ecological effects [53,82,84,88].

In addition, the nanomaterials that have been moved to the biological or ecology system may be chemically transformed, aggregated or degraded and this influences its ecological impact. Interdisciplinary approaches, which are founded on the combination of nanochemistry, toxicology, environmental science, and life-cycle assessment, are used as the scientific method of the evaluation of these processes. Upon exposure to biological environments, nanomaterials interact with surrounding biomolecules, leading to the formation of a protein corona that defines their biological identity, as illustrated in Figure 7.

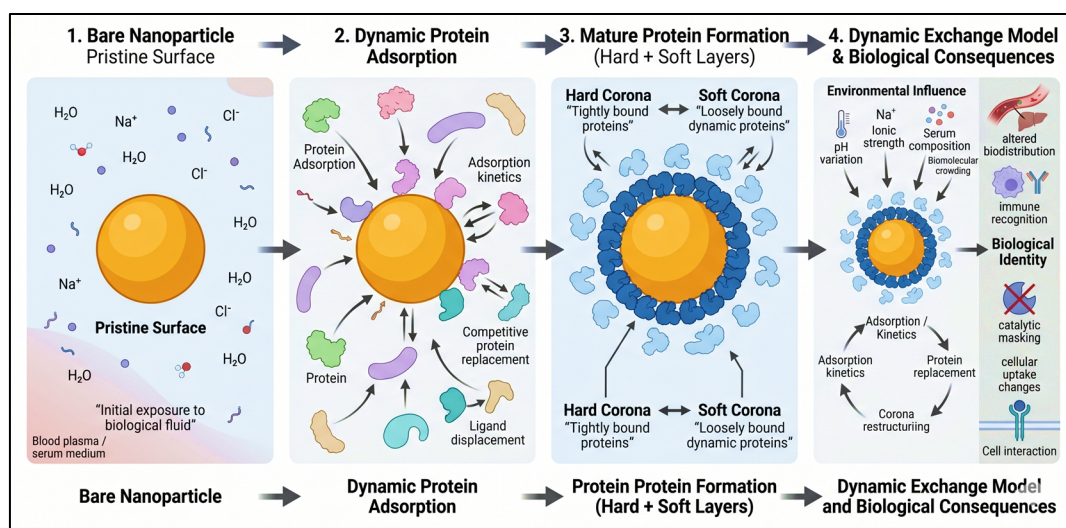


Figure 7. Competitive adsorption, soft/hard corona transition, protein exchange process, hydration effect, and environment modulation effect are shown in the dynamic model of protein corona formation at biomolecule–nanomaterial interfaces. The scheme also highlights the impact of corona remodelling on the identity, biodistribution, immune interactions, catalytic accessibility, biosensing performance, and drug delivery behaviour of a nanoparticle.

A recurring theme in both stability and environmental issues is that safety and functionality are not independent design parameters, but rather interrelated outcomes of interface chemistry. Surface functionalization, type of linker used, charge tuning, biomolecule coating, nanomaterial composition, cellular interactions, environmental persistence, and biomedical viability are all affected by decisions made during the surface functionalization of nanoparticles, all of which influence catalytic performance, biomolecular stability, protein/eco-corona formation, and cellular interactions. It is therefore crucial to implement these relationships in the design of interfaces for the development of safer, more efficient and more sustainable nano–bio systems [31,53,64,82,84,88].

9.1. Chemical and Structural Stability of Interfaces

Biomolecule nanomaterials intermolecular interactions define the stability of the biomolecule nanomaterial interface, the environmental conditions and surface chemistry of the nanomaterial. The covalent bonds and coordination interactions usually are more stable compared to noncovalent interactions, such as electrostatic attraction and hydrogen bonding. Interactions by creating noncovalent interactions can provide dynamic and

reversible binding also which may be useful in certain biological applications. Conditions that can substantially affect interface stability include environmental conditions like pH, ionic strength, temperature as well as the presence of competing biomolecules. Within the biological fluids, nanoparticles tend to come in contact with complicated mixtures of proteins, lipids and metabolites that can rival surface binding sites. This could result in the development of dynamic protein coronas that could displace originally bound biomolecules and modify the behaviour of nanoparticles.

Surface functionalization of nanomaterials is an important nano-biological interface stabilizer. Physiological aggregation can be prevented by protecting the agglutination of the colloidal particle using protective coatings e.g., polymers, biomolecules or inorganic shells. These approaches are especially necessary in biomedical uses where a nanoparticle has to be stable in complicated biological conditions. Besides, the catalytic activity, electronic properties and the biomolecule functionality can be affected by structural stability at the nano- bio interface. Indicatively, adsorption of proteins can cause changes in protein conformation that can inhibit the action of the enzyme, or modify biomolecular recognition properties. So, the creation of constant interfaces that do not destroy the structure of biomolecules is one of the key challenges in nano- bio engineering.

9.2. Biocompatibility and Toxicological Aspects

The use of nanomaterials on biological systems is one area where biocompatibility is a critical issue. Most nanomaterials have potential biomedical benefits, but they can also have undesirable biological consequences, especially when not carefully engineered, when interacting with cells and tissues. Nanomaterials are toxic materials whose toxicity depends on various factors such as the size of the particles, the charges of the surface of the particle, the chemical composition or surface functionalization. Smaller nanoparticles are often more reactive and are able to penetrate biological membranes with greater ease, which could result in greater cellular uptake and cellular toxicity. In the same way, surface charge contributes significantly to cell membrane and protein interactions.

Some nanomaterials also could produce reactive oxygen species (ROS) or leech metal ions which could cause oxidative stress, inflammation, or damage to cells. For example, metal oxide nanoparticles can go through dissolution mechanisms releasing metal ions that can interact with biological molecules [119]. Strategies to modify the surface of nanoparticles can greatly enhance the biocompatibility of nanoparticles. Nonspecific interactions with biological components can be minimized by functionalization with biocompatible molecules, e.g., polymers, peptides, polysaccharides, etc. to minimize immune responses. Moreover, biomolecule coating can assist in replication of natural biological settings, hence enhancing nanoparticle bio-compatibility with living organisms [18,21,22,120,121]. To guarantee safe uses of biomolecule-nanomaterial hybrids in medicine, diagnostics and biotechnology, understanding of the toxicological behavior of these hybrids is therefore necessary.

9.3. Degradation, Transformation, and Lifecycle Analysis

Nanomaterials do not often exist in a chemical form after being incorporated into a biological or environmental system. Rather, they usually experience chemical conversion, disintegration, aggregation, or dissolution that changes their characteristics and determines their behaviour in the long term. Here are examples of effects that can occur owing to interaction with biomolecules or environmental components: nanoparticle surface chemistry or structural integrity can change. Metal nanoparticles can be oxidized or dissolved, whereas nanomaterials made of polymers can be degraded by hydrolysis or enzyme actions. Such changes may change the toxicity, mobility and persistence of nanoparticles in the environment.

Nanomaterials can also react with natural organic matter, microorganisms and mineral surfaces in the environment. These interactions can cause the creation of so-called eco-coronas, similar to protein coronas in biology, and they can have a strong impact on the fate and transport of nanoparticles in the environment [13,15,122–124]. Lifecycle analysis (LCA) offers a valuable model of assessing the environmental effects of nanomaterials in their manufacturing and use, to their disposal or degradation. After evaluating the environmental impact of the production and use of nanomaterials, scientists can determine the risks involved and come up with a more environmentally friendly solution in nanotechnology. This means that the pathways of degradation and lifecycle dynamics are important in the design of nanomaterials that can be considered as functional and as well as environmentally responsible. Besides the enhancement of catalytic efficiency, future development of nanozyme technologies will rely on the ability to overcome a number of major limitations, such as specificity, mechanistic understanding, reproducibility, and translational value. The identification of the real catalytic active sites and the development of quantitative structure–activity relationships and benchmarking with natural enzyme systems in realistic operational conditions are needed. Combining advanced characterization, computational modelling, and

AI-driven catalyst optimization could open up new avenues that address current limitations and predict nanozyme design [47,53,64,82,83].

10. Challenges and Current Limitations

Although impressive progress has been achieved in the construction of interfaces between biomolecules and nanomaterials, there are a number of scientific and technological implications, which restrict the application of these interfaces to more practical use. The main problems are associated with the complexity of nano- bio systems, with the dynamical interaction of biological macromolecules with engineered nanomaterials in changing environmental conditions. The ability to control interfacial structure on a very fine scale, to reproducibly fabricate systems, and the ability to describe these complex systems accurately are still challenges.

A key challenge is the dynamism and heterogeneity of nano- bio interfaces in which biomolecules can change their structure or exchange with the surrounding biomolecular species. These types of processes may modify the functional behaviour of nanomaterials and provide variability in the performance of these materials across various experimental conditions [1,4,31,99]. These limitations can be met by further insights into the mechanics of molecular interactions at the nano-bio interface and by creating new powerful characterization methods which can probe these systems in realistic biological conditions.

10.1. Control over Orientation and Density of Biomolecules

The ability to control accurately the orientation and surface density of biomolecules on nanomaterial surfaces is one of the primary challenges to nano- bio interface engineering. The spatial orientation and accessibility of biomolecules at the interface is often essential to the functional performance of biomolecule -nanomaterial systems. In one example, enzymes immobilized on nanomaterial surfaces need to have an orientation that does not block access to their catalytic active sites. Proteins can be adsorbed randomly onto the surface of nanoparticles resulting in unfavourable orientations or conformational alterations that decrease catalytic reactivity or alter molecular recognition properties. Likewise, the antibody-functionalized nanoparticles to be used as biosensors must be oriented correctly with regard to the antigen-binding sites in order to be used effectively. The strategies most commonly used to control the orientation of biomolecules include site-specific conjugation, affinity based immobilization or the design of molecular linkers. But, the strategies are usually constrained by the complexity of biomolecular structures and the number of nanomaterial surface chemistries. Attaining consistent and uncontrolled biomolecule coverage on nanoparticle surfaces thus continues to be a major field of study.

10.2. Reproducibility and Scalability

A second critical issue in the biomolecule-nanomaterial interface engineering is the challenge of reproducing and scaling fabrication methods. Nano-bio hybrid systems are being developed in large numbers today under tightly controlled conditions in the laboratory and might not eagerly be shifted to industrial scale manufacture. The synthesis of nanoparticles, their functionalization of surfaces, and immobilization of biomolecules can be variable, which makes nanoparticles, surface, and functionality erratic. Minor differences in the size of nanomaterials or their surface characteristics may lead to huge biological reaction and catalysis differences [55,57,59,67]. There are also technical difficulties associated with scaling up the fabrication of nano- bio interfaces. Mass production should provide quality, stability and safety with cost effective production processes. This is especially critical in the use of nanomaterials in nanomedicine where stringent regulations are applied in the development of therapeutic nanomaterials. The progress in automated synthesis, microfluidic fabrication technologies and standardized characterization protocols can be used to enhance reproducibility and scalability in nano- bio interface engineering.

10.3. Interface Complexity and Characterization Gaps

The nano bio interface is a very complicated system with several interacting components, such as nano material surface, biomolecules, solvent molecules and the biological environments surrounding them. This is very complicated such that one can hardly understand and predict the behaviour of these systems. The development of biomolecular coronas that involve the dynamic adsorption of proteins and other biomolecules on the surfaces of nanoparticles is one of the most significant challenges in biological conditions. Such coronas have the potential to dramatically manipulate nanoparticles to affect cellular uptake, biodistribution and immune responses. Nevertheless, it is still hard to describe the structure and the dynamics of these coronas because of their non-persistent and non-homogenous character [32,53,54].

The other constraint is the existence of analytical methods that can investigate nano- bio interfaces at the required spatial and temporal resolution. Although microscopy and spectroscopy techniques can give useful information, the sample preparation procedures used in many methods can modify the natural structure of the interface. The emerging characterization gaps will be filled by developing advanced in-situ characterization techniques, computational characterization techniques and multiscale characterization techniques. Such tools will empower the researchers to gain a bigger understanding of the dynamics in the process occurring at the nano- bio interfaces and develop the rational design of more dependable and operative hybrid systems. Real-world validation, operational stability, and techno-economic evaluations must be emphasized in the future development of environmental and energy-related biomolecule–nanomaterial systems. More focus should be given to performance evaluation in realistic environmental matrices, to catalyst recovery strategies, and to performance in the long term and scale up compatibility. The inclusion of sustainability metrics, life cycle analysis and circular design considerations could help in the creation of environmentally-friendly nano–bio technologies [84,88,122–124]. Lifecycle analysis (LCA) is a new tool that increasingly is being used to help inform the development of safer and more sustainable nanomaterials. Lifecycle approaches are not only based on performance but also take into account raw-material sourcing, synthesis requirements, operational stability, environmental release, degradation behavior, recyclability and end-of-life management. These evaluations can help inform the material-selection process, by establishing trade-offs between functional efficiency, toxicity risk, economic costs, and environmental footprint. The integration of lifecycle thinking with interface engineering can then be used to help design a more responsible next generation biomolecule–nanomaterial system [31,53,88,122–124].

11. Future Perspectives and Emerging Directions

The rapid growth of the interface engineering of biomolecules with the nanomaterials created the opportunities connected with the design of the hybrid materials with new functions. However to transform these improvements into practical technologies, several scientific and technological challenges are to be overcome. The future studies in this area will hopefully deal with rational interface design, predictive modelling, combination of computational and artificial intelligence applications, and large scale application of nano bio systems. Nano-bio interaction complexity, together with the dynamic behaviour of a biological environment, requires a more detailed mechano-level perspective of molecular interactions at the interface. The development of materials science, computational science and interdisciplinary teamwork will be significant in informing the future generation of biomolecule-nanomaterials platforms to be used in healthcare, energy and environmental technologies.

11.1. Rational Interface Design and Predictive Models

The approach of creating rational design strategies that would allow controlling interfacial structure and functionality is one of the most important avenues of biomolecule-nanomaterial interface research in the future. Conventional methods usually make use of experimental trial and error to determine appropriate biomolecule and nanomaterial combinations. Nevertheless, these methods are time consuming and may not be able to properly represent the dynamics that take place within the nano-bio interface.

The future studies are anticipated to be on predictive models which can be developed by integrating physicochemical principles with experimental data to direct the development of functional nano-bio systems. These models are useful to predict the interaction of biomolecules with the surface of nanomaterials, the effects of adsorption processes on biomolecular conformation, and the effects of interfaces on catalysis or electronics.

The ability to appreciate the effects of parameters like nanoparticle size, surface charge, ligand chemistry and environmental conditions will enable the researchers to develop interfaces that optimize functional activity whilst staying stable and being biocompatible. The models will also find their application in optimization of nanozyme catalysts, biosensor platforms and nano-particles of therapy.

11.2. Computational and AI-Assisted Interface Engineering

Computational chemistry and artificial intelligence will probably make a breakthrough in interface engineering with biomolecules and nanomaterials. The interactions between molecules at the nano- bio interfaces can also be studied at the level of computational tools such as molecular dynamics simulations, density functional theory (DFT) and multiscale modelling. Such simulations may give information regarding the binding of biomolecules on the nanomaterial's surface, disclose conformational alterations of biomolecules in adsorption, and forecast the effects of surface functionalization on electronic and catalytic functions. These kinds of computational methods are used to supplement experimental research by providing atomistic-level insights into interface development.

These simulations may assist in understanding the binding mechanism of biomolecules on the nanomaterial surfaces, the conformational variations of biomolecules upon adsorption, and determine the effect of surface functionalization on electronic and catalytic characteristics. These methods of computation are used to complement experimental research to provide atomic-level insight into the formation of interfaces. Machine learning and artificial intelligence are also becoming potent solutions to accelerate the materials discovery and interface optimization. Through statistical methods of examining big data sets of experimental and computer-based research, AI algorithms can communicate trends and determine the best combinations of nanomaterials, ligands, and biomolecules to utilize in certain applications. The ability to predict the structure, energetics, and functional characteristics of interfacial structure and behaviour before synthesis is becoming possible with computational modelling, which is increasingly appearing as a powerful complement to experimental nano–bio interface engineering. Molecular dynamics (MD) simulations can be used to investigate biomolecule adsorption pathways, conformational stability, organization of water molecules in the hydration layer, evolution of the protein corona and the orientation of enzymes on the surface of nanomaterials under realistic environmental conditions. In smaller spatial and electronic dimensions, density functional theory (DFT) calculations could be employed to develop mechanistic understanding of the binding energetics, charge redistribution, catalytic active sites, and changes in surface reactivity upon biomolecule attachment. These strategies provide molecular-level understanding of the role of interface chemistry on function [1,55,62–64,82]. Coarse-grained and multiscale simulation methods are becoming more significant in bridging the atomistic detail and the experimentally-relevant complexity that is common to biomolecule–nanomaterial systems. Employing coarse-grained models, long timescale adsorption dynamics, corona evolution, and aggregation of nanoparticles, which are currently beyond the simulation range of fully atomistic models, can be simulated. These are especially effective for the study of competitive adsorption processes, restructuring of interfaces, and collective nano–bio behaviours in crowded biological environments. The use of machine learning (ML) and data-driven materials discovery frameworks for enhancing the rational design of nano–bio interfaces is growing in popularity. The establishment of relationships between the surface chemistry, physicochemical descriptors, biomolecular properties and experimentally determined performance metrics enables ML models to help predict binding affinity, adsorption propensity, toxicity profile, catalytic activity, and colloidal stability. These strategies can significantly decrease the number of empirical screens and help to identify the best material–biomolecule combinations in vast design spaces. Emerging graph neural network (GNN) approaches provide further possibilities to represent complex nano–bio systems as these can naturally encode molecular connectivity, surface topology and interaction networks. By combining structural and physicochemical information into predictive learning architectures, GNN-based frameworks could help to predict biomolecule–surface binding behavior, protein corona composition, adsorption selectivity, enzyme orientation, and the toxicity at the interface. While still in its infancy, these kinds of methods could be useful in the inverse design of application-specific biomolecule–nanomaterial interfaces [53,62–64,82].

Computational tools are now becoming a more important means for the rational design of the biomolecule–nanomaterial interface. The opportunities to correlate surface chemistry, biomolecular structure and functional performance through the use of machine learning, molecular dynamics simulations and data-driven predictive modelling are endless, with the ability to span complex multidimensional datasets. AI-powered methods can be used to speed up prediction of binding affinity, interface stability, toxicity profile, and catalytic behaviour and minimize trial-and-error experiments. Nevertheless, there are still barriers that need to be overcome for widespread adoption, such as the quality of available high-quality datasets, model transferability and interpretability [31,53,62–64,82].

Such methods can facilitate the construction of knowledge-based design models that can quickly discover high-performance nano–bio structures in the use of biosensing, nanomedicine, and catalytic technologies [125]. Closed loop interface design workflows with the integration of simulation, machine learning and experimental validation are promising directions. For instance, DFT calculations can be used to identify binding motifs which are energetically favourable, MD simulations can be used to assess the conformational stability and the dynamics of adsorption, and machine-learning algorithms can then optimize candidate materials based on experimental-performance datasets. These hybrid approaches may enable a predictive design of nano–bio systems with specific properties such as controlled orientation of biomolecules, minimization of non-specific adsorption, increased catalytic activity and improved safety profiles [53,55,62–64,82,88].

11.3. Toward Real-World and Industrial Implementation

Although several biomolecule–nanomaterial systems have been shown to perform well in laboratory, their implementation in practical conditions needs to consider some practical concerns. These are large scale production, long term consistency, government regulation and environmental protection. The nano–bio technologies

implemented in industrial practice will entail a means of scaling the synthesis, consistent fabrication, and a uniform quality control procedure. Improvement in methods of synthesizing nanomaterials through continuous-flow reactors and automated systems of nanofabrication can be employed to bring consistency to the production of high quality nanomaterials. Moreover, the regulatory frameworks should focus on the safety and environmental effects of nanomaterials in use in biomedical and industrial use. To be able to develop nano-enabled technologies in a responsible way, risk assessment, nano-enabled technologies lifecycle analysis and sustainable design will be required.

The translation of biomolecule-nanomaterial interfaces to practical devices will probably result in the creation of new products in such domains as personalized medicine, environmental systems, renewable energy systems, and smart materials. Interdisciplinary interactions will continue to intensify and improve next generation nano-bio systems with additional interactions between nanotechnology, biotechnology, computational science and engineering. Table 14. Recent advances and emerging trends in biomolecule-nanomaterial interface research, summarizing representative studies with respect to nanomaterial type, biomolecule integration, key innovations, and application domains. The table highlights current progress and future directions in the field.

Table 14. Recent advances and emerging trends in biomolecule-nanomaterial interfaces.

Year	Nanomaterial	Biomolecule	Key Innovation	Application	Reference
2022	Metal nanoparticles	Proteins (corona systems)	Understanding protein corona dynamics and bioidentity	Nanomedicine, drug delivery	[10]
2022	Nanomaterials (general)	Multiple biomolecules	Engineering nano–bio interfaces for functional nanomedicine	Biomedical applications	[9]
2023	Carbon-based nanomaterials	Biomolecules (general)	Biomimetic design strategies for sensing and therapeutics	Biosensing, theranostics	[60]
2023	Self-assembled nanomaterials	Proteins, peptides	Interfacial colloidal self-assembly for functional materials	Smart materials	[39]
2024	Graphene-based systems	Proteins (bioelectrodes)	Advanced electrical stimulation and biointerface control	Bioelectronics	[22]
2024	MOFs/porous frameworks	Biomolecules	Tunable porous nano–bio interfaces	Drug delivery, catalysis	[86,87]
2025	DNA-directed nanomaterials	DNA	Programmable assembly of photonic nanostructures	Diagnostics, therapeutics	[25,79]
2025	Nanozymes	Proteins/biomolecular corona	Advanced nanozyme–bio interface engineering	Catalysis, biomedical therapy	[32,35]
2025	Liquid–liquid phase separation systems	Proteins, biomolecular condensates	LLPS-inspired nanomaterials and adaptive systems	Drug delivery, smart materials	[42,46]
2026	Bio-inspired nanomaterials	Biomolecular condensates	Adaptive nano–bio materials from biomolecular assemblies	Next-generation nanomaterials	[42]

Computational methods also provide valuable possibilities for predictive nanotoxicology. Modelling, molecular simulations, and quantitative structure–activity relationship (QSAR)-type approaches may be able to predict biological response, immune interactions, environmental toxicity and other properties based on the surface charge, dissolution properties, size, surface functionalization, and composition of the corona. These predictive capabilities can guide the development of nanomaterials to be safer and minimize the need for extensive experimental screening. For biomolecule-nanomaterial systems, that covers a range of spatial, temporal and physicochemical scales, there is no single computational approach that will be able to predict the behavior of the interface. Rather, atomistic simulations and machine learning-driven predictive models are becoming more interwoven to model adsorption dynamics, corona formation, binding energetics, toxicity and interface optimization in complementary ways [53,62–64,82,88]. Table 15 summarizes a comparative overview of the major computational tools that are relevant to predictive biomolecule-nanomaterial engineering.

Table 15. Computational tools for predictive biomolecule–nanomaterial engineering.

Method	Primary Capability	Example Application	Major Limitation	Reference(s)
MD (Molecular Dynamics)	Adsorption dynamics	Protein orientation/corona formation	Computational cost	[5,10,31,57]
DFT (Density Functional Theory)	Electronic structure	Binding energetics/catalysis	Small system size	[5,31,32,48]
Coarse-grained simulation	Large-scale dynamics	Aggregation/long-timescale adsorption	Reduced molecular detail	[5,10,31,38]
ML (Machine Learning)	Property prediction	Toxicity/affinity prediction	Dataset quality dependence	[12,27,53,64]
GNN (Graph Neural Networks)	Structure-aware prediction	Corona composition/adsorption	Data availability	[27,53,64,91]
Data-driven discovery	Material screening	Interface optimization	Model interpretability	[9,12,55,82]

12. Conclusions

Biomolecule–nanomaterial interfaces are an emerging field of research at the crossroads of chemistry, materials science, biotechnology and nanomedicine. Yet, the systems must not only combine biomolecules and nanomaterials, but also control the architecture and chemistry of the interfaces. The biomolecule orientation, surface density, linker design, charge characteristics and properties of the hydration state, as well as dynamic protein/eco-corona formation, plays a significant role in the sensing performance, catalysis efficiency, charge-transfer behavior, therapeutic efficacy, environment response, and operational stability of the biosensors, as highlighted throughout this review.

The desire to achieve functional improvements, combined with robustness, biocompatibility and the feasibility of translation towards the clinic, is a recurring challenge in all biomolecule classes, nanomaterial platforms and applications. However, several challenges remain that limit the widespread use of nano–bio technologies, such as controlling the evolution of the corona, loss of biomolecular activity upon adsorption, toxicity, transformation of nano–materials, batch-to-batch variation, limitations of reproducibility and scalability. Decisions on these issues must be made beyond simple selection of materials, and requires a shift toward rational interface engineering based on chemistry–structure–function relationships.

Advanced multi-modal characterization strategies, which can co-resolve the organization, dynamics, and biological complexity at the interface across multiple length and time scales are going to be important to future advances. Computational modelling, molecular simulation, machine learning, graph neural networks, and data-driven materials discovery are anticipated to become more important approaches for predicting biomolecule adsorption, binding affinity, enzyme orientation, corona behavior, surface reactivity, and safety profiles.

In the future, the next generation of biomolecule–nanomaterial systems will likely be a product of integrated design systems, which incorporate controlled interface chemistry, predictive modelling, sustainability, and safer-by-design principles. The application of such strategies is very promising for the development of biomolecule–nanomaterial interfaces from their laboratory-scale innovations toward robust, scalable, and application-ready technologies in sensing, catalysis, medicine, environmental remediation and energy systems. “Instead, the future of biomolecule–nanomaterial engineering is in predictive, quantitatively-guided control of dynamic nano–bio interfaces in complex real-world settings”.

Author Contributions

S.S. (Sandhya Savita): writing—original draft preparation, conceptualization, methodology; S.B.: data curation, validation, software; A.M.: visualization, investigation; A.S.: data curation, investigation; P.B.: validation, software; S.K.S.: validation, visualization; S.S. (Suryabhan Singh): data curation, validation; A.K.S.: data curation, investigation; U.P.A.: validation, supervision; writing—reviewing and editing; B.M.: funding, validation, supervision, writing—reviewing and editing; B.L.S.: validation, supervision, writing—reviewing and editing. All authors have read and agreed to the published version of the manuscript.

Funding

This research was funded by ANRF, India [IRG No. ANRF/IRG/2024/001761/CS].

Institutional Review Board Statement

Not applicable.

Informed Consent Statement

Not applicable.

Data Availability Statement

All the data used in the review are already present in public domains and no new data has been generated.

Acknowledgments

U.P.A., B.L.S. and B.M. sincerely grateful to the central library of Guru Ghasidas Vishwavidyalaya for the access of various books and journals. B.M. gratefully acknowledges the financial support provided by ANRF, India (Inclusivity Research Grant No. ANRF/IRG/2024/001761/CS).

Conflicts of Interest

The authors declare no conflict of interest.

Use of AI and AI-Assisted Technologies

Artificial intelligence (AI)-assisted tool (Gemini Pro) was used for the generation and graphical design of the schematic figures (Figures 1–7) presented in this review. All AI-generated figures were developed under the direction of the authors and subsequently reviewed, edited, and scientifically verified by all the authors. The authors take full responsibility for the accuracy, interpretation, originality, and final presentation of all figures in the manuscript.

References

- Rodríguez-Quijada, C.; Sánchez-Purrà, M.; de Puig, H.; et al. Physical properties of biomolecules at the nanomaterial interface. *J. Phys. Chem. B* **2018**, *122*, 2827–2840.
- Bobo, D.; Robinson, K.J.; Islam, J.; et al. Nanoparticle-Based Medicines: A Review of FDA-Approved Materials and Clinical Trials to Date. *Pharm. Res.* **2016**, *33*, 2373–2387.
- Anselmo, A.C.; Mitragotri, S. Nanoparticles in the Clinic. *Bioeng. Transl. Med.* **2016**, *1*, 10–29.
- Bekdemir, A.; Stellacci, F. A Centrifugation-Based Physicochemical Characterization Method for the Interaction Between Proteins and Nanoparticles. *Nat. Commun.* **2016**, *7*, 13121.
- Willner, I.; Willner, B. Biomolecule-based nanomaterials and nanostructures. *Nano Lett.* **2010**, *10*, 3805–3815.
- Willner, I.; Basnar, B.; Willner, B. Nanoparticle–Enzyme Hybrid Systems for Nanobiotechnology Applications. *FEBS J.* **2007**, *274*, 302–309.
- Freeman, R.; Finder, T.; Willner, I. Multiplexed Analysis of Hg²⁺ and Ag⁺ Ions by DNA-Based Sensors. *Angew. Chem. Int. Ed.* **2009**, *48*, 7818–7821.
- Freeman, R.; Gill, R.; Shweky, I.; et al. Biosensing and Probing of Intracellular Metabolic Pathways by NADH-Sensitive Quantum Dots. *Angew. Chem. Int. Ed.* **2009**, *48*, 309–313.
- Zhao, H.; Wang, Y.; Bao, L.; et al. Engineering nano–bio interfaces from nanomaterials to nanomedicines. *Acc. Mater. Res.* **2022**, *3*, 812–829.
- Ren, J.; Andrikopoulos, N.; Velonia, K.; et al. Chemical and Biophysical Signatures of the Protein Corona in Nanomedicine. *J. Am. Chem. Soc.* **2022**, *144*, 9184–9205.
- Liu, J.; Guo, M.; Chen, C. Nano-Bio Interactions: A Major Principle in the Dynamic Biological Processes of Nano-Assemblies. *Adv. Drug Deliv. Rev.* **2022**, *186*, 114318.
- Bondarenko, O.; Mortimer, M.; Kahru, A.; et al. Nanotoxicology and Nanomedicine: The Yin and Yang of Nano-Bio Interactions for the New Decade. *Nano Today* **2021**, *39*, 101184.
- Pulido-Reyes, G.; Leganés, F.; Fernández-Piñas, F.; et al. Bio-nano interface and environment: A critical review. *Environ. Toxicol. Chem.* **2017**, *36*, 3181–3193.
- Kuruvilla, J.; Farinha, A.P.; Bayat, N.; et al. Surface proteomics on nanoparticles: A step to simplify the rapid prototyping of nanoparticles. *Nanoscale Horiz.* **2017**, *2*, 55–64.
- Yue, Y.; Li, X.; Sigg, L.; et al. Interaction of silver nanoparticles with algae and fish cells: A side by side comparison. *J. Nanobiotechnol.* **2017**, *15*, 16.
- Hayashi, Y.; Miclaus, T.; Murugadoss, S.; et al. Female versus male biological identities of nanoparticles determine the interaction with immune cells in fish. *Environ. Sci. Nano* **2017**, *4*, 895–906.
- Yang, Y.-S.S.; Atukorale, P.U.; Moynihan, K.D.; et al. High-throughput quantitation of inorganic nanoparticle biodistribution at the single-cell level using mass cytometry. *Nat. Commun.* **2017**, *8*, 14069.
- Ma, J.; Yang, S.; Yang, Z.; et al. Functional nanomaterials for advanced bioelectrode interfaces: Recent advances in disease detection and metabolic monitoring. *Sensors* **2025**, *25*, 4412.
- Irimes, M.B.; Pusta, A.; Cernat, A.; et al. Multiplexed electrochemical sensing devices for chronic diseases diagnosis and monitoring. *TrAC Trends Anal. Chem.* **2024**, *172*, 117560.
- Han, Q.; Wang, H.; Wang, J. Multi-Mode/Signal Biosensors: Electrochemical Integrated Sensing Techniques. *Adv. Funct. Mater.* **2024**, *34*, 2403122.
- Keles, G.; Sifa Ataman, E.; Taskin, S.B.; et al. Nanostructured Metal Oxide-Based Electrochemical Biosensors in Medical Diagnosis. *Biosensors* **2024**, *14*, 238.
- Fabbri, R.; Scida, A.; Saracino, E.; et al. Graphene oxide electrodes enable electrical stimulation of distinct calcium signalling in brain astrocytes. *Nat. Nanotechnol.* **2024**, *19*, 1344–1353.
- Song, J.; Chen, H.; Sun, Y.; et al. Layered MXene Films via Self-Assembly. *Small* **2024**, *20*, e2406855.

24. Jawad, M.S.; Rahman, W.U.; Khan, M.; et al. Nanomaterial assembly pathways: Comparative insights into self-assembly and directed assembly techniques. *Next Mater.* **2026**, *10*, 101422.
25. Karachevtsev, V.A.; Stepanian, S.G. Biomolecules and their complexes with nanostructures. *Low Temp. Phys.* **2021**, *47*, 179–180. <https://doi.org/10.1063/10.0003517>.
26. Miao, H.; Chen, K.; Yu, H.; et al. DNA-directed assembly of graphene homostructures. *J. Appl. Phys.* **2025**, *138*, 044303.
27. Kuznetsova, V.; Kadar, A.; Gaenko, A.; et al. Graph property relationships for complex chiral nanodendrimers. *ACS Nano* **2025**, *19*, 6095–6106.
28. Bhaskar, S.; Wang, W.; Lee, H.; et al. Photonic crystal grating resonance and interfaces for health diagnostic technologies. *Chem. Rev.* **2025**, *125*, 6435–6540.
29. Yeo, E.L.L.; Cheah, J.U.J.; Neo, D.J.H.; et al. Exploiting the Protein Corona around Gold Nanorods for Low-Dose Combined Photothermal and Photodynamic Therapy. *J. Mater. Chem. B* **2017**, *5*, 254–268.
30. Chan, K.P.; Gao, Y.; Goh, J.X.; et al. Exploiting the Protein Corona from Cell Lysate on DNA Functionalized Gold Nanoparticles for Enhanced mRNA Translation. *ACS Appl. Mater. Interfaces* **2017**, *9*, 10408–10417.
31. Ozboyaci, M.; Kokh, D.B.; Corni, S.; et al. Modeling and Simulation of Protein–Surface Interactions: Achievements and Challenges. *Q. Rev. Biophys.* **2016**, *49*, e4. <https://doi.org/10.1017/S0033583515000256>.
32. Gao, Q.; Liu, S.; Guo, Z.; et al. Nanozyme-biological interfaces: A review on mechanisms and applications from natural biomolecular corona to artificial surface modifications. *ACS Appl. Nano Mater.* **2025**, *8*, 22070–22085.
33. Wu, G.; Li, S.; Luo, L.; et al. Exploring Single-Atom Nanozymes toward Environmental Pollutants: Monitoring and Control. *Nano Micro Lett.* **2025**, *17*, 238.
34. Ma, L.; Liang, Z.; Hou, Y.; et al. Nanozymes and Their Potential Roles in the Origin of Life. *Adv. Mater.* **2025**, *37*, 2412211.
35. Jia, X.; Wang, E.; Wang, J.; et al. Rational Design of Nanozymes for Engineered Cascade Catalytic Cancer Therapy. *Chem. Rev.* **2025**, *125*, 2908–2952.
36. Yang, X.; Hu, Y.; Zhou, H.; et al. Specific Capture, Detection, and Killing of *Enterococcus faecalis* Based on Aptamer-Modified Peroxidase Mimetic Nanozymes. *Chem. Eng. J.* **2025**, *511*, 161848.
37. Wu, Y.; Zhang, Y.; Han, J.; et al. Nanozyme-Based Biomimetic Intelligent Immune Organelles for the Treatment of Bladder-Metastasized Tumors. *Adv. Mater.* **2025**, *37*, e11181.
38. Miller, M.A.; Medina, S. Life at the interface: Engineering bio-nanomaterials through interfacial molecular self-assembly. *Wiley Interdiscip. Rev. Nanomed. Nanobiotechnol.* **2024**, *16*, e1966. <https://doi.org/10.1002/wnan.1966>.
39. Hou, S.; Bai, L.; Lu, D.; et al. Interfacial Colloidal Self-Assembly for Functional Materials. *Acc. Chem. Res.* **2023**, *56*, 740–751.
40. Lawanprasert, A.; Sloand, J.N.; González Vargas, M.; et al. Deciphering the Mechanistic Basis for Perfluoroalkyl-Protein Interactions. *ChemBioChem* **2023**, *24*, e202300159. <https://doi.org/10.1002/cbic.202300159>.
41. Ornithopoulou, E.; Åstrand, C.; Gustafsson, L.; et al. Self-assembly of RGD-functionalized recombinant spider silk protein into microspheres in physiological buffer and in the presence of hyaluronic acid. *ACS Appl. Bio Mater.* **2023**, *6*, 3696–3705.
42. Dang, M.; Yang, C.; Jin, G.; et al. From biomolecular condensates to functional nanomaterials: LLPS-inspired frameworks for nanoscale hydrogels and adaptive materials. *J. Nanobiotechnol.* **2026**, *24*, 280.
43. Pu, J.; Wu, S.; Chen, L. AFM multiparametric characterization of LLPS in plants. *Trends Plant Sci.* **2025**, *30*, 437–438.
44. Arbiol Enguita, A.M.; Zöllner, L.; Tomberg, T.; et al. Chemically Specific Coherent Raman Imaging of Liquid–Liquid Phase Separation and Its Sequelae. *Anal. Chem.* **2025**, *97*, 3242–3252.
45. Qian, J.; Li, X.; Ruan, H.; et al. Design and development of drug delivery nanocarriers based on liquid–liquid phase separation: Improved stability, cell penetration and anti-cancer effect. *Int. J. Biol. Macromol.* **2025**, *307*, 142023.
46. Wen, P.; Huang, H.; Zhang, R.; et al. Coacervate vesicles assembled by liquid–liquid phase separation improve delivery of biopharmaceuticals. *Nat. Chem.* **2025**, *17*, 279–288.
47. Caselli, L.; Conti, L.; De Santis, I.; et al. Small-angle X-ray and neutron scattering applied to lipid-based nanoparticles: Recent advancements across different length scales. *Adv. Colloid Interface Sci.* **2024**, *327*, 103156.
48. Yang, H.; Yao, L.; Wang, Y.; et al. Advancing cell surface modification in mammalian cells with synthetic molecules. *Chem. Sci.* **2023**, *14*, 13325–13345.
49. Zhang, F.L.; Hou, Y.; Zhu, M.H.; et al. Death Pathways of Cancer Cells Modulated by Surface Molecule Density on Gold Nanorods. *Adv. Sci.* **2021**, *8*, 2102666.
50. Tiwari, V.K.; Jaiswal, M.K.; Rajkhowa, S.; et al. Bioorthogonal Click Chemistry: Invention to Applications in Living Systems. In *Click Chemistry*; Springer Nature: Singapore 2024; pp. 175–203.
51. Swierczewski, M.; Bürgi, T. Langmuir and Langmuir–Blodgett Films of Gold and Silver Nanoparticles. *Langmuir* **2023**, *39*, 2135–2151.
52. Pinals, R.L. On Protein Corona Formation: Understanding Nano-Bio Interactions Toward Engineering Optical Nanomaterials. Ph.D. Thesis, in Chemical Engineering, University of California, Berkeley, CA, USA, 2021.

53. Baimanov, D.; Wang, J.; Liu, Y.; et al. Identification of Cell Receptors Responsible for Recognition and Binding of Lipid Nanoparticles. *J. Am. Chem. Soc.* **2025**, *147*, 7604–7616.
54. Balashev, K., Interfacial Interactions of Nanoparticles and Molecular Nanostructures with Model Membrane Systems: Mechanisms, Methods, and Applications. *Membranes* **2026**, *16*, 134. <https://doi.org/10.3390/membranes16040134>.
55. Guo, P.; Wang, Y.; Cui, H.; et al. Nano-bio-interface: Unleashing the potential of noble nanometals. *Small Sci.* **2024**, *4*, 2300227.
56. Azharuddin, M.; Zhu, G.H.; Das, D.; et al. A repertoire of biomedical applications of noble metal nanoparticles. *Chem. Commun.* **2019**, *55*, 6964–6996.
57. Wang, Z.; Tang, C.; Huang, M.; et al. One-Step Synthesis of Peptide–Gold Nanoclusters with Tunable Fluorescence and Enhanced Gene Delivery Efficiency. *Langmuir* **2022**, *38*, 14799–14807.
58. Cao, C.; Yang, N.; Wang, X.; et al. Biomedicine meets nanozyme catalytic chemistry. *Coord. Chem. Rev.* **2023**, *491*, 215245.
59. Chen, W.-Q.; Wu, W.-J.; Yu, Y.-Q.; et al. New Insights on the Size-Dependent Inhibition of Enzymes by Gold Nanoparticles. *Langmuir* **2023**, *39*, 9595–9603.
60. Ramezani Farani, M.; Zandi, A.; Shojaeian, F.; et al. Biomimetic carbon-based nanomaterials: From design strategies to next-generation biosensing and theranostic applications. *Small* **2025**, *21*, e02711.
61. Farani, M.R.; Zare, I.; Mirshafiei, M.; et al. Graphene oxide-engineered chitosan nanoparticles: Synthesis, properties, and antibacterial activity for tissue engineering and regenerative medicine. *Chem. Eng. J.* **2025**, *483*, 160852.
62. Jabbari, F.; Farani, M.R.; Abednejad, A.; et al. Chitosan Nanoparticles in Tissue Engineering and Regenerative Medicine. In *Fundamentals and Biomedical Applications of Chitosan Nanoparticles*; Elsevier: Amsterdam, The Netherlands, 2025; pp. 497–526.
63. Banazadeh, M.; Ilaghi, M.; Abadi, B.; et al. Chitosan Nanoparticles-Hydrogel Composites for Biomedical Applications. In *Fundamentals and Biomedical Applications of Chitosan Nanoparticles*; Elsevier: Amsterdam, The Netherlands, 2025; pp. 633–678.
64. Kang, N.; Hwang, J.; Jeong, D.; et al. Ligand Interrelation Analysis via Graph Theory Predicts Macrophage Response. *Adv. Mater.* **2025**, *37*, 2414356.
65. Yang, B.; Wang, R.; Li, W.; et al. On-Origami Molecular Crowding Control of G-Quadruplex DNAzymes. *Small Methods* **2025**, *9*, 2401401.
66. Gou, Q.; Lu, L.; Lin, S.; et al. Multifunctional Chitosan–Covalent Bonded Multi-Walled Carbon Nanotubes Composite Binder for Enhanced Electrochemical Performances of Lithium–Sulfur Batteries. *Macromol. Rapid Commun.* **2025**, *46*, 2500155.
67. Li, T.; Sun, M.; Zhou, Q.; et al. Endogenous Enzyme-Powered DNA Nanomotor Operating in Living Cells for microRNA Imaging. *Anal. Chem.* **2023**, *95*, 15025–15032.
68. Freeman, R.; Finder, T.; Gill, R.; et al. Probing Protein Kinase (CK2) and Alkaline Phosphatase with CdSe/ZnS Quantum Dots. *Nano Lett.* **2010**, *10*, 2192–2196.
69. Lo, P.K.; Karam, P.; Aldaye, F.A.; et al. Loading and selective release of cargo in DNA nanotubes with longitudinal variation. *Nat. Chem.* **2010**, *2*, 319–328.
70. Gu, H.; Chao, J.; Xiao, S.-J.; et al. A proximity-based programmable DNA nanoscale assembly line. *Nature* **2010**, *465*, 202–205.
71. Yehezkeli, O.; Yan, Y.-M.; Baravik, I.; et al. Integrated oligoaniline-cross-linked composites of Au nanoparticles/glucose oxidase electrodes: A generic paradigm for electrically contacted enzyme systems. *Chem. Eur. J.* **2009**, *15*, 2674–2679.
72. Freeman, R.; Li, Y.; Tel-Vered, R.; et al. Self-assembly of supramolecular aptamer structures for optical or electrochemical sensing. *Analyst* **2009**, *134*, 653–656.
73. Andersen, E.S.; Dong, M.; Nielsen, M.M.; et al. Self-Assembly of a Nanoscale DNA Box with a Controllable Lid. *Nature* **2009**, *459*, 73–76.
74. Sharma, J.; Chhabra, R.; Cheng, A.; et al. Control of Self-Assembly of DNA Tubules Through Sequence Design. *Science* **2009**, *323*, 112–116.
75. Yang, Q.; Ren, W.J.; Yu, X.H.; et al. Arachin amyloid-like fibrils and chitosan composite coatings with antimicrobial and antioxidant activities for effective preservation of perishable fruits. *Int. J. Biol. Macromol.* **2025**, *308*, 142704.
76. Feng, N.; Zhang, J.; Tian, J.; et al. Preserving fruit freshness with amyloid-like protein coatings. *Nat. Commun.* **2025**, *16*, 5060.
77. Liu, G.; Liu, R.; Fu, C.; et al. Protein/polysaccharide composite nanocoating based on amyloid-like aggregation for fresh-cut fruits preservation. *Chem. Eng. J.* **2025**, *519*, 165048.
78. Marshall, K.E.; Mengham, K.; Spink, M.C.; et al. Correlative cryo-soft X-ray tomography and cryo-structured illumination microscopy reveal changes to lysosomes in amyloid- β -treated neurons. *Structure* **2024**, *32*, 585–593.
79. Ding, L.; Bing, L.; Peil, A.; et al. DNA-directed assembly of photonic nanomaterials for diagnostic and therapeutic applications. *Adv. Mater.* **2025**, *37*, 2500086. <https://doi.org/10.1002/adma.202500086>.

80. Zhang, T.-K.; Yi, Z.-Q.; Huang, Y.-Q.; et al. Natural biomolecules for cell-interface engineering. *Chem. Sci.* **2025**, *16*, 3019–3044.
81. Zhang, T.-K.; Geng, W.; Huang, Y.-Q.; et al. Specific recognition to create nanofunctionalized cells for precise functions. *Coord. Chem. Rev.* **2024**, *498*, 215471.
82. Gao, M.; Wang, H.; Zhang, Y.; et al. Unveiling Trends: Nanoscale Materials Shaping Emerging Biomedical Applications. *ACS Nano* **2024**, *18*, 16325–16342.
83. Yang, H.; Xiong, Z.; Heng, X.; et al. Click-Chemistry-Mediated Cell Membrane Glycopolymer Engineering to Potentiate Dendritic Cell Vaccines. *Angew. Chem. Int. Ed.* **2024**, *63*, e202315782.
84. Niu, Y.; Xue, D.; Dai, X.; et al. Sustainable power generation from sewage with engineered microorganisms as electrocatalysts. *Nat. Sustain.* **2024**, *7*, 1182–1189.
85. Zhou, W.; Zhang, W.; Geng, W.; et al. External Electrons Directly Stimulate Escherichia coli for Enhancing Biological Hydrogen Production. *ACS Nano* **2024**, *18*, 10840–10849.
86. Li, Z.; Xue, L.; Yang, J.; et al. Metal–Organic Framework-Based Nanoplatfoms for Biomedical Applications. *Adv. Sci.* **2024**, *11*, 2305126.
87. Wei, X.; Wang, Y.; Liu, Y.; et al. Biomimetic design strategies for biomedical applications. *Matter* **2024**, *7*, 826–854.
88. Ma, B.; Bianco, A. Regulation of biological processes by intrinsically chiral engineered materials. *Nat. Rev. Mater.* **2023**, *8*, 403–413.
89. Li, J.; Chen, C.; Xia, T. Understanding Nanomaterial–Liver Interactions to Facilitate the Development of Safer Nanoapplications. *Adv. Mater.* **2022**, *34*, 2106456.
90. Liu, J.M.; Wang, A.Z.; Liu, S.H.; et al. A Titanium Nitride Nanozyme for pH-Responsive and Irradiation-Enhanced Cascade Catalytic Tumor Therapy. *Angew. Chem. Int. Ed.* **2021**, *60*, 25328–25338.
91. Ni, B.; González-Rubio, G.; Van Gordon, K.; et al. Seed-Mediated Growth and Advanced Characterization of Chiral Gold Nanorods. *Adv. Mater.* **2024**, *36*, 2412473.
92. Li, B.; Yu, S.; Feng, R.; et al. Dual-Mode Gold Nanocluster-Based Nanoprobe Platform for Two-Photon Fluorescence Imaging and Fluorescence Lifetime Imaging of Intracellular Endogenous miRNA. *Anal. Chem.* **2023**, *95*, 14925–14933.
93. Habeeb, H.; Vengateswaran, H.T.; Tripathi, A.K.; et al. Enhancing biomedical imaging: the role of nanoparticle-based contrast agents. *Biomed. Microdevices* **2024**, *26*, 42.
94. Li, H.; Zhang, Y.; Ding, J.; et al. Synthesis of Carbon Quantum Dots for Application of Alleviating Amyloid- β Mediated Neurotoxicity. *Colloids Surf. B* **2022**, *212*, 112373.
95. Huang, H.; Liu, R.; Yang, J.; et al. Gold Nanoparticles: Construction for Drug Delivery and Application in Cancer Immunotherapy. *Pharmaceutics* **2023**, *15*, 1868.
96. Li, Q.; Li, Y.; Xu, P.; et al. One-Step Fabrication Bioinspired Flexible Hierarchical Micro–Nano Structures with Different Morphologies. *ACS Appl. Mater. Interfaces* **2023**, *15*, 43016–43025.
97. Kumar, N.; Samal, P.P.; Mahapatra, A.; et al. Deciphering pressure induced nanoarchitectonics in a monolayer of heterocoronene-based discotics at air–water and air–solid interfaces. *Soft Matter* **2023**, *19*, 1513–1522.
98. Sánchez-Purrà, M.; Carré-Camps, M.; de Puig, H.; et al. SERS-Based Sandwich Immunoassays for Multiplexed Detection of Zika and Dengue Viral Biomarkers. *ACS Infect. Dis.* **2017**, *3*, 767–776.
99. Wilhelm, S.; Tavares, A.J.; Dai, Q.; et al. Analysis of nanoparticle delivery to tumours. *Nat. Rev. Mater.* **2016**, *1*, 16014.
100. Liu, C.; Liang, L.; Yu, Y.; et al. Multiple Recognition-Powered Abiotic Bimetallic Oxide Nanozyme-Based Colorimetric Platform for the Selective Detection of Allergen β -Lactoglobulin. *Food Chem.* **2025**, *468*, 142400.
101. Bhaskar, S.; Liu, L.; Liu, W.; et al. Photonic crystal band edge coupled enhanced fluorescence from magneto-plasmonic cryosoret nano-assemblies for ultra-sensitive detection. *APL Mater.* **2025**, *13*, 041103.
102. Bhaskar, S.; Liu, L.; Liu, W.; et al. Photonic-crystal-enhanced fluorescence: Template-free gold cryosoret nanoassembly steering, dequenching, and augmenting the quenched emission from radiating dipoles. *MRS Bull.* **2025**, *50*, 585–598.
103. Bhaskar, S.; Liu, W.; Shepherd, S.; et al. Integrated metallo-dielectric 3D hotspots from high refractive index nanodiamond-plasmonic gold cryosoret nano-assemblies for photonic crystal enhanced fluorescence. *Adv. Funct. Mater.* **2025**, *36*, e12295.
104. Jia, B.; Yao, Y.; Xu, Y.; et al. Multi-Functional Copper-Based Nanoenzyme System with Antioxidant and Ferroptosis Inhibition for Traumatic Brain Injury. *Chem. Eng. J.* **2025**, *515*, 163592.
105. Kim, K.; Min, S.; Thangam, R.; et al. Dynamic hierarchical ligand anisotropy for competing macrophage regulation in vivo. *Bioact. Mater.* **2025**, *47*, 121–135.
106. Miller, M.A.; Medina, S. Synthetic Colonic Mucus Enables the Development of Modular Microbiome Organoids. *Adv. Funct. Mater.* **2024**, *34*, 2402514.
107. Kim, I.; Elliott, J.C.; Lawanprasert, A.; et al. Real-Time, In Situ Imaging of Macrophages via Phase-Change Peptide Nanoemulsions. *Small* **2023**, *19*, 2301673.

108. Tawakey, S.H.; Mansour, M.; Soltan, A.; et al. Early detection of hypo/hyperglycemia using a microneedle electrode array-based biosensor for glucose ultrasensitive monitoring in interstitial fluid. *Lab Chip* **2024**, *24*, 3958–3972.
109. Wang, X.; Xu, M.; Yang, H.; et al. Ultraflexible Neural Electrodes Enabled Synchronized Long-Term Dopamine Detection and Wideband Chronic Recording Deep in Brain. *ACS Nano* **2024**, *18*, 34272–34287.
110. Du, X.; Yang, L.; Shi, X.; et al. Ultrathin Bioelectrode Array with Improved Electrochemical Performance for Electrophysiological Sensing and Modulation. *ACS Nano* **2024**, *18*, 34971–34985.
111. Kirtane, A.R.; Bi, J.; Rajesh, N.U.; et al. Radioprotection of healthy tissue via nanoparticle-delivered mRNA encoding for a damage-suppressor protein found in tardigrades. *Nat. Biomed. Eng.* **2025**, *9*, 1240–1253.
112. Garg, T.; Arora, S.; Pahwa, R. Cellulose and its derivatives: Structure, modification, and application in controlled drug delivery. *Future J. Pharm. Sci.* **2025**, *11*, 76.
113. Donthi, M.R.; Munnangi, S.R.; Krishna, K.V.; et al. Nanoemulgel: A Novel Nano Carrier as a Tool for Topical Drug Delivery. *Pharmaceutics* **2023**, *15*, 164.
114. Zhao, H.; Xu, J.B.; Wang, Y.Q.; et al. A Photosensitizer Discretely Loaded Nanoaggregate with Robust Photodynamic Effect for Local Treatment Triggers Systemic Antitumor Responses. *ACS Nano* **2022**, *16*, 3070–3080.
115. Zhao, H.; Xu, J.B.; Feng, C.; et al. Tailoring Aggregation Extent of Photosensitizers to Boost Phototherapy Potency for Eliciting Systemic Antitumor Immunity. *Adv. Mater.* **2022**, *34*, 2106390.
116. Li, Y.; Feng, H.; Yang, H.; et al. A Self-Priming Pyroptosis-Inducing Agent for Activating Anticancer Immunity. *Adv. Healthcare Mater.* **2025**, *14*, 2500610.
117. Ma, J.; Wu, Y.; Zou, H.; et al. Acid-Responsive Aggregation of Gold Nanoparticles for the Photothermal Treatment of Bacterial Infections. *ACS Infect. Dis.* **2023**, *9*, 2538–2547.
118. de Puig, H.; Carré-Camps, M.; Bosch, I. Effect of the Protein Corona on Antibody–Antigen Binding in Nanoparticle Sandwich Immunoassays. *Bioconjugate Chem.* **2017**, *28*, 230–238.
119. Picchiatti, S.; Bernini, C.; Stocchi, V.; et al. Engineered nanoparticles of titanium dioxide (TiO₂): Uptake and biological effects in a sea bass cell line. *Fish Shellfish Immunol.* **2017**, *63*, 53–67.
120. Shen, Z.; Liang, Q.; Chang, Q.; et al. Topological Hydrogels for Long-Term Brain Signal Monitoring, Neuromodulation, and Stroke Treatment. *Adv. Mater.* **2024**, *36*, e2310365.
121. Fu, F.; Liu, C.; Jiang, Z.; et al. Polymeric silk fibroin hydrogel as a conductive and multifunctional adhesive for durable skin and epidermal electronics. *Smart Med.* **2024**, *3*, e20240027.
122. Mitrano, D.M.; Nowack, B. The need for a life-cycle-based aging paradigm for nanomaterials: Importance of real-world test systems to identify realistic particle transformations. *Nanotechnology* **2017**, *28*, 072001.
123. Berry, T.D.; Filley, T.R.; Clavijo, A.P.; et al. Degradation and microbial uptake of C60 fullerols in contrasting agricultural soils. *Environ. Sci. Technol.* **2017**, *51*, 1387–1394.
124. Martín-de-Lucía, I.; Campos-Mañas, M.C.; Agüera, A.; et al. Reverse Trojan-horse effect decreased wastewater toxicity in the presence of inorganic nanoparticles. *Environ. Sci. Nano* **2017**, *4*, 1273–1282.
125. Elfiky, A.A.; Ibrahim, I.M.; Elghareib, A.M.; et al. Simulation of gold nanoparticle movement through normal and cancer cell membranes. *Comput. Biol. Med.* **2023**, *164*, 107363.