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Antimicrobial nanomaterials at the nanoscale: design principles, mechanisms, and global challenges

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The rapid escalation of antimicrobial resistance (AMR) represents a major global health challenge, undermining the clinical efficacy of conventional antibiotics and driving the demand for alternative therapeutic strategies. Advances in nanoscience have positioned nanoscale materials as a powerful platform for the development of next generation antimicrobial systems, owing to their precisely tunable physicochemical features and ability to operate through multiple resistance evasive modes of action. This review provides a comprehensive overview of recent progress in antimicrobial nanomaterials, encompassing metallic and metal oxide nanoparticles, polymeric and carbon based nanostructures, as well as multifunctional hybrid architectures. Key antimicrobial mechanisms, including controlled ion release, reactive oxygen species generation, membrane perturbation, and synergistic chemical and physical interactions at the nano bio interface, are critically discussed. Particular attention is given to the growing class of stimuli responsive and smart nanomaterials that enable spatiotemporal activation, enhanced targeting, and improved therapeutic precision. The integration of antimicrobial functionality with anti-inflammatory, regenerative, and diagnostic capabilities highlights the versatility of nanoscale systems for biomedical applications such as wound management, implant surface engineering, biofilm mitigation, and targeted drug delivery. Despite significant advances, challenges related to cytocompatibility, biodegradability, scalable manufacturing, and regulatory harmonization remain. Finally, emerging directions including nanozymes, metal organic frameworks, artificial intelligence assisted material design, and antimicrobial 3D printed nanocomposites are discussed, underscoring the pivotal role of nanoscale innovation in addressing AMR and advancing future antimicrobial technologies.

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1. Introduction

Global assessments indicate that AMR is no longer a prospective or emerging concern but has become a major contributor to morbidity and mortality worldwide. Substantial evidence demonstrates a continuous rise in the prevalence of drug-resistant pathogens across both community and healthcare settings. Resistant bacterial infections, including those caused by *Escherichia coli*, *Klebsiella pneumoniae*, and methicillin resistant *Staphylococcus aureus* (MRSA), are now widespread, significantly compromising the effectiveness of first line antimicrobial therapies and leading to increased case fatality rates. Systematic investigations have documented the global dissemination of AMR, revealing the presence of drug-resistant pathogens in critical infectious syndromes across diverse geographical regions, encompassing low, middle, and high income countries alike. Extensive epidemiological evaluations indicate that resistant infections are a persistent and rapidly growing worldwide health issue.^{1,2}

AMR can develop through several mechanisms, including the prevention of drug penetration into the cell, alterations in antibiotic targets, inactivation of enzymes in antibiotics, and active efflux of antibiotics from the cell. Also, biofilm formation provides a protective environment for microbes that shield them from antibiotics, which is a further complicating treatment.³ Fig. 1 shows the mechanism of AMR of microbes.

The spread of antimicrobial resistance has had a direct impact on the increased incidence of treatment failure and undermined clinical outcomes. Previous studies on the comparison of resistant and susceptible infections show that patients with resistant infections have longer hospital stays, more complicated care, and higher mortality rates. Systematic and meta-analytic data indicate that resistant infections are always linked with worse clinical trajectories, which can be more resource-intensive and second or third-line treatments, which are less available or more toxic. The clinical burden of AMR consequently exerts further strain on already over-stretched healthcare services, especially in areas that already have limited capacity to handle advanced infectious disease treatment.^{4,5}

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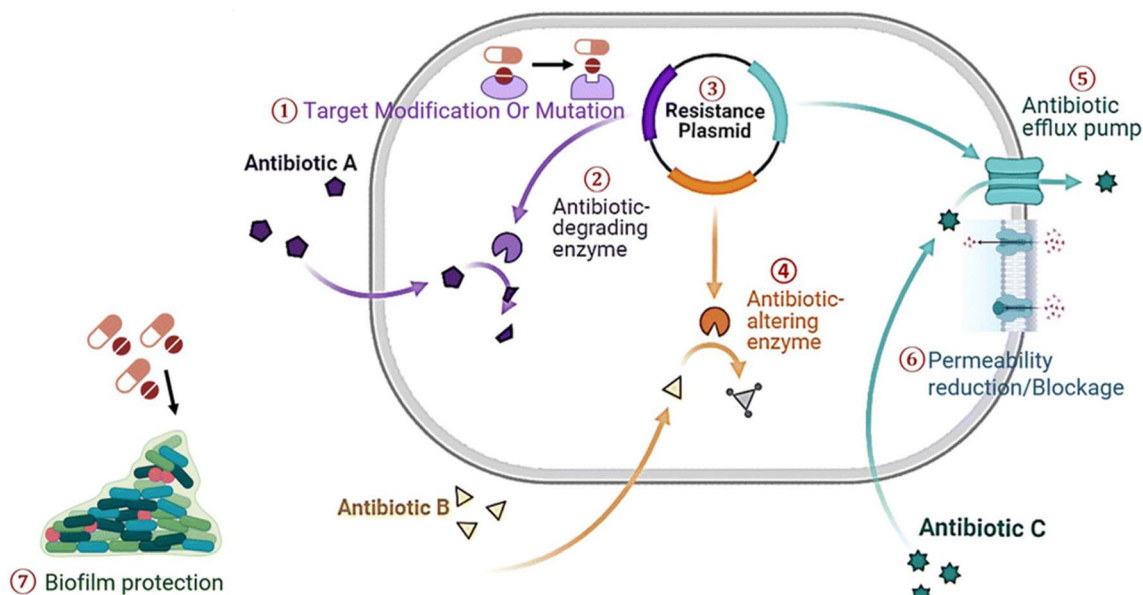


Fig. 1 Mechanism of AMR of microbes, including (1) target modification or mutation reducing antibiotic binding, (2) antibiotic-degrading enzymes, (3) resistance plasmids enabling gene transfer, (4) antibiotic-modifying enzymes, (5) active efflux pumps expelling antibiotics, (6) reduced membrane permeability limiting drug entry, and (7) biofilm formation that protects bacterial communities from antibiotic penetration. The figure is obtained from ref. 3 under a Creative Common (CC BY) License.

The socio-economic consequences of AMR are profound and complex. In a health systems perspective, resistant infections cause significantly greater direct costs because of the extended treatment, longer hospital stay, and greater use of costly or advanced therapeutics. Economic studies have revealed that the total cost of antimicrobial resistance involves high labour productivity losses, decreased workforce, and high out-of-pocket costs to patients and health insurers. Economic models around the world estimate that uncontrolled AMR may cause a significant decline in gross domestic products (GDP) and cause chronic financial strain on the economies of countries and households at risk, especially in low and middle-income countries (LMICs) where most AMR-related morbidity and mortality are experienced. The wider socio-economic effect is increased health inequities and possible retrogression of the gains in the public health, which underscores the overlapping of AMR with social determinants of health and economic development.^{2,5}

The use of conventional antibiotics is severely limited and is threatening its further use in clinical practice: first, the efficacy of the current antibiotics is decreasing as resistant strains of bacteria are becoming more common in the world, decreasing the usefulness of even long established drugs and causing higher rates of treatment failure; surveillance data and previous studies have reported significant increases in resistance among common bacterial pathogens, with resistance now being observed against multiple first and second line agents, limiting the usefulness and reducing the reliability of the traditional antibiotic regimens.⁶ Second, the pipeline in antibiotic discovery is slow and lacks sufficient innovation with only a small number of truly new antibacterial agents undergoing clinical development and most new candidates being

derivatives of existing classes and not an entirely novel chemical scaffold that can be used against resistant pathogens; pipeline evaluations of the world show a low number of innovative antibacterials and diagnostic agents, and this is due to scientific, economic, and regulatory obstacles that have limited investment and slowed the translation of research into approved therapeutics.⁷ Third, microbial populations can quickly evade even newly introduced or in development antibiotics, as experimental studies have shown that priority Gram negative “ESKAPE” pathogens can develop resistance to antibiotics including those recently introduced to the market or under development in short exposure times and by selection of already existing genetic variants. It means that microbial evasion continues to be a challenge to conventional drug paradigms.⁸ These interrelated constraints underscore the fact that the use of traditional antibiotics is no longer adequate to control infectious disease and why there is an immediate necessity to develop alternative therapeutic approaches in addition to long-term investment in antibiotic research and development.

Nanotechnology has emerged as a powerful strategy to address AMR by enabling the design of nanoscale materials that overcome the inherent limitations of conventional antibiotics and traditional drug delivery systems. Recent advances demonstrate that a wide range of nanomaterials, including metallic nanoparticles, polymeric nanostructures, carbon-based materials and hybrid nanosystems, possess unique physicochemical characteristics such as nanoscale dimensions, high surface-to-volume ratios and tunable surface chemistries. These features enable interactions with pathogens and the biological milieu that are unattainable using bulk antimicrobial agents alone. Consequently, antimicrobial nanomaterials have



been extensively investigated for their ability to enhance drug stability, improve targeted delivery to infection sites and function as standalone or synergistic antimicrobial agents. Such approaches offer effective solutions to critical challenges including poor penetration into biofilms and intracellular compartments, limited therapeutic stability and the inherently static nature of conventional antimicrobial formulations.^{9,10}

For examples, previous works have outlined how nanomaterials can be used to improve the efficacy of already existing antimicrobial treatments by improving the delivery of drugs, decreasing required dosage, and possibly avoiding certain conventional resistance mechanisms, including those related to efflux pumps and biofilm barriers, by allowing localized, targeted, and controlled therapeutic effect.¹¹ Likewise, previous studies outline a broad range of engineered nanomaterials (*e.g.*, polymeric nanoparticles, lipid based carriers, metal and metal ion nanoparticles, carbon dots and dendrimers) that are under investigation to be used to enhance antimicrobial therapy in human, veterinary and environmental settings.¹² These works point to the possibility of nanotechnology to transform antimicrobial approaches through the principles of multifunctionality and cross disciplinary design to enhance effectiveness against multidrug resistant organisms.¹⁰

Besides therapeutic delivery, nanoparticle mediated platforms have also been studied in the treatment of recalcitrant infections and biofilm associated pathogens which are hard to treat with conventional antibiotics because of physical and physiological barriers. Surveys of nanoparticle based antimicrobial strategies highlight the importance of nanoparticle based approaches in increasing penetration of biofilms and hard to reach cellular niches, allowing new formulations to be developed that combine antimicrobial agents with carrier systems that are designed to be more stable and bi-odistributed.¹³ Collectively, this review demonstrates that nanotechnology has significant potential as a next generation toolset in the fight against AMR, which can be used to enhance current therapies and design novel therapeutic approaches, although current efforts are dealing with issues of safety, scalability, and clinical translation.^{9,10}

2. Antimicrobial nanomaterials

2.1 Nanomaterial types

2.1.1 Metals. Metal nanomaterials consist of elemental metallic nanoparticles which are widely researched in the field of antimicrobial use. Silver, gold, copper and iron in nanoscale are common examples. Silver nanoparticles specifically have been one of the most studied metal antimicrobials and have been added to a variety of products including medical device coatings, antimicrobial textiles and wound dressings. Nanoparticles of gold and copper have also been created to be used in biological and healthcare applications, and studies have been done on the incorporation of nanoparticles into sensors, medical devices, and therapeutic materials. Metal nanomaterials are valued due to their tunable physicochemical characteristics at the nanoscale, simple synthesis with size and

shape control, and their ability to be further functionalized to meet a particular application.^{13,14}

2.1.2 Metal oxides. Metal oxide nanomaterials are metal-oxygen atom composites that are formed into nanoscale particles that are not similar to their bulk counterparts in terms of surface area and structural properties. Examples of these are oxides of zinc (ZnO), titanium (TiO₂), copper (CuO) and iron (*e.g.*, Fe₃O₄). The nanomaterials have been of interest to be incorporated in coating, composites and biomedical formulations where antimicrobial properties are required. They are robust materials to use in their applications in healthcare, environmental remediation materials and consumer products because of their stability, the range of available metal oxide compositions, and their ability to be manufactured with controlled size distributions.¹⁵

2.1.3. Polymeric nanomaterials. Polymeric nanomaterials are organic, nanoscale structures built from polymers, which may be natural or synthetic. Biocompatible polymers like chitosan and biodegradable polymers like poly(lactic-co-glycolic acid) (PLGA) can be made into nanoparticles, nanofibers or nanocomposites. They are commonly discussed in biomedical applications where their biocompatibility and structural flexibility allow them to be used as drug delivery vehicles, wound care systems and implantable antimicrobial systems. The polymer matrix itself may also be used as a nanoscale scaffold or carrier of active agents or functional components, and this offers flexibility in design and application.¹⁴ Functional antimicrobial polymers, including cationic polymers, amphiphilic macromolecules, and polymeric hydrogels, have demonstrated significant potential for preventing microbial colonization and biofilm formation while maintaining favorable biocompatibility profiles.

2.1.4 Carbon-based nanomaterials. Carbon-based nanomaterials represent a wide range of structures that are mainly made of carbon atoms in specific nanoscale structures. Some of the most common ones are graphene and its derivatives (*e.g.*, graphene oxide), carbon nanotubes (single-walled and multi-walled), fullerenes, nanodiamonds and carbon quantum dots. These carbon nanostructures are characterized by large surface area, unique mechanical and electrical characteristics, and a large variety of potential functionalizations. Due to these properties, carbon-based materials are used in composites, coatings, and other systems where antimicrobial properties are needed in the design, such as biomedical interfaces and high-performance filtration.¹⁶

2.1.5 Hybrid materials. Hybrid nanomaterials are composites, which consist of two or more different material classes to achieve superior or multifunctional characteristics. Hybrids are commonly used in the antimicrobial field where metallic or metal oxide materials are combined with polymers or carbon nanostructures to make systems that leverage on the advantages of each constituent material type. Such examples are metal nanoparticles incorporated into polymer scaffolds, and metal-functionalized carbon nanotube or graphene scaffolds.¹⁶ Hybrid systems can also include metal-organic frameworks (MOFs), where metal nodes are connected with organic ligands to form porous structures. These hybrid strategies



enable the investigator to customize the material properties including stability, biocompatibility, and incorporation into application-specific formats including coatings to drug delivery vehicles.^{16,17} The recent developments in antimicrobial biomaterials have been more directed towards multifunctional surface designs that incorporate antimicrobial activity with antifouling properties to inhibit microbial colonization and biofilm formation on biomedical devices. As an example, a previous study discusses the creation of multifunctional antimicrobial surfaces that combine bactericidal agents with antiadhesive or antifouling agents. Such systems frequently include nitric oxide (NO) releasing substances along with hydrophilic polymers, zwitterionic coating, or topographical alterations of surfaces to prevent bacterial adhesion as well as to remove adhesive microorganisms. These multifunctional strategies have been shown to perform better than traditional single function antibacterial materials and are now being considered in biomedical applications such as implant coating, catheters and other medical devices where biofilm formation is a major problem.¹⁸

2.2 Functional categories

2.2.1 Active nanomaterials. Active antimicrobial nanomaterials are nanomaterials that are designed to have a specific antimicrobial activity by containing or attaching active agents or functions. Nanomaterials in this category, can be specifically designed to contain or deliver antimicrobial ingredients in the form of metal nanoparticles, functional polymer coatings or other incorporated bioactive entities, which are designed to react with microbial contaminants upon contact.¹⁹ Active nanomaterials are commonly incorporated into coatings, composites, or delivery vehicles in which their desired activity is directly involved in the reduction of microbial burden or contamination. A wide variety of active nanoscale materials have been used in antimicrobial applications, with their physicochemical properties being tailored and their functions as active agents in therapeutic, environmental, and device-related applications.²⁰

2.2.2 Passive nanomaterials. Passive antimicrobial nanomaterials are designed in such a way that they prevent microbial adhesion, colonization or persistence without the release of active antimicrobial agents. Rather, these nanomaterials are characterized by surface properties, topographies or interfacial chemistry that discourage microbial settlement or growth on contact.²¹ Examples are nanostructured surfaces that foul or resist bacterial attachment because of their nanoscale engineered properties, *e.g.* surface patterns or hydrophilic/hydrophobic balances that do not promote stable microbial adherence. Passive designs are also commonly discussed in the previously reported literature on antimicrobial coatings, in which the focus is on physical and chemical interface design, rather than on the incorporation of soluble biocidal agents.²²

2.2.3 Stimuli-responsive nanomaterials. Stimuli-responsive nanomaterials are designed to alter their structural or functional state in response to environmental conditions allowing them to activate or be activated precisely at their site of action. In biomedical and antimicrobial contexts, smart

nanomaterials are engineered to react to endogenous stimuli including local pH variations, overexpressed enzymes and altered redox conditions which are typical of sites of infection or pathological microenvironment.²³ As an example, microenvironmental conditions such as slightly acidic pH and elevated concentrations of certain enzymes and metabolic byproducts are frequently observed in infection microenvironments and can be used to induce changes in nanomaterial structure, surface characteristics, or cargo release only where necessary.²⁴ In the case of endogenous stimuli, pH-responsive systems are one of the most commonly studied due to the slightly acidic microenvironment (pH ~ 5–6.5) of infected tissues and biofilms in comparison with normal physiological pH (~7.4).²³ To exploit this difference, scientists add pH-sensitive linkers like hydrazone, acetal, ketal, imine, or *cis*-aconityl bonds to the nanoparticle or polymeric carrier. These linkers are stable at physiological pH, but cleave or rearrange their structure in acidic environments, allowing antimicrobial payloads to be released at specific sites.^{25,26} Conversely, enzyme-responsive nanomaterials utilize peptide sequences or polymer backbones that can be cleaved selectively by enzymes that are overexpressed at sites of infection, including collagenases, hyaluronidases or proteases. The antimicrobial activity can be activated by enzymatic cleavage of these sequences to disassemble nanoparticles or reveal active functional groups.²⁷

Importantly, different chemical strategies involve varying stability, specificity and release kinetics trade-offs. As an example, pH-labile linkers are typically relatively predictable in release profiles and easy to synthesize chemically but can also be unintentionally activated in other acidic body locations. Enzyme-responsive systems can provide increased biological specificity since they depend on the presence of specific enzymes, but peptide linkers can be cleaved by non-specific proteases and thus activated prematurely.²⁸ This means that it needs to be carefully designed to ensure that it is structurally stable during circulation and fast activating at the target site.

The other important design factor is to quantitatively match the response threshold of the nanomaterial to the pathological microenvironment. As an example, the pK_a of pH-responsive polymers ionizable groups should preferably match the pH range of the targeted infection environment to be selectively activated. Normal tissues have a pH of about 7.4, and tumor tissues tend to have pH of about 6.5–7.0 and intracellular endosomal compartments may have a pH of 4.5–6.5.²⁹ Similarly, infected wounds can exhibit pH ranges of about 5.4 to 7.4 based on the level of infection.³⁰ Materials with an activation threshold that matches these microenvironmental conditions are designed to enhance the accuracy of targeting and reduce off-target activation. Smart nanomaterials can be used to target these localized biological signals, thereby enhancing targeting and minimizing off-target effects by incorporating chemical groups or structural motifs that are responsive to these localized biological signals.³¹

It is also possible to design stimuli-responsive nanomaterials that react to exogenous physical stimuli to increase their capabilities beyond intrinsic environmental signals. Nanomaterial features can be triggered by exogenous stimuli,



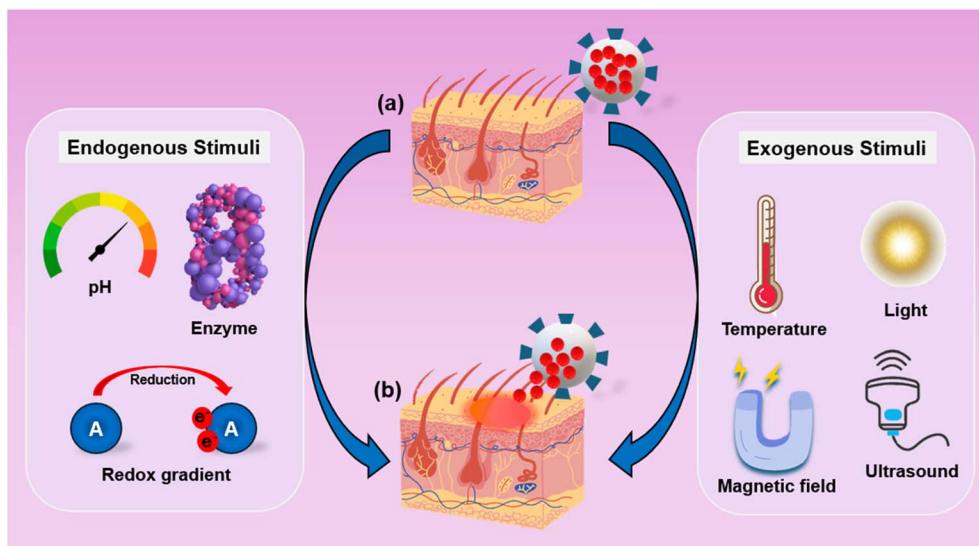


Fig. 2 (a) Under normal tissues, the nanoparticles are in an inactive state, as there are no activating endogenous or exogenous stimuli, preventing premature release of therapeutic contents. (b) At the wound site, in the presence of signals of the pathological microenvironment, together with endogenous and exogenous stimuli, activate nanoparticle and produce localized delivery of the therapeutic cargo, which highlights the concept of stimulus-regulated delivery. (Figure created by the authors; some graphical components were sourced from Canva and adapted).

e.g. light, temperature variations, magnetic fields or ultrasound, which can be delivered intentionally to achieve a desired response, *e.g.* conformational changes, controlled release of therapeutic contents or enhanced interaction with microbial targets at the desired time and location.^{23,32} Light-responsive systems utilize photo-sensitive molecules or photothermal nanomaterials that convert light energy into heat or reactive species, enabling localized antibacterial treatment upon irradiation. Magnetic-responsive nanomaterials, often based on iron oxide nanoparticles, can be manipulated using external magnetic fields for targeted delivery or controlled heating. Such externally triggered reactions enable on-demand control of nanomaterial behaviour, providing clinicians or researchers with the capability to exert spatiotemporal control of treatment with great precision. Smart nanomaterials can be used to enhance the results of antimicrobial therapy and other biomedical interventions by integrating endogenous and exogenous stimuli responsiveness into one design, which provides multifunctional adaptability.^{32,33}

Other recent developments in antimicrobial materials are stimuli-responsive polymeric biohybrids based on amino acids and biomacromolecules. As an example, Mohammad *et al.*³⁴ have created an L-histidine-based amphiphilic triblock copolymer biohybrid (PHisMAM-*b*-PB-*b*-PHisMAM) that is pH and temperature responsive. The material can change between zwitterionic, cationic and anionic states based on the environmental conditions, which allows adaptive antifouling behavior and controllable interactions with biological systems. These histidine-based polymeric structures illustrate how biomolecule-based materials may be designed to form smart antimicrobial and antifouling surfaces with physicochemical properties that can be controlled to be used in biomedical applications.

Fig. 2 depicts the endogenous and exogenous stimuli which cause to controlled release of therapeutic contents at the wound site.

2.2.4 Multifunctional and hybrid nanomaterials. Recent studies in antimicrobial nanotechnology are moving towards the development of multifunctional and hybrid nanomaterials beyond microbial inhibitory effects to aid in anti-inflammatory and tissue-regenerative effects in therapeutic applications. As an example, the recent electrospun nanohybrid membranes incorporate trimetallic nanoparticles with bioactive curcuminoids, which combine antimicrobial activity with high anti-inflammatory effects, showing how hybrid nanomaterials can be used to treat both infection control and inflammatory reactions in the same platform. The systems can be fabricated typically by biodegradable polymers that mimic the natural extracellular matrices not only in the inhibition of infections, but also the increased tissue compatibility and healing capacity. These multifunctional nanomaterials are especially applicable to biomedical applications in wound-related contexts, where inflammation reduction and cell adhesion and growth in the presence of antimicrobial activity can be of significant benefit to the overall therapeutic outcome.³⁵

Besides anti-inflammatory incorporation, multifunctional nanocomposite hydrogels and other hybrid systems have been developed to facilitate regenerative events and at the same time offer antimicrobial protection. An example is the use of advanced multifunctional hydrogels with antimicrobial and osteogenic agents that have been demonstrated to stimulate bone regeneration in defect repair models, which deal with microbial contamination and tissue regeneration in one construct. These hybrid scaffolds are commonly used together with bioactive molecules like growth factors or biomineral components and antimicrobial nanoparticles and allow the synergistic effect of promoting regenerative signaling and



inhibiting infection. The combination of antimicrobial activity and regenerative support is an example of how hybrid nanomaterials can be used to bridge the gap between infection control and tissue engineering and expand their application to clinical applications where infection and tissue damage are simultaneous.³⁶

In addition to therapeutic and regenerative applications, multifunctional nanomaterials are also considered to have integrated diagnostic applications, where real-time infection status or therapeutic response can be monitored *in vivo*. Recent nanomedicine literature has focused on innovative micro and nanoarchitectures that are designed to deliver imaging agents, biosensors, or responsive markers reporting on biological conditions with co-located antimicrobial activity. Such a theranostic combination, therapeutic antimicrobial activity with diagnostic outputs, can be used to support individualized therapy plans, whereby the clinician can customize interventions to local biological responses on the nanomaterial itself. These dual-function systems have a great potential to complement the normal care pathways with actionable information regarding the progress or resolution of the infection in parallel with direct antimicrobial therapy.³⁷

3 Mechanisms of action in antimicrobial nanomaterials

3.1 Chemical pathways

3.1.1 Ion release. In ion releasing antimicrobial nanomaterials, the process starts at the surface of the nanoparticle where the interaction with water, dissolved oxygen or biological fluids triggers oxidative dissolution or surface ionization. The high surface energy of metallic nanoparticles like silver, copper or zinc is because of their nanoscale size, which causes the surface atoms to be more chemically reactive than in bulk materials. Surface metal atoms are oxidized (*e.g.*, Ag^0 to Ag^+) when they are exposed to aqueous or physiological conditions and release metal ions into the surrounding medium. This is usually gradual and sustained because the nanoparticle serves as a reservoir, which constantly replenishes ions by redox reactions at the surface. The rate and degree of ion release is dependent on the particle size, surface coating, crystallinity and the environmental conditions like pH and ionic strength. Since the liberation of ions is directly due to the surface of the nanoparticle, this route allows localized chemical activity at the nano-bio interface instead of diffusive activity throughout the bulk.³²

3.1.2 Reactive oxygen species (ROS) generation. The electron excitation and redox cycling at the nanoparticle surface are the basic processes that drive the ROS generation by antimicrobial nanomaterials. In metal oxide nanoparticles of semiconductors like TiO_2 or ZnO , light of energy equal or higher than the band gap of the material causes electrons to be excited out of the valence band into the conduction band leaving positively charged holes behind.³⁸ These excited electrons (e^-) and holes (h^+) move to the surface of the nanoparticle, where they undergo redox reactions with adsorbed molecules. The

electrons in the conduction band decrease the oxygen in the molecules to form superoxide radicals ($\text{O}_2^{\cdot-}$), and the holes in the valence band oxidize water or hydroxide ions to form hydroxyl radicals (OH^{\cdot}). The further reactions produce secondary ROS like hydrogen peroxide (H_2O_2) and singlet oxygen. Transition-metal nanoparticles can also be used to produce ROS in non-photoactivated systems by Fenton-like or redox cycling reactions, in which subsequent oxidation and reduction of metal ions in the nanoparticles catalyze the formation of oxygen-derived radicals. The small size of the nanoscale increases the yield of ROS by increasing the number of reaction sites on the surface and reducing the diffusion distance of reactive intermediates. Nanomaterials have been investigated in medical, environmental, and industrial systems to support the production of ROS as a mechanism of ensuring long-term antimicrobial activity.^{39,40} Fig. 3 shows the mechanism of ROS generation which causes to kill microbes by inducing widespread oxidative destruction of vital cellular constituents, such as membranes, proteins, and nucleic acids. Membrane lipids oxidation destabilizes the membrane integrity and permeability, whereas protein and DNA damage disrupts the metabolic processes, replication, and repair.³⁵

3.1.3 Catalytic activity. Catalytic antimicrobial nanomaterials work by enzyme-like chemical reactions whereby the nanoparticle surface repeatedly transforms benign substrates into reactive chemical products without being used up. Numerous metal and metal-oxide nanoparticles are used as nanozymes, which mimic natural enzymes, including peroxidase, oxidase, or catalase. Under normal conditions of a peroxidase-like reaction, the nanoparticle reacts with hydrogen peroxide to its active sites, reduces the activation energy barrier to O–O bond cleavage, and catalyzes its transformation to highly reactive hydroxyl radicals, which are useful for the killing of bacteria.⁴¹ The oxidase-like nanozymes have the capability to catalyze the production of reactive oxygen species by the direct use of molecular oxygen without the need for hydrogen peroxide. This is useful for applications in environments with low levels of hydrogen peroxide. The catalase-like nanozymes have the capability to degrade hydrogen peroxide into water and oxygen, which is useful for the regulation of oxidative stress as well as the enhancement of oxygen supply to tissues.⁴² Multi-step redox cascades are catalyzed by other catalytic systems, in which electrons are passed between substrates and surface metal centers, to regenerate the active catalytic state after each cycle. Since the nanomaterial is not consumed during the production of reactive species, catalytic pathways will enhance the antimicrobial activity by repeatedly converting the nanomaterial, making them fundamentally different from stoichiometric antimicrobial agents.⁴³

A comparative summary of the physicochemical parameters influencing different antimicrobial mechanisms, including catalytic nanozyme activity and photothermal antibacterial strategies, is provided in Table 1.

3.1.4 Nanoscale carriers for controlled antibiotic delivery. Antibiotic carriers based on nanomaterials are based on chemically programmed release mechanisms, as opposed to direct antimicrobial chemistry. Antibiotics are encapsulated,



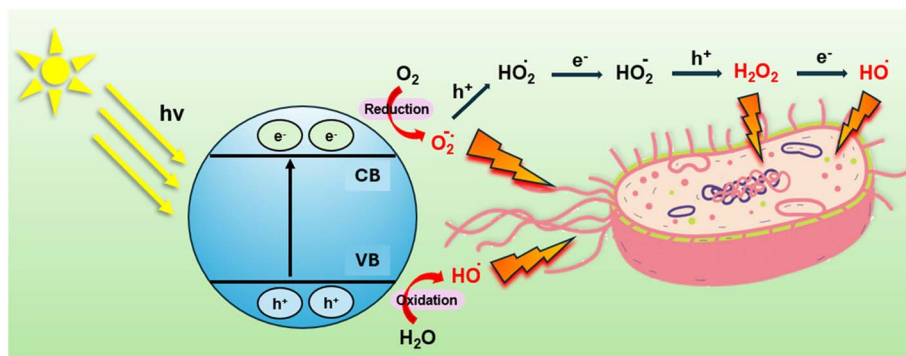


Fig. 3 Mechanism of ROS generation against microbes. Under light irradiation ($h\nu$), electrons are excited from the valence band (VB) to the conduction band (CB) of the semiconductor, generating electron–hole pairs. The electrons reduce O_2 to form $O_2^{\cdot-}$, while holes oxidize H_2O to produce $\cdot OH$. Subsequent reactions generate HO_2^{\cdot} and H_2O_2 , and the resulting reactive oxygen species damage bacterial membranes and cellular components, leading to bacterial inactivation. (Figure created by the authors; some graphical components were sourced from Canva and adapted).

adsorbed or chemically conjugated into nanoscale carriers like polymeric nanoparticles, lipid vesicles, dendrimers or inorganic shells. The release is controlled by specified chemical stimuli, such as degradation of polymers, breaking of bonds or environmental sensitivity. As an example, biodegradable polymer carriers are hydrolyzed by breaking ester or amide bonds, which break down the matrix and releases the drug that is encapsulated. Antibiotics can be conjugated in stimuli-responsive systems, where the conjugation is done through pH-sensitive or redox-cleavable bonds that are stable in circulation, but dissociate in response to infection-related chemical conditions. This is a controlled delivery system which enables antibiotics to be discharged in a spatially and time regulated fashion,

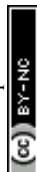
enhancing the local concentration of the drug and reducing premature degradation or systemic exposure.⁴⁸

3.2 Physical interactions

3.2.1 Membrane disruption. One of the most documented types of physical interactions between antimicrobial nanomaterials and microbes are direct interactions with microbial cell membranes. The intensive contact of engineered nanostructures like nanoparticles, nanosheets or textured surfaces with the outer envelope of bacteria and other pathogens results in structural compromise of the membrane layer. Studies in antimicrobial nanotechnology point to the fact that this disruptive contact, which is provided by the high surface area

Table 1 Physicochemical parameters influencing dominant antimicrobial mechanisms in nanomaterials

Mechanism	Typical nanomaterials	Key physicochemical parameters	Environmental requirement	Typical biomedical applications	References
Peroxidase-like nanozyme activity	Fe ₃ O ₄ , CuO, Pt, Au nanozymes, metal–organic frameworks	Small particle size (≈ 1 –20 nm), high density of catalytic active sites, redox-active metal centers, tunable surface charge	Requires H ₂ O ₂ for catalytic activation	Antibacterial wound dressings, infection-site therapy, biofilm disruption	44 and 45
Oxidase-like nanozyme activity	Au nanoclusters, MnO ₂ , CeO ₂ nanoparticles	Surface catalytic activity, oxygen activation capability, defect-rich surfaces	Uses dissolved O ₂ (no H ₂ O ₂ required)	Antimicrobial coatings, surface sterilization, biosensing platforms	44
Catalase-like nanozyme activity	CeO ₂ , MnO ₂ nanoparticles	Oxygen vacancies, redox switching (e.g., Ce ³⁺ /Ce ⁴⁺), stable surface chemistry	Requires H ₂ O ₂ but decomposes it to O ₂ and H ₂ O	Wound healing materials, oxidative stress regulation, tissue-protective antimicrobial systems	44
Photothermal antibacterial therapy	Au nanorods, CuS nanoparticles, graphene derivatives, polydopamine nanoparticles	Strong near-infrared (NIR) absorption, plasmonic properties, high photothermal conversion efficiency, optimized particle size (10–100 nm)	External NIR light irradiation	Implant coatings, localized infection therapy, biofilm eradication	44 and 45
Photocatalytic antimicrobial activity	TiO ₂ , ZnO, g-C ₃ N ₄	Semiconductor band gap (≈ 2 –3.5 eV), high surface area, efficient charge separation	UV or visible light irradiation	Antimicrobial coatings, water disinfection, medical device sterilization	46 and 47



and shape control of nanoscale materials, can cause the cell boundary of the microbe to become compromised, leaving the cell incapable of sustaining normal physiological balance. A significant number of modern investigations describe this type of interaction as a characteristic of physical antimicrobial activity in complex materials, in particular, surface finishes and biomedical interfaces where contact with pathogens is common.^{49,50}

3.2.2 Nano-sharp interface effects. Some antimicrobial nanomaterials are designed or have nanoscale sharp edges, *e.g.* the edges of graphene derivatives, nano-pillars, or other high-aspect-ratio structures, which form highly localized points of contacts with microbial surfaces. Recent literature explains the interface of these nano-sharp geometries, which are commonly referred to as nano knives or nano-spikes, with cells at very fine length scales, which lead to strong interactions with microbial envelopes when pathogens come into contact with them. These nanostructured surfaces are based on naturally occurring antimicrobial topographies (*e.g.* insect wings) and are actively studied as broad-spectrum antimicrobial coating and material surfaces.^{51,52}

3.2.3 Mechanical stress. Mechanical stress on pathogens can also be exerted by antimicrobial nanomaterials using engineered topographies or structures that put mechanical stress on microbial cells upon contact. As an example, nanostructured surfaces with arrays of high-aspect-ratio protrusions or patterned nanofeatures produce stresses on cells during adhesion or exposure to external forces, which causes deformation which is part of inactivation. Recent studies in engineered bactericidal surfaces comment on the potential of the interaction between nanofeature geometry and external conditions (*e.g.*, fluid flow, cell adhesion forces) to increase mechanical stress on microbial envelopes, and discuss the application to water disinfection materials, self-sterilizing surfaces, and implant coating.^{52,53}

3.2.4 Surface-mediated pathogen inactivation. Besides discrete structural characteristics, the general surface architecture of nanomaterials such as patterns, textures and nanotopographies can mediate pathogen inactivation by providing an environment that compromises microbial persistence or viability on contact. This classification includes engineered surfaces of controlled roughness, nanoscale arrays of pillars or other morphological structures that favour pathogen adhesion in undesirable shapes as well as physical interactions that prevent survival. Extensive surveys of antimicrobial surface technologies highlight that surface-based methods are a unique functional category in nanomaterials studies since they do not rely much on chemical release to generate antimicrobial effects but instead on physical design.³⁸

3.3 Synergistic mechanisms

In the recent studies in antimicrobial nanotechnology, there has been a keen focus on incorporating more than one functional component in engineered nanomaterials in order to realize cooperative chemical-physical effects that are superior to those of single-mode systems. These synergistic nanoplatforms

can be based on a wide range of different materials. As an example, metal nanoparticles incorporated into polymeric or hybrid matrices to form multifunctional assemblies that act on multiple antimicrobial pathways.⁵⁴ These cooperative architectures are based on the complementary physicochemical characteristics of each of the constituents and make it possible to achieve greater antimicrobial activity in environments where the monofunctional methods are less efficient. Research has demonstrated that these multi-component nanomaterials can be used to offer superior protection against a wide variety of microbes than their individual components, through the incorporation of customized surface properties, embedded antimicrobial agents and responsive components that respond to environmental conditions, and are under active development in wound care, surface coating and biomedical devices.^{35,55}

In addition to simple combinations, multi-modal antimicrobial nanomaterials are being designed to deliver multiple antimicrobial activities simultaneously or in series, that is, a single nanoplatform can be used to execute different functional activities in inhibiting microbial growth. For example, studies on multifunctional nanomaterials focus on materials that combine antimicrobial agents with physical barrier properties or that offer improved delivery of therapeutics and other functional properties, which lead to materials that are effective against a variety of microbial phenotypes and conditions.^{35,55} This multi-modal approach does not only expand the range of microbial inhibition but can also decrease the chances of resistance development by exposing the pathogens to different challenges that are harder to overcome by the traditional resistance mechanisms. Multi-modal nanomaterials can be considered a priority of modern research to overcome the limitations of traditional antimicrobial treatment and help to implement next-generation infection control approaches by decreasing the dependence on one mechanism of action and increasing the overall antimicrobial efficacy.^{56,57}

Table 2 shows the different nanomaterial types, examples for them, microbes tested and their mechanism of antimicrobial activity.

4 Applications in biomedical and healthcare industries

4.1 Clinical therapeutics

Antimicrobial nanomaterials have been clinically relevant in a number of critical fields of contemporary healthcare with benefits over traditional therapies in that they provide antimicrobial activity with improved functional performance to meet the needs of particular medical issues.⁶⁹ Wound healing and infection control is one of the significant fields of application, as nanomaterials are integrated into the advanced wound dressings and scaffolds to decrease the microbial burden and promote tissue repair.⁷⁰ Previous works note that nanomaterials, such as metal and metal-oxide nanoparticles, polymeric carriers, and composite systems, have been widely investigated as having the capacity to enhance infection control, promote wound healing, and tissue regeneration in chronic,



Table 2 Nanomaterial types, examples for them, microbes tested and mechanism of antimicrobial activity

Nanomaterial type	Example nanomaterials and microbes tested	Mechanism of antimicrobial activity	Key nanomaterial properties	Ref.
Metals (metallic NPs)	Silver nanoparticles (AgNPs)- <i>E. coli</i> & <i>S. aureus</i>	AgNPs interact with bacterial membranes and internal components, generate ROS, disrupt membranes, and interfere with cellular processes and replication	Size typically 5–100 nm; spherical morphology; high surface area; size-dependent Ag ⁺ release and ROS generation	58
	Silver-reduced graphene oxide nanocomposite (rGO/AgNPs)- <i>S. aureus</i> , <i>P. aeruginosa</i> , <i>S. mutans</i> , <i>C. albicans</i>	Nanocomposite binds to biofilm proteins and disrupts microbial biofilms and growth	AgNPs anchored on rGO sheets; large surface area; improved dispersion; synergistic interface interactions	59
Metal oxides	Zinc oxide nanocomposites (ZnO/EggShell)- <i>E. coli</i> , <i>P. aeruginosa</i> , <i>S. aureus</i> , <i>C. albicans</i>	ZnO nanostructures interact with microbial membranes and cellular components; biofilm inhibition observed in mixed bacteria and fungi	Nanostructured ZnO (typically <100 nm); high crystallinity; morphology-dependent activity (rods/spheres)	60
	α -Fe ₂ O ₃ nanoparticles- <i>S. aureus</i> , <i>E. coli</i> , <i>P. aeruginosa</i>	Iron oxide NPs increase membrane permeability, disrupt efflux pumps, and inhibit biofilm formation	Particle size ~20–100 nm; stable oxide phase; possible magnetic behavior	61
Polymeric nanomaterials	Selenium/CuO nanocomposite (SeNPs/CuO)-multi-drug resistant clinical pathogens	Metal-oxide nanocomposite disrupts resistant bacterial growth across multiple clinical isolates	Bimetallic composite; high surface reactivity; synergistic redox activity	62
	Chitosan nanoparticles- <i>E. coli</i> , <i>S. aureus</i> , <i>Listeria</i>	Cationic polymeric NPs interact with negatively charged bacterial membranes, leading to membrane disruption and inhibition	Size ~50–200 nm; strong positive surface charge; biodegradable polymer matrix	63
Carbon-based nanomaterials	Chitosan/Ag/GO composite (ChAgG)- <i>E. coli</i> , <i>S. aureus</i>	Combined polymer, Ag nanocrystals, and GO provide enhanced membrane interactions and microbial inhibition	Hybrid system; high surface area; combined metallic and polymeric functionalities	64
	Graphene-based TiO ₂ /CaO nanocomposite- <i>E. coli</i> , <i>S. aureus</i> , molds/yeasts	Graphene supports embedded metal oxides for microbial membrane interaction and ROS-linked effects	2D sheet structure; high conductivity; large surface area; nanoparticle anchoring	65
Hybrid materials	GO/AgNP and GO/CuNP composites- <i>E. coli</i> , <i>S. aureus</i>	Graphene oxide functionalised with metallic NPs enhances contact interactions with microbes and inhibits growth	Functionalized GO sheets; metal NP decoration; high surface reactivity	66
	ZnO–Ag–MWCNT nanocomposite- <i>E. coli</i> , <i>S. aureus</i>	Hybrid nanocomposite exhibits enhanced antimicrobial efficacy via combined effects of metal ions, surface interactions, and potential ROS pathways	Multi-component system; high aspect ratio nanotubes; enhanced electron transfer	67
	Metal oxide/alginate nanocomposites- <i>E. coli</i> , <i>P. aeruginosa</i> , <i>S. aureus</i> , <i>E. faecalis</i>	Alginate matrix with metal oxide NPs enables sustained interactions and inhibits both gram-negative and gram-positive bacteria	Polymer-embedded NPs; controlled release; improved stability and dispersion	68

acute, and burn wounds, which is a major advancement in wound care practices.⁷¹

Antimicrobial nanomaterials are used as nanocoatings and surface modifications in the biomedical implants and device

protection field to minimize the risk of postoperative infections and biofilm formation, which are the leading causes of implant failure and clinical complications. Surface functionalization with nanotechnology-through metallic, metal-oxide, and two-



dimensional nanomaterials-offers better antimicrobial performance at the device-tissue interface than untreated implants and is currently being explored as a method to reduce the risk of infection, increase biocompatibility, and overall clinical outcomes of orthopedic, dental, and other implanted devices.^{72,73}

The use of antimicrobial nanomaterials is also becoming increasingly important in the treatment of drug-resistant infections, especially in cases where traditional antibiotics are not as effective. Nano-delivery engineered antimicrobial nanostructures are under development to target multidrug resistant pathogens, disrupt biofilms, and localize therapeutic effect to infection sites, as alternatives and or adjuncts to systemic antibiotic therapy in challenging-to-treat clinical situations. Their possible application in improving the effectiveness of treatment of resistant bacterial infections is mentioned in reviews of nano-enabled anti-biofilm strategies and antimicrobial nanoparticle applications, which is a major issue in modern clinical therapeutics.^{74,75}

4.2 Biofilm prevention and control

Antimicrobial nanomaterials are becoming more significant in the prevention and control of biofilms in biomedical and healthcare sectors, especially by the creation of surface-engineered materials that prevent microbial adhesion and destabilize mature biofilms on medical devices. Biofilms-organized microbial communities that are enclosed in an extracellular polymeric substance that they produce themselves, are a significant cause of device-associated infections and are extremely resistant to traditional antimicrobial interventions.⁷⁶ Nanotechnology approaches have been designed to alter the surfaces of implants and medical devices at the nanoscale to form coating or textured surfaces that can inhibit initial bacterial adhesion, reduce colonization, and inhibit the formation of biofilms on the surfaces of medical devices. These surface modifications include anti-adhesive, bactericidal and disruptive surface modifications that operate at the interface between the device and biological fluids to reduce surface colonization and hinder the conversion of planktonic cells to mature biofilm communities, which is a major challenge in enhancing the longevity and safety of indwelling medical materials.^{77,78}

Recent studies review multifunctional antibiofilm materials that integrate nanostructured surface engineering with novel nanotherapeutic approaches to prevent biofilm formation as well as treat established biofilms on biomedical devices, highlighting the scope of strategies currently being explored to be used in clinical practice.⁷⁹ These are the application of nano-coatings, *e.g.*, metal and metal-oxide nanoparticles, through physical deposition methods to impart long-term antibiofilm characteristics, and the development of surface physicochemical characteristics that suppress microbial adhesion and promote biocompatibility.⁸⁰ With the help of the physicochemical properties of nanoscale materials, including high surface area and tunable surface chemistry, engineered surfaces can resist initial microbial adhesion and destabilize the

protective architecture of pre-existing biofilms, which can also help to improve the infection control process and reduce device-associated morbidity.⁸¹

4.3 Targeted nanocarrier systems

Nanomaterials have been extensively investigated in targeted nanocarrier systems to enhance the accuracy of antimicrobial agent delivery, to enhance bioavailability, local drug concentration at the sites of infection, and reduce systemic toxicity relative to traditional therapies. Nanoscale carriers like liposomes, polymeric nanoparticles, micelles, dendrimers and nanogels offer a platform to encapsulate a wide variety of antimicrobial compounds, shield them against premature degradation and enhance their pharmacokinetic characteristics by enhancing their distribution within the body and increasing their circulation time. Surface-modified versions of these nanocarriers can be used to regulate biological interactions and tissue affinity, which enhances more focused delivery to infected tissues and minimizes exposure to healthy organs-a major benefit in the treatment of complex infections and the minimization of undesired systemic side effects.^{82,83}

The recent developments in the targeted nanocarrier studies have focused on the increased bioavailability and decreased toxicity by employing strategic physicochemical design and functionalization. As an example, polymeric and lipid-based carriers can be programmed to deliver hydrophilic and hydrophobic antimicrobial agents, enhance drug stability, and enhance penetration of biological barriers, which eventually enhances therapeutic efficacy against persistent pathogens.⁸⁴ Surface functionalization with ligands or receptor-binding moieties allow nanocarriers to specifically target cells or microbial habitats and further concentrate the therapeutic into the area of need and reduce off-target delivery. Such advances in nanocarrier engineering are reported in the recent studies as potentially effective solutions to the shortcomings of low solubility, rapid systemic clearance, and non-specific distribution of conventional antimicrobial therapies.^{82,85}

Table 3 provides a summary of recent studies utilizing different nanocarrier types, examples, targeted places, and mechanism of antimicrobial delivery.

5 Current challenges and considerations

Fig. 4 shows the current limitations of nanomaterials in AMR. A few of the are discussed below considering recent studies as examples.

5.1 Cytotoxicity and biocompatibility

Antimicrobial nanomaterials are promising solutions to healthcare problems, yet the trade-off between high antimicrobial efficacy and safety to human cells and tissues is a major challenge to clinical translation. Nanomaterials, particularly metal-based nanoparticles that are commonly investigated as antimicrobial agents, are dose-dependently cytotoxic and can induce adverse biological responses, including oxidative stress,



Table 3 Summary of nanocarrier types, their targeted places and mechanism of action

Nanocarrier type	Example	Targeted place/infection site	Mechanism of targeted antimicrobial delivery	Stage of development	Ref.
Liposomes	Antibiotic-loaded liposomal formulations (<i>e.g.</i> , colistin, tobramycin)	Pulmonary infections, biofilms	Encapsulation of antibiotics within lipid bilayers enhances accumulation at infection sites and improves biodistribution and retention at infected tissues relative to free drug; functionalization with ligands or surface charge can further promote targeted binding to bacterial cells or infected tissue	<i>In vitro/In vivo</i> (preclinical; some clinically translated analogues)	86
	Liposomal antibiotic delivery to intracellular pathogens	Intracellular infections (<i>e.g.</i> , macrophage-harbored bacteria)	Liposomes enable internalization into host cells <i>via</i> endocytosis, increasing intracellular antibiotic concentration and thereby targeting pathogens within host cells	<i>In vitro/In vivo</i>	87
Polymeric nanoparticles	PEG- <i>b</i> -PCL (poly(ethylene glycol)- <i>b</i> -poly(ϵ -caprolactone) nanoparticles loaded with antibiotics)	Biofilm-associated infections and deep tissue infections	Size, surface chemistry, and ligand modification allow enhanced retention at infection sites, improved penetration through biological barriers (<i>e.g.</i> , biofilm matrices), and sustained release of antibiotic payloads	<i>In vitro/In vivo</i> (preclinical)	88
	PLGA or other polymeric antibiotic nanocarriers	Lung, systemic infections	Surface modifications (<i>e.g.</i> , antibodies or receptors) facilitate active targeting to pathogen-associated markers or infected tissue, improving local drug accumulation and bactericidal efficacy	<i>In vitro/In vivo</i> (preclinical)	88
Micelles	Amphiphilic block copolymer micelles carrying antibiotics	Respiratory or systemic bacterial infections	Micelles self-assemble to encapsulate hydrophobic antimicrobial agents, and functional ligands (<i>e.g.</i> , antibiotic moieties such as vancomycin) can be attached to the micelle surface to target bacterial cell walls and trigger site-specific delivery and release	<i>In vitro/In vivo</i> (early preclinical)	88 and 3
Dendrimers	Poly(amidoamine) (PAMAM) and related dendrimer nanocarriers	MRSA, gram-negative & positive infections	Highly branched polymer structures with surface functionalization allow targeted interactions with bacterial membranes or receptors, enhancing delivery of antibiotics to microbial communities	<i>In vitro/In vivo</i> (preclinical)	89
	Nanocarriers combining dendrimers with antibiotics	Biofilms and systemic infections	Dendrimers can be engineered to increase cell uptake and interaction, permitting deeper penetration into bacterial biofilms and infected tissues compared to non-targeted carriers	<i>In vitro/In vivo</i>	89



Table 3 (Contd.)

Nanocarrier type	Example	Targeted place/infection site	Mechanism of targeted antimicrobial delivery	Stage of development	Ref.
Nanogels	Antibiotic-loaded nanogels (e.g., camphor/thymol or other agents)	Skin, soft tissue and localized infections	Nanogels encapsulate therapeutic agents in a polymer network that facilitates controlled release at infection sites; surface chemistry or stimuli responsiveness (e.g., pH) can be modified to enhance targeting and release kinetics	<i>In vitro</i> / <i>In vivo</i> (preclinical)	90
	Nanogel delivery of essential oil derivatives	<i>Pseudomonas aeruginosa</i> infections	Nanogels use controlled release in local environments (e.g., pH-dependent dissolution) to concentrate antimicrobial agents where needed	<i>In vitro</i>	91 and 92

inflammation, and immune activation in mammalian cells; physicochemical properties such as size, surface charge, and chemical composition have a strong impact on these effects. Smaller particles are usually more efficient in penetrating biological barriers but are more likely to be reactive and potentially genotoxic, and some surface chemistries can make some cytotoxic to non-target cells, so care should be taken before considering their effects on human tissues in clinical use.⁹⁴ One of the major hurdles in cytotoxicity involves immunological

responses triggered by nanoparticles. Once nanomaterials are introduced into the systemic circulation, they quickly react with plasma proteins, creating a protein corona that may trigger the elements of the innate immune system. Specifically, nanoparticles can initiate the complement activation, a cascade of plasma proteins, which is typically used in host defense. Complement activation may result in the production of inflammatory mediators like C3a and C5a and opsonization of

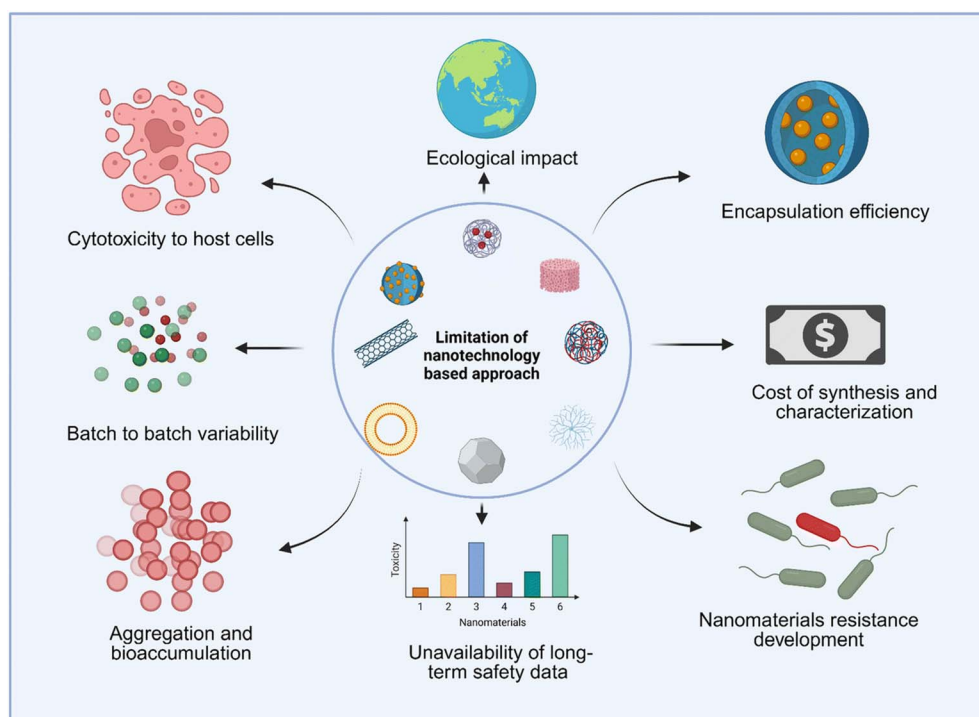


Fig. 4 Limitations of nanomaterials in AMR. The key challenges associated with nanotechnology-based approaches in combating AMR, including cytotoxicity to host cells, batch-to-batch variability, aggregation and bioaccumulation, ecological impacts, limited encapsulation efficiency, high costs of synthesis and characterization, potential development of resistance, and the lack of long-term safety data. The figure is obtained from ref. 93 under a Creative Common (CC BY) License.



nanoparticles, which are then rapidly taken up by macrophages in the liver and spleen and have lower therapeutic efficacy.^{95,96}

The other clinically relevant effect is the accelerated blood clearance (ABC) effect, which has been extensively reported in PEGylated liposomes and other nanocarriers. Following initial exposure to PEG-coated nanoparticles, the immune system can generate anti-PEG antibodies that identify the subsequent doses, leading to rapid clearance of the circulation and liver accumulation.⁹⁷ This effect can greatly impair the performance of the therapeutic process and has brought up the issue of repeated dosing regimens. Moreover, complement activation can cause hypersensitivity reactions or pseudoallergic reactions, which have been observed in certain clinical nanomedicine preparations. Such immunological responses are especially pertinent to antimicrobial nanomaterials that are to be used in systemic therapy because unintended immune responses may cause inflammation, shortened circulation time, and bi-distribution changes.

To guarantee biocompatibility and reduce toxicity, a systematic evaluation based on *in vitro* and *in vivo* research and the application of design measures to minimize the adverse effects and preserve antimicrobial activity are necessary. To reduce immunogenicity and off-target toxicity, approaches like surface functionalization with biocompatible polymers, encapsulation with benign matrices and use of inherently biocompatible nanomaterial can be used, although long-term safety data is still limited on most nanoplatforms. Previous works on safety of nanomaterials highlight that reciprocal interactions with host proteins, lipids and immune cells are key determinants of biocompatibility and that toxicological profiling, in terms of effects on major organs and immune responses, is a key step for advancing nanomaterials towards clinical applications.^{71,98}

5.2 Biodegradability and clearance

The antimicrobial nanomaterials pose serious issues regarding biodegradability and biological clearance, which are important factors to consider regarding the long-term safety and sustainability of their use in healthcare and the environment. Nanomaterials with many clinically relevant properties (including metallic nanoparticles or larger inorganic structures) do not undergo biodegradation in biological systems, and thus accumulate in organs (such as liver, spleen and kidneys) and may persist longer than they should, which may complicate long-term toxicity profiles and depend on physicochemical properties such as size, surface chemistry and coating materials.^{99,100} Also, biodegradable polymers like PLGA, polylactic acid (PLA) and some natural polymers are engineered to degrade into metabolizable byproducts that can more predictably be eliminated out of the body, but even these systems may leave residual materials based on the kinetics of degradation and the environmental situation.⁹⁹ In addition to human health, the environmental fate of the nanomaterials discharged through medical waste, manufacturing effluents or disposal is also a concern, with materials such as silver and zinc oxide nanoparticles potentially being retained in soils and aquatic environments and accumulating in the sediments and affecting

ecological communities long after their primary use.^{101,102} Due to the fact that the long-term fate, clearance mechanisms, and possible bioaccumulation of nanomaterials are not fully comprehended, the biodegradation pathways, organ distribution, and environmental transformation processes should be thoroughly studied, and the standards of assessing clearance and ecological impact should be developed to make sure that nanotechnology is used responsibly in healthcare and beyond.¹⁰³

5.3 Manufacturing and standardization

In addition to biological safety, biodegradability and clearance, scalability of manufacturing and standardization of regulatory requirements are other obstacles to clinical translation. The products of nanomedicine should be manufactured under Good Manufacturing Practice (GMP) conditions, and their characteristics should be regulated by clearly defined critical quality attributes (CQAs). Significant CQAs are particle size, size distribution (polydispersity index), surface charge (zeta potential), morphology, chemical composition, drug loading efficiency, and release kinetics, which can have a strong impact on pharmacokinetics, biodistribution, and therapeutic performance. Even small changes in the size or surface chemistry of nanoparticles between production batches can cause profound changes in biological behavior, and tight control of the process is a requirement to be approved by the regulatory authorities.¹⁰⁴

A combination of sophisticated physicochemical characterization methods is needed to achieve batch-to-batch consistency. Dynamic light scattering (DLS) is a common method of measuring hydrodynamic size and polydispersity, whereas transmission electron microscopy (TEM) or scanning electron microscopy (SEM) can be used to give detailed information on particle morphology and structural integrity. Zeta potential measurements are used to measure surface charge and colloidal stability, and spectroscopic and chromatographic methods can be used to measure chemical composition, surface functionalization, and drug loading. Nevertheless, these traditional techniques usually give incomplete data on the behavior of nanoparticles in physiological conditions. As an example, nanoparticles can be aggregated, degraded, or form protein corona in biological fluids, which can significantly change their biological identity relative to measurements conducted in simple buffer systems.¹⁰⁴

At the same time, regulatory challenges are also an added obstacle, with regulatory authorities like the U.S. Food and Drug Administration (FDA) and European agencies yet to have fully adapted frameworks and standardized safety and characterization procedures specific to nanomaterials, and developers are unsure of definitions, data required, and acceptable evidence to be approved.^{105,106} Besides, the lack of harmonized international testing guidelines and metrological standards of nanomaterial characterization including agreed measures of size, surface characteristics, and biological interactions, impedes the comparison of results across laboratories and regulatory jurisdictions, impedes the mutual acceptance of safety data, and slows regulatory review.^{107,108} These two issues are



interconnected to show that scalable, reproducible manufacturing and regulatory clarity is necessary to move antimicrobial nanomaterials into widespread clinical use and to make them safe and reliable in their clinical use.⁹³

6 Future directions in antimicrobial nanotechnology

6.1 Emerging platforms

The current state of antimicrobial nanotechnology is quickly progressing to the next generation of platforms incorporating novel architectures including nanozymes, MOFs and other emerging composites to overcome drug resistance and expand functional capabilities. Another major innovation area is MOF-based materials, which consist of inorganic metal ions and organic linkers, which can be assembled into highly porous and tunable structures that can be designed to improve antimicrobial activity, controlled release of agents and multifunctionality in biomedical applications.^{109,110} Recent studies point out that MOFs have been designed as flexible antimicrobial agents with tunable surface area, pore architecture and accessibility of active sites, which can be used as scaffolds to entrap antimicrobial compounds or metal ions and overcome the issues of sustained activity and biocompatibility in wound dressings and targeted therapeutics.¹¹¹

Simultaneously, nanozymes, artificial enzyme-like nanomaterials are also becoming potent antimicrobial platforms that combine the natural enzyme functionality with synthetic strength, frequently based on MOF structures or composites as the scaffold on which the catalytic activity is improved. In the recent literature, it is reported that MOF-derived nanozymes are under investigation to detect bacteria, targeted antimicrobial activity, and potential theranostic, with high structural stability and tunable catalytic activity that can be used in certain biomedical applications and surpass traditional antimicrobial agents.¹¹²

Also, studies have shown hybrid nanozyme/MOF systems, *e.g.* MOF-stabilized metal nanoparticle composites with enzyme-like catalytic activity, which have potential in antibacterial wound healing applications without the use of traditional antibiotics, demonstrating how these new architectures can be used to deliver multifunctional antimicrobial activity and overcome major challenges such as resistance and delivery.^{113,114}

These advances suggest that nanozymes and MOF-based systems are promising directions to antimicrobial nanotechnology of the future, which can provide novel solutions to combine catalytic activity, structural control, and therapeutic flexibility to next-generation infection control and clinical translation.^{115,116}

6.2 AI-enabled design

To overcome the long-standing limitations of trial-and-error methods of nanomaterial development, antimicrobial nanotechnology is adopting AI-enabled design and machine-learning-guided discovery to develop nanomaterials with enhanced antimicrobial properties faster and more efficiently.

As an example, machine learning models under supervision have been used to predict the bactericidal activity of nanostructured surfaces by using experimental data and determining which physical characteristics have the strongest impact on antimicrobial activity, allowing more effective choice of design parameters prior to synthesis and saving on experimental cost and time.¹¹⁷ In another recent work, machine learning methods were applied to simulate bacterial survival *versus* various doped metal oxide nanoparticles, which can be used to map the complex interactions between nanoparticle characteristics and microbial response, which can be used to optimize nanomaterial formulations to specific pathogens.¹¹⁸ The literature is broader in its focus on the fact that AI models, such as predictive algorithms and data-driven optimization frameworks, are currently being exploited to determine the best nanoparticle size, shape, surface chemistry, and functionalization to achieve the highest therapeutic performance and minimize toxicity and development time.¹¹⁹ These developments demonstrate that AI can be used to rationally design antimicrobial nanomaterials by combining high-dimensional experimental and computational information to reveal patterns and relationships that would be hard to identify by hand to direct researchers to more effective and efficient nanomaterial candidates with increased antimicrobial activity and translational potential.¹²⁰

In addition to predictive modeling, an even more ambitious goal of artificial intelligence in antimicrobial nanomaterials is inverse design, in which algorithms are given a desired biological performance, such as high activity against drug-resistant pathogens (*e.g.*, MRSA), low cytotoxicity to mammalian cells, and stability in physiological conditions, and the optimal nanomaterial composition, size, morphology, and surface chemistry to achieve these results are computed. Unlike traditional “forward” methods, which test materials and subsequently assess their properties, inverse design inverts the process by beginning with desired properties and searching for chemical space with candidate structures that are predicted to satisfy those properties.¹²¹

Inverse design using AI usually combines predictive and generative machine-learning models. Predictive models are trained to understand the correlation between nanomaterial characteristics, including particle size, surface functional groups, metal composition, and synthesis parameters, and biological responses such as minimum inhibitory concentration (MIC), cytotoxicity, or biofilm inhibition. Variational autoencoders, generative adversarial networks, reinforcement-learning algorithms, or Bayesian optimization frameworks can then be used to search the vast chemical design space and suggest new nanomaterial architecture that meet pre-specified performance criteria. These algorithms are useful to allow multi-objective optimization, which is a balance between antimicrobial efficacy, safety, stability, and manufacturability.¹²²

6.3 3D-printed antimicrobial materials

Antimicrobial nanotechnology is also adopting 3D-printed antimicrobial materials as a platform of the future that uses



additive manufacturing to create custom nanocomposite structures with inherent and durable antimicrobial properties. To illustrate how nanofillers can be incorporated into printable matrices to produce multifunctional biomedical components, recent studies have made 3D-printed resin nanocomposites that are modified with graphene nanoplatelets, which when produced by additive manufacturing have superior mechanical properties and long-term antimicrobial activity against *Candida albicans* without causing cytotoxic reactions, illustrating how nanofillers can be integrated into printable matrices to yield multifunctional biomedical parts.¹²³ Studies of nanocoated 3D-printed polymer composites also indicate that antimicrobial nanoparticles like silver (Ag) and zinc oxide (ZnO) can be coated onto additive manufacturing scaffolds to provide antimicrobial functionality against a variety of microbial strains, and thus are promising candidates to be used in patient-specific orthotic and medical applications.¹²⁴ Clinically, 3D-printed denture base resins reinforced with nano-zirconia and other nanoparticles have demonstrated a reduced microbial colonization of prosthetic devices, which has demonstrated the potential of 3D-printed nanocomposite materials in the real world in the fight against device-associated infections.¹²⁵ Also, another study of biodegradable polymer-based composites with metallic particles added (e.g., copper, aluminum, stainless steel, bronze) demonstrated that 3D-printed PLA-based antimicrobial sheets fabricated by fused filament fabrication had a high level of reduction in bacterial viability against common pathogens like *Escherichia coli* and *Staphylococcus aureus*, meaning that 3D-printed polymer matrices can host a variety of antimicrobial reinforcements to achieve effective antimicrobial performance in printed parts.¹²⁶ Taken together, these illustrations of recent studies and preprint studies demonstrate that additive manufacturing of nanocomposite materials can be used to create complex, customized structures with long-lasting antimicrobial functionality, and that such structures can be used in a wider range of applications in personalized medical devices, prosthetics, and healthcare components with embedded infection-resistant capabilities.^{127–129}

Conclusion

To sum up, antimicrobial nanomaterials are a highly versatile and fast-evolving category of materials that have a high potential to resolve the global problem of antimicrobial resistance. The various platforms mentioned in this review such as metallic and metal oxide nanoparticles, polymeric nanomaterials, carbon-based systems, and multifunctional hybrid materials provide distinct physicochemical characteristics that allow enhanced antimicrobial functionality and versatility to different biomedical settings. They are very appropriate in the development of advanced antimicrobial technologies due to their tunable structures and multifunctional capabilities. Notably, these materials have a great potential in real-life healthcare and biomedical applications. Antimicrobial nanomaterials are also finding application in advanced wound dressings to control infections and promote faster tissue healing, antimicrobial coating of implants and medical devices to prevent device-

associated infections, and biofilm-resistant surfaces of long-term biomedical equipment. Moreover, nanomaterial-based systems may be used as targeted drug delivery systems, which allow localized antimicrobial therapy with enhanced therapeutic efficacy and reduced systemic side effects. These applications are further extended by smart and stimuli-responsive nanomaterials that enable controlled activation at the sites of infection to enhance the precision of treatment. To enhance biocompatibility, scalability, and regulatory readiness to enable clinical translation, future studies should be directed towards enhancing the biocompatibility of nanomaterials, whereas emerging technologies, including nanozyme-based systems, metal-organic frameworks, and AI-guided nanomaterial design, can provide new opportunities to next generation antimicrobial solutions.

Author contributions

Dinithi Senanayake – literature search and drafted the manuscript. Imalka Munaweera – conceptualization, supervision, writing, review, and editing the manuscript. All authors have given approval to the final version of the manuscript.

Conflicts of interest

The authors declare that there is no conflict of interest.

Data availability

No primary research results, software or code have been included, and no new data were generated or analyzed as part of this review.

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