Nanotechnology, Science and Applications

Dovepress
Taylor & Francis Group

Open Access Full Text Article

REVIEW

Tiny Diamonds, Big Impact: Unlocking the Structure-Activity Relationship of Antimicrobial Nanodiamonds

Gabriela Wyszogrodzka-Gaweł, Maciej Stróżyk, Marta Skoda, Aleksander Mendyk

Faculty of Pharmacy, Jagiellonian University Medical College, Kraków, Poland

Correspondence: Gabriela Wyszogrodzka-Gaweł, Faculty of Pharmacy, Jagiellonian University Medical College, Kraków, Poland, Email gabriela.wyszogrodzka@uj.edu.pl

Abstract: Nanodiamonds (NDs) have emerged as a highly promising nanomaterial due to their intrinsic biocompatibility and remarkable antimicrobial and anti-adhesive properties, which result from their unique surface morphology. NDs serve as an excellent platform for extensive functionalization with diverse chemical groups and complex bioactive molecules, including peptides, photosensitizers, antibiotics and polycations. The antimicrobial potential of NDs has gained considerable attention in recent years across numerous application areas, including drug-delivery platforms, wound dressings, dentistry, surface coatings, biomedical implants, the food industry and water treatment technologies. This article compiles and critically evaluates the current microbiological evidence on ND antimicrobial activity. However, translating these findings into practical guidelines remains challenging due to the wide variability in reported results and the limited diversity of bacterial strains employed. The antimicrobial mechanisms of NDs in the context of Gram positive, Gram negative, and flagellated bacteria are examined, and it is demonstrated that key factors, including particle size, surface charge, and the composition of testing media, profoundly influence experimental outcomes and underlie many apparent contradictions in the field. Moreover, this review summarizes the functionalization strategies available for NDs, their reported biomedical and industrial applications, and current knowledge regarding their cytotoxicity and biocompatibility. Collectively, the article provides an integrated view of the structure-activity relationship governing ND antimicrobial performance.

Keywords: nanodiamonds, drug resistance, antibacterial nanomaterials, surface functionalization, biocompatibility

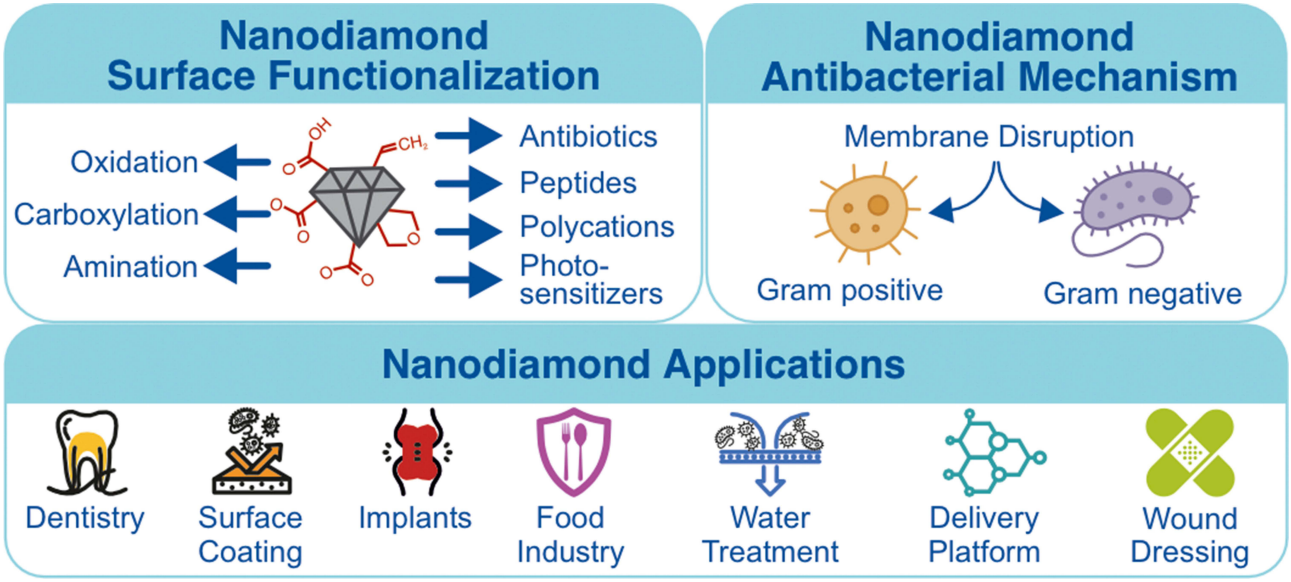
Introduction

Nanodiamonds (NDs) are a fascinating class of biocompatible, carbon-based nanomaterials, characterized by their distinctive physical, chemical, and optical properties. Their chemical inertness and exceptional hardness confer remarkable wear and corrosion resistance, all expressed at the nanoscale.[1,2] Typically measuring between 1 and 10 nm, NDs consist of carbon atoms arranged in a diamond-like lattice. They are said to have been synthesised since the 1960s, however the first report containing information on NDs was published in 1988.[3] Over time, they have attracted a lot of interest thanks to their unique features, including fluorescence, paramagnetism, and versatile surface morphology, resulting in a lot of research on their potential for biomedical applications.

Among their diverse applications, the antimicrobial properties of NDs have amassed significant attention in recent years. They are now being recognized as promising candidates for addressing the growing challenge of infectious diseases and microbial resistance. These issues represent a significant global health burden, according to a report published by WHO in 2025, they account for approximately 4.95 million deaths in 2019.[4] The increasing prevalence of antibiotic-resistant bacteria has necessitated the search for alternative antimicrobial agents. This urgency is driven by the declining efficacy of conventional antibiotics, largely due to the rapid evolution of resistant strains.[5,6] NDs, with their unique surface characteristics and ability to be functionalized, offer a novel approach to combat microbial infections, potentially reducing the amount of antibiotics.[7,8]

Received: 17 October 2025
Accepted: 12 December 2025
Published: 26 December 2025

Graphical Abstract



NDs are carbon-based nanoparticles (NP) predominantly composed of sp^3-hybridized carbon atoms, exhibiting a crystalline diamond core structure. The two principal synthesis routes are controlled detonation, which yields detonation nanodiamonds (DNDs), and high-pressure high-temperature (HPHT) carbon processing, which produces HPHT NDs. These ND types differ primarily in particle size (DNDs: 2–10 nm; HPHT NDs: 18–210 nm) and the uniformity of their surface groups following synthesis.[3,9] NDs exhibit antimicrobial activity due to physical, chemical and electrostatic interactions with bacterial membranes, which can compromise cell surface integrity, disrupt membrane permeability and interfere with nutrient transport or essential cellular processes.[5,10–14] While the studies referenced in this review employ either or both types of NDs, the analysis within this study emphasizes key particle attributes, specifically surface functional groups and particle size, as the principal determinants of antimicrobial activity and biocompatibility. Consequently, NDs will be differentiated according to these characteristics rather than their synthesis method, although these properties are intrinsically influenced by the synthesis process.

This review integrates current evidence on nanodiamond antibacterial mechanisms, applications, and biocompatibility into a single coherent overview, and it addresses a critical gap by consolidating contradictory information scattered across numerous studies and by proposing a coherent mechanistic explanation for how ND size, surface charge, and experimental conditions govern antimicrobial activity. Thereby clarifying previously inconsistent findings, with some scientists claiming antimicrobial activity and others negating it, which in our opinion is a result of differences in material tested, as well as the testing conditions. Furthermore, the article calls for future research to be conducted with standardized testing methods, under clinically relevant conditions.

Surface Functionalization

This section highlights surface homogenization or functionalization strategies of NDs, which allow for control over their hydrophilicity, surface charge, colloidal stability, and reactivity for targeted applications. Focus will be placed on oxidation, amination, carboxylation, conjugation with antibiotics, peptides, photosensitizers and polycations/metallic NPs.

Pristine NDs have a heterogeneous surface with low-density oxygen-containing groups (eg, hydroxyl, carbonyl, carboxyl) and non-diamond carbon (graphitic) impurities. Surface functionalization or homogenization is meant to enrich the ND surface with reactive groups (eg oxygen, carbonyl), remove graphitic carbon, reduce variability between the groups on the NDs surface, creating a uniform and reactive platform for further modifications. Functionalized NDs can

be conjugated with various bioactive agents, including antibiotics, polycations, photosensitizers or peptides. Surface functionalization is therefore a key step in enhancing the antibacterial efficacy of NDs and broadening their applicability in biomedical and technological contexts. A graphical overview of the discussed approaches is shown in Figure 1.

Oxidation

Oxidation of NDs, resulting in oxidated nanodiamonds (O-NDs), is a widely employed strategy to introduce oxygen-containing groups to their surfaces, enhancing reactivity, hydrophilicity, and colloidal stability. These modifications improve dispersion and enable further functionalization for applications ranging from biomedical imaging to drug delivery.

Thermal air oxidation, typically conducted at 450–550 °C, is a common method for modifying ND surfaces. Lazovic et al showed that treatment at 520 °C for 65 min reduces particle size (from approximately 4.4 nm to 3.4 nm), removes sp^2 carbon, and introduces oxygen-containing groups. Shifting the zeta potential from +6.4 mV to –50 mV and markedly improving colloidal stability.[15]

Mochalin et al demonstrated that thermal air oxidation of commercial NDs at 425 °C for 5 h yields a product with approximately 95 wt% diamond content and abundant surface C=O and O–H groups, as confirmed by IR spectroscopy. Subsequent hydrochloric acid (HCl) treatment effectively removed residual metal impurities.[16]

Chemical oxidation with strong acid mixtures is a common method for functionalizing NDs. Colon et al treated NDs in a heated 3:1 sulfuric acid (H_2SO_4) and hydrogen peroxide (H_2O_2) solution at 120 °C for 30–40 minutes, effectively adding oxygen groups and reducing aggregation, followed by washing and lyophilization for stable particles.[17] Similarly, Fang et al used a two-step process: first treating NDs with a 3:1 sulfuric and nitric acid (HNO_3) mixture at 90 °C for

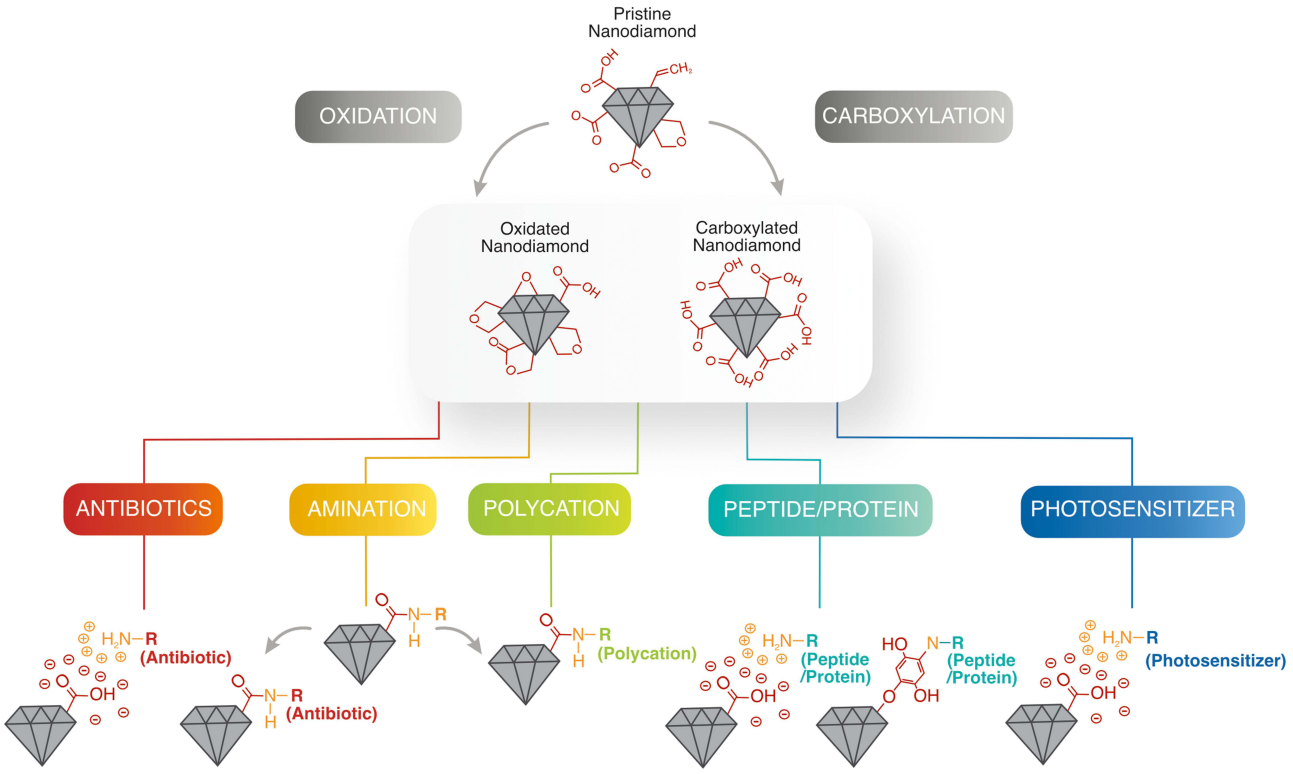


Figure 1 Graphical breakdown of surface functionalization possibilities. (i) Pristine NDs may be oxidated (O-NDs), approaches vary on method followed, most commonly via air oxidation. (ii) Pristine ND may be carboxylated (C-NDs), approaches vary on method followed, most commonly via acid treatment. (iii) O-NDs/C-NDs may be functionalised with antibiotics, most commonly via electrostatic interactions or hydrogen bonding, as well as amide bonds utilising linkers such as polyethylenimine (PEI). (iv) O-NDs/C-NDs may be functionalised with various amides via amide bond formation, most commonly done to enable further modifications. (v) O-NDs/C-NDs are usually functionalised with polycations via the initial amination of the NDs (vi) O-NDs/C-NDs may be functionalised with peptides/proteins, most commonly via electrostatic interactions, or covalent linkages utilising linkers such as benzotriazole (vii) O-NDs/C-NDs may be functionalised with photosensitizers such as phthalocyanine, most commonly via electrostatic interactions.

https://doi.org/10.2147/NSA.S575116

2 hours, then neutralizing, dialyzing, and heating in 0.1 M HCl at 90 °C for 2 more hours, yielding NDs rich in oxygenated groups with improved dispersibility and reactivity.[18]

Collectively, these oxidation techniques serve as critical preparatory steps, not only removing graphitic and amorphous carbon contaminants but also generating surfaces rich in carboxyl, hydroxyl, and carbonyl groups.

Carboxylation

NDs carboxylation introduces carboxyl (–COOH) groups to their surface, facilitating conjugation with amines, peptides, drugs, and polymers, primarily via amide bond formation. Additionally, carboxylation impacts biological interactions, including antimicrobial activity, by modifying surface charge and hydrophilicity.

A common carboxylation method involves oxidative acid treatment. Lei et al used a two-step protocol: oxidation in concentrated H_2SO_4/HNO_3 at 80 °C for 24 hours, neutralization with 0.1 M potassium hydroxide (KOH), followed by acidification in 0.1 M HCl at 90 °C for 6 hours. NDs were then washed until neutral to slightly alkaline pH, ensuring reagent removal and carboxyl group formation. Treated NDs showed strong Fourier transform infrared spectroscopy (FTIR) signals for C=O, C–O, and O–H, and increased oxygen content by energy-dispersive X-ray spectroscopy (EDS).[19]

Astuti et al carboxylated pristine NDs using a 3:1 H_2SO_4/HNO_3 mixture at 90 °C for 10 hours.[20] Garg et al applied the same acid blend at room temperature for 48 hours, followed by refluxing in 0.1 M sodium hydroxide (NaOH) and 0.1 M HCl at 90 °C for 2 hours each.[21] This post-treatment improved surface group stability and enhanced colloidal behaviour of the carboxylated nanodiamonds (C-NDs).

The presence of carboxyl groups on ND surfaces is linked to antimicrobial activity while acting as versatile intermediates for amination and biofunctionalization.

Amination

Amination of NDs is a strategy used to introduce nitrogen-containing functionalities, primarily amide or amine groups, onto the ND surface, thereby enabling further chemical derivatization, enhancing compatibility with polymers, and modulating biological interactions.

A streamlined solvent-free thermal method developed by Basiuk et al enables direct amidation of C-NDs with various diamines. The mixture of NDs and amines (eg, 1,12-diaminododecane, PEG-diamine) is heated under vacuum at 170–220 °C, depending on the amine. This simple approach requires minimal purification and is adaptable to diverse amine structures. Amide bond formation and functionalization were confirmed by FTIR, thermogravimetric analysis, and electron microscopy.[22] The method was later applied with octadecylamine to study the effect of NDs surface hydrophobicity on bacterial interactions.[23]

Complementing this thermally driven approach, Burleson et al employed hydrothermal and solvothermal reactions under acidic and basic conditions to aminate commercial detonation and synthetic NDs. FTIR confirmed NH_2 group incorporation, and the modified NDs showed enhanced loading of hydrophobic drugs such as doxorubicin and paclitaxel.[24]

Antibiotics

The immobilization of antibiotics onto NDs is a widely explored strategy to develop stable antimicrobial systems with prolonged therapeutic activity. Surface functional groups enable binding through electrostatic interactions, hydrogen bonding. Surface oxidation and carboxylation are often essential preparatory steps to ensure chemical compatibility between NDs and the antibiotics intended for immobilization.

One of the most employed strategies for antibiotic loading onto NDs is passive adsorption, involving direct incubation of antibiotics with aqueous ND suspensions. Shen et al[25,26] and Chernysheva et al[27] utilized this approach with antibiotics such as vancomycin, amikacin, and levofloxacin. Adsorption efficiency was primarily governed by the surface charge of both NDs and drug molecules: cationic antibiotics (eg, amikacin) bound to negatively charged C-NDs via electrostatic interactions, while anionic drugs (eg, levofloxacin) interacted through hydrogen bonding. Zeta potential analysis confirmed surface charge shifts upon adsorption, supported by FTIR spectroscopy indicating characteristic drug-related vibrations. Shen et al also

utilized this approach within their studies. Notably, the resulting ND-antibiotic complexes remained stable after extensive washing and incubation in physiological media, indicating strong and durable binding.[28,29]

Rouhani et al developed a PEI (polyethylenimine) mediated strategy for antibiotic loading. NDs were first acid-treated with H_2SO_4/HNO_3 to introduce surface carboxyl groups, followed by functionalization with PEI. The amine-rich PEI chains facilitated binding of amoxicillin via hydrogen bonding and electrostatic interactions. FTIR analysis confirmed the presence of characteristic β-lactam and amide peaks associated with amoxicillin, and the ND-drug complexes remained stable after repeated washing, indicating strong and durable binding.[30]

Peptides & Proteins

The conjugation of peptides onto ND surfaces represents a promising strategy to endow these particles with specific bioactivity, improved biocompatibility, and targeted antimicrobial or cell-interactive properties. Owing to their rich surface chemistry and high surface-to-volume ratio, NDs offer an attractive platform for anchoring short peptides and small proteins through both noncovalent adsorption and covalent coupling. Notably, the oxidation and carboxylation treatments discussed in previous sections generate surface-bound functional groups, particularly carboxyl moieties, that facilitate peptide immobilization via electrostatic interactions, hydrogen bonding, or covalent linkage.

Daskalova et al demonstrated this approach by incubating various types of NDs with bioactive fractions from *Cornu aspersum* snail mucus, containing peptides (<10 kDa) and proteins (>30 kDa). In the absence of crosslinking agents, biomolecules adsorbed onto ND surfaces within 30 minutes at room temperature. Although direct quantification of surface attachment was not performed, the observed enhancement in antibacterial activity against *Bacillus laterosporus*, relative to the mucus alone, indicated successful adsorption and preservation of bioactivity.[31]

In contrast, covalent conjugation offers greater stability and tunable attachment chemistry. Knapinska et al synthesized collagen-mimetic peptides and covalently linked them to C-NDs using a solid-phase peptide synthesis (SPPS) based coupling protocol. The NDs were first oxidized and acid-treated to introduce surface carboxyl groups, which were then conjugated to peptide N-termini using benzotriazole-based coupling agents in tetrahydrofuran. Successful attachment and structural integrity of the peptides were confirmed by fluorescence spectroscopy, matrix-assisted laser desorption/ionization time-of-flight (MALDI–TOF) mass spectrometry, and Raman spectroscopy. A zeta potential shift from –32 mV to +17 mV further indicated the incorporation of positively charged peptide residues.[32]

Mogil'naya et al compared two methods of binding of lysozyme, an antimicrobial enzyme, to NDs. Noncovalent adsorption enabled rapid attachment via brief vortexing and incubation but allowed desorption upon buffer washing. In contrast, covalent immobilization achieved by ND surface activation with benzoquinone produced stable ND-lysozyme conjugates with a loading capacity of approximately 0.2–0.25 mg of lysozyme on 1 mg of ND. Both methods preserved enzymatic activity. Antimicrobial assays against *Bacillus subtilis* and *Photobacterium phosphoreum*, supported by scanning electron microscopy (SEM) imaging, confirmed bacterial cell wall disruption.[33]

Lin et al systematically examined protein adsorption onto different ND types by varying pH to optimize electrostatic binding. Their findings, while focused on model proteins like myoglobin, bovine serum albumin (BSA), and insulin, offer direct relevance for peptide systems. Proteins were successfully immobilized through adsorption, as verified by zeta potential shifts, UV–vis spectroscopy, and MALDI–TOF mass spectrometry. Myoglobin and BSA formed stable 1:1 monolayer complex with the ND surface, while insulin displayed multimerization upon binding, indicating that protein or peptide structure can influence the binding mode and final complex configuration.[34]

Together, these studies demonstrate that peptide immobilization onto NDs can be achieved through either rapid, noncovalent adsorption or chemically driven covalent conjugation, each offering distinct advantages in terms of stability and ease of preparation.

Photosensitizers

Photosensitizers such as phthalocyanines (Pc) and porphyrins, which are aromatic macrocycles known for their light-activated production of reactive oxygen species (ROS), have been explored for photodynamic antimicrobial therapy (PACT). When interfaced with NDs, these compounds benefit from enhanced stability, dispersibility, and delivery potential.

A widely adopted strategy involves noncovalent adsorption of phthalocyanines onto NDs via extended π-π stacking. Openda et al[35] and Gvozdev et al[36] demonstrated that aromatic zinc and indium phthalocyanines spontaneously adsorb onto the graphitic surface domains of NDs when mixed in dry solvents such as dimetyloformamid (DMF) or dimethyl sulfoxide (DMSO), followed by prolonged stirring and sonication. These interactions are further stabilized by electrostatic attraction when the phthalocyanines are polycationic, enabling efficient complexation with the negatively charged ND surface. Gvozdev et al showed that titration with polycationic ZnPc dyes results in progressive red shifts and quenching of the Q-band absorbance, confirming face-to-face interactions. Dynamic light scattering and zeta potential measurements additionally confirmed formation of stable complexes with altered surface charge and hydrodynamic diameter.[36]

Covalent conjugation offers a more permanent attachment route. Openda et al synthesized asymmetrical porphyrin and phthalocyanine derivatives containing reactive –OH groups and conjugated them to C-NDs via carbodiimide chemistry (via DCC/NHS (N,N′-Dicyclohexylcarbodiimide/N-hydroxysuccinimide) activation in DMF). In this case, ester linkages were formed between the photosensitizer and the ND surface, while silver nanoparticles (AgNP) were co-incorporated to create ternary nanohybrids.[37] This multi-component assembly strategy was also explored in Openda et al, where the covalent ND-photosensitizer constructs were subsequently linked to chitosan-AgNP via amide bonds to form stable, multifunctional conjugates.[38] Confirmation of successful coupling across these studies was achieved using FTIR, Raman spectroscopy, and UV–vis absorbance, alongside transmission electron microscopy (TEM) imaging that revealed size increases from ~2.4 nm for pristine NDs to over 20 nm post-conjugation.[36,38]

Polycations and Silver

Surface cationization of NDs with polycations or metal NPs such as AgNP is another strategy to endow them with antimicrobial functionality. By introducing permanent or pH-responsive positive charges, these modified NDs disrupt negatively charged bacterial membranes through electrostatic interactions.

Li et al synthesized cationic NDs (cNDs) by coupling branched PEI to acid-oxidized NDs via carbodiimide chemistry. Pristine NDs were oxidized at 425 °C for 5 h, treated with H_2SO_4/HNO_3 (3:1) at 80 °C for 48 h, and subsequently heated in 0.1 M NaOH and HCl (100 °C, 1 h each). The cNDs were reacted with PEI in 2-(N-morpholino)ethanesulfonic acid (MES) buffer (0.1 M, pH 6.0) using EDC/NHS (1-ethyl-3-(3-dimethylaminopropyl)carbodiimide/N-Hydroxysuccinimide) activation. Functionalization was confirmed by a zeta potential shift (–36 mV to +31 mV), C–N and N–H FTIR bands, and nitrogen signals in X-ray photoelectron spectroscopy (XPS).[39]

Wang et al prepared quaternary ammonium-modified NDs (QNDs) via a three-step process comprising silanization, polymer grafting, and quaternization, followed by in situ silver reduction to yield QND-Ag hybrids. The polycationic shell was formed by coupling 3-bromopropyltrimethoxysilane to O-NDs, grafting with poly(4-vinylpyridinium-co-2-hydroxyethyl methacrylate), and quaternization using alkyl halides.[40]

Chang et al employed a comparable approach, functionalizing cNDs with poly-L-arginine via EDC/NHS coupling, followed by electrostatic assembly of citrate-stabilized AgNPs onto the cationic surface. The resulting Ag-ND hybrids were further coated with BSA, yielding Ag-ND@BSA constructs with improved colloidal stability in biological media.[41]

Cao et al similarly used QNDs as a precursor to in situ silver deposition, forming Ag/QND composites for use in dental materials. Their approach grafted a quaternized pyridinium-based polymer onto O-NDs using carbodiimide and silane-link chemistry, resulting in contact-killing QNDs. These were then loaded with Ag via hydroquinone-mediated reduction in the presence of sodium citrate.[42]

NDs Antibacterial Mechanism

Recent studies have highlighted the intrinsic antimicrobial properties of NDs, primarily arising from their physicochemical characteristics, including nanoscale morphology, surface functionalization, and associated surface charge. Physical and chemical interactions with bacterial membranes can compromise cell surface integrity, disrupt membrane permeability, interfere with nutrient transport, and induce electrostatic disturbances that impair essential cellular processes.[5,10–14] A schematic representation of the antibacterial mechanism of NDs against Gram-positive and Gram-negative bacteria is presented in Figure 2.

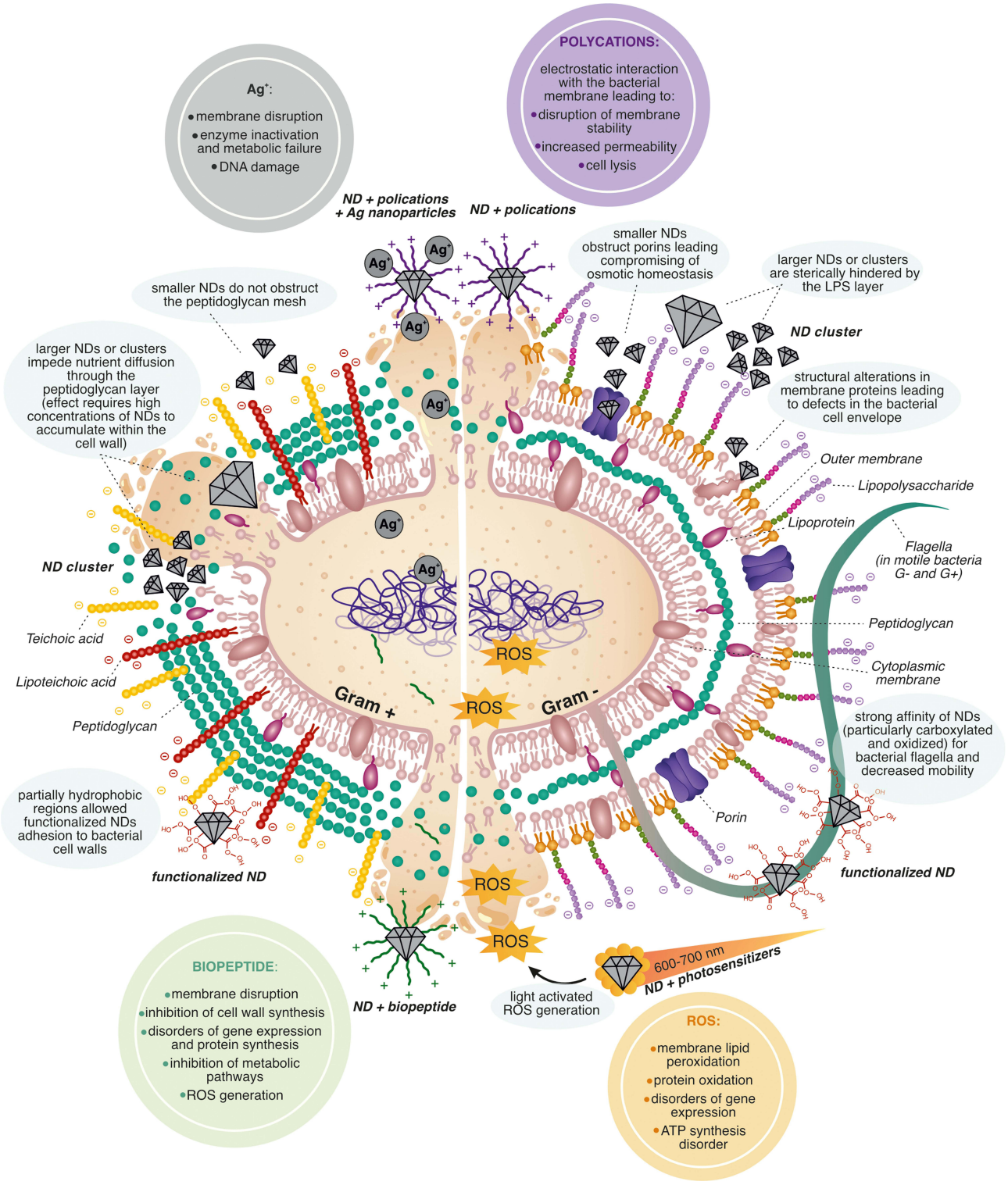


Figure 2 Schematic representation of the antibacterial mechanism of NDs against Gram-positive and Gram-negative bacteria.

Influence of Cell Wall Structure

As the microbial cell wall is the primary target of NDs, it is also therefore their most important line of defence. The differences in the cell envelope arising from whether a bacteria is Gram positive or negative influence their susceptibility to NDs. For instance, Gram-positive bacteria possess a thick peptidoglycan layer, within which teichoic and lipoteichoic

https://doi.org/10.2147/NSA.S575116

acids are embedded, whereas Gram-negative bacteria feature a markedly thinner peptidoglycan layer located within the periplasmic space, between the inner cytoplasmic membrane and an outer membrane enriched with lipopolysaccharides (LPS).[43] The thickness of the peptidoglycan layer varies significantly, ranging from a few nanometres in Gram-negative bacteria to about 100 nm in Gram-positive bacteria, representing a difference of more than an order of magnitude.

Beranova et al proposed that in Gram-negative bacteria, the initial interaction between NPs and the cell surface likely involves LPS moieties protruding from the outer membrane.[11] They further suggested that porins may constitute an additional target structure affected by NDs. These porin channels function as molecular sieves, permitting the passive diffusion of hydrophilic molecules up to around 600 Da into the periplasmic space. Given that the typical diameter of *Escherichia coli* porins is approximately 1.1–1.2 nm, the authors hypothesized that 5 nm NDs could obstruct these channels, thereby disrupting solute transport and compromising osmotic homeostasis. In contrast, larger NDs or aggregates may be sterically hindered by the LPS layer, which can act as a physical barrier, trapping ND agglomerates or larger particles and preventing their access to porin sites. Chatterjee et al further reported that interaction with NDs may induce structural alterations in membrane-associated proteins, leading to defects in the bacterial cell envelope. Raman spectral shifts provide additional evidence of such protein structural changes at the cell wall. Since the LPS layer in *E. coli* plays a critical role in protecting cells from osmotic stress, even minor structural disruptions could compromise its integrity. Such damage would facilitate the influx of water molecules into the cytoplasm, resulting in an internal buildup of osmotic pressure that ultimately leads to cell lysis.[14]

The thick peptidoglycan layer of Gram-positive bacteria acts as a robust barrier against external agents, including NPs, and is the primary site of interaction with NDs. Beranová et al proposed that NDs can substantially impede nutrient diffusion through the peptidoglycan layer; however, such a nonspecific effect requires high concentrations of NDs to accumulate within the cell wall. Authors observed increased susceptibility of *B. subtilis*, to larger NDs, which tend to form sizable aggregates. The authors proposed that these larger NDs (100 nm) or clusters are more effective at obstructing the otherwise permeable peptidoglycan mesh, thereby exerting a stronger antibacterial effect against Gram-positive bacteria than smaller NPs.[11] The findings of Norouzi et al conducted on Gram-positive strains of *Staphylococcus aureus* and *Staphylococcus epidermidis*, demonstrating that the aggregation of NDs and their adhesion to bacterial cell walls play a critical role in inhibiting bacterial growth. The authors examined NDs of 18, 25, 75, and 125 nm in size and demonstrated that colony-forming ability was not affected by particle size, but rather by concentration, as a clear dose-dependent inhibition of colony formation was observed in both bacterial strains. Additionally, 125 nm NDs exhibited the least reduction in bacterial colony-forming ability, indicating lower antibacterial efficacy.[44] Ong et al demonstrated that NDs ranging between sizes 18–125 nm exhibit antibacterial properties, particularly against Gram-positive bacteria like *S. aureus*, while showing minimal effects on Gram-negative *E. coli*.[45]

Previous observations regarding the size-dependent NDs antibacterial activity, where NDs with smaller NDs (approximately 5nm) exhibit greater efficacy against Gram-negative bacteria, while larger NDs (approximately 100nm) or their aggregates are more effective against Gram-positive strains were further corroborated by Sousa et al. In their study, negatively charged C-NDs with an average diameter of approximately 100 nm and a zeta potential of –29.21 mV were investigated using Langmuir monolayers as biomimetic models of bacterial cytoplasmic membranes. C-NDs demonstrated a more pronounced detrimental effect on Gram-positive bacteria, such as *S. aureus*, compared to Gram-negative bacteria. This enhanced activity was attributed to the deeper penetration of C-NDs into the hydrocarbon chains of the bacterial cell membranes, resulting in significant alterations to membrane structure and potential integrity. In contrast, interactions with Gram-negative bacteria, such as *E. coli*, were predominantly limited to the polar head groups of membrane lipids, rendering their membranes less susceptible to damage.[46]

As reported by Beranova et al,[47] Sawosz et al[48] and Chwalibog et al,[49] NDs can bind to bacterial cell membranes and flagella, are located non-specifically on the cell wall and in some cases even penetrate the cells. These interactions are species-dependent: *Salmonella enteritidis* (Gram-negative) and *Listeria monocytogenes* (Gram-positive) exhibited pronounced structural damage following exposure to NDs, whereas *S. aureus* (Gram-positive) appeared unaffected. It is important to note that these studies utilized NDs in the 2–10 nm size range. Consistent with earlier findings, NDs of this size demonstrate clear antibacterial activity against Gram-negative bacteria, such as *E. coli* and *S. enteritidis*. In this context, the observed strong inhibitory effect against *L. monocytogenes* appears to be an exception. However, despite

both being Gram-positive, *L. monocytogenes* and *S. aureus* differ significantly in morphology and motility: the former is a motile (has flagella), rod-shaped bacterium typically occurring singly, in pairs, or short chains, while the latter is a non-motile, spherical bacterium forming grape-like clusters. Sawosz et al observed a strong affinity of NDs for bacterial flagella. These filamentous appendages are composed of proteins rich in acidic amino acids, primarily aspartic and glutamic acid. The surfaces of C-NDs functionalized with carboxyl groups promote preferential attachment to flagellar structures, as carboxyl groups readily react with amino ($–NH_2$) groups found in amino acids. Therefore, the enhanced sensitivity of *L. monocytogenes* to NDs may be attributed to the presence of flagella, which serve as additional binding sites and facilitate stronger or more targeted interactions with the ND surface.[48]

Influence of Surface Charge

Bacterial hydrophobicity and surface charge are key physicochemical properties influencing susceptibility to NDs and adhesion to surfaces. Though distinct, these parameters are interrelated and reflect the bacterial cell surface composition. Hydrophobicity depends on lipids, surface proteins, LPS, and cell wall architecture, while surface charge arises from ionizable groups on the envelope. Usually, Gram-positive bacteria, with a thick peptidoglycan layer and teichoic acids, exhibit lower negative charge and higher hydrophobicity, facilitating adhesion to hydrophobic surfaces such as plastics and implants. In contrast, Gram-negative bacteria have a thinner peptidoglycan layer and LPS-rich outer membrane, resulting in a more negative surface charge and reduced hydrophobicity.

The surface charge of NDs, reflected by their zeta potential, critically influences antibacterial activity by modulating electrostatic interactions with bacterial membranes. This charge is pH-dependent, varies with ND size, and is strongly affected by the synthesis method. Surface modifications enable tuning of the zeta potential: oxygen-containing groups lowering the potential, while amination introduces positive charges. Since bacterial membranes are predominantly negatively charged, positively charged NDs (eg, amine-functionalized) demonstrate enhanced electrostatic attraction to bacterial surfaces.

Although both bacteria and C-NDs possess negative surface charges, C-NDs exhibit strong antibacterial activity. This paradox is explained by their ability to penetrate bacterial membranes through hydrophobic interactions. Upon adsorption, the partially hydrophobic regions of C-NDs insert into the lipid bilayer, disrupting van der Waals forces between hydrocarbon chains and inducing localized membrane thinning and increased permeability, effects especially pronounced in Gram-positive bacteria lacking an outer LPS membrane barrier.[46] Quan et al[50] and Sousa et al[46] showed that C-NDs significantly inhibit the growth of *Streptococcus mutans* and *S. aureus*, likely by disrupting bacterial membranes through surface charge interactions. Moskvitina et al reported that 5 nm C-NDs exhibited significant antibacterial activity against both *E. coli* and *S. aureus*.[51] Chatterjee et al revealed that C-NDs enhance antibacterial effects against *E. coli* compared to their pristine counterparts.[14] Similar findings were reported by Yang et al for 5 nm oxygen-terminated NDs. Proteomic analysis revealed significant changes in *S. aureus* protein expression, including upregulation of peptidoglycan-degrading enzymes and downregulation of a key biosynthetic enzyme, indicating impaired cell wall integrity and activation of bacterial stress responses. Wehling et al[52] established a direct correlation between oxygen-containing surface groups and reduced bacterial viability, highlighting the pronounced effect of highly reactive carboxylic acid groups in acid anhydride forms. The authors identified surface functionalization as a key determinant of the NDs antibacterial activity.[53]

Another approach to modifying ND surface charge is functionalization with polycations, which impart a strong positive charge. This enhances electrostatic attraction to negatively charged bacterial surfaces, promoting effective adsorption. The primary target of polycations is the bacterial cytoplasmic membrane, whose integrity is compromised, resulting in increased permeability, destabilization, and ultimately cell lysis. NDs functionalized with polycations and silver ions (Ag+) exhibit dual biocidal activity. Ag+ disrupts bacterial membranes via electrostatic interactions and binds to thiol (-SH) groups in proteins, leading to enzyme inactivation and metabolic failure. It also interacts with bacterial DNA, destabilizing the double helix, and promotes ROS generation, enhancing oxidative stress. Silver-loaded, polycation-functionalized NDs show strong antibacterial activity against both Gram-positive and Gram-negative bacteria, even at low concentrations.[40–42]

Role of Biological Media in Antimicrobial Efficacy Evaluation

Wehling et al[52] findings demonstrate that the NDs antibacterial activity is strongly influenced by their chemical environment and surface chemistry. Even simple cell culture media, such as Dulbecco's Modified Eagle Medium (DMEM), significantly reduce the NDs bactericidal effect. This suggests that the presence of proteins or small organic molecules interferes with the reactive surface groups of NDs, thereby inhibiting their activity. These reactive groups appear to form covalent or nonspecific interactions with proteins and other biomolecules, including those on cell membranes or intracellular targets. When such surface functionalities are neutralized or removed through binding, the antibacterial potential of NDs is diminished. This is consistent with the idea mentioned before, that certain functionalizations of NDs involving proteins occur through simple and rapid non-covalent adsorption. Dunseath et al reported only modest differences in antibacterial efficacy among NDs with various surface terminations (–COOH, –OH, –H), attributing this to passivation of reactive surface groups by organic components present in Tryptic Soy Broth.[54] Similarly, Budil et al demonstrated that the anti-adhesive properties of –H and –F terminated nanocrystalline diamond films, which reduced *E. coli* adhesion by around 50% in mineral media, were markedly attenuated in organic-rich environments.[55]

In conclusion, these findings suggest that in complex biological media, the apparent antimicrobial activity of diamond-based materials may result from indirect effects such as nutrient adsorption onto the diamond surface, rather than from direct bactericidal mechanisms, especially when high concentrations of NDs are tested. However, other researchers may observe and report low bactericidal activity of NDs, that may be a result of the adsorption of proteins or other media ingredients to the NDs surface, effectively inhibiting the NDs bactericidal activity. This is evidenced by the inverse relationship between fetal bovine serum (FBS) concentrations and antimicrobial activity of NDs.[44,52]

Antimicrobial Applications

NDs have emerged as multifunctional nanomaterials with intrinsic antimicrobial activity, as summarized in Table 1. In addition to their antimicrobial properties, NDs offer high biocompatibility, chemical stability, and versatile surface functionalization. These features support their growing investigation for applications in medicine, dentistry, food safety, water purification, and materials science, particularly for preventing microbial colonization and biofilm formation (Figure 3).

Delivery Platform

In medicine, NDs are being explored both for their antimicrobial properties and as drug delivery carriers. Their surface can be functionalized and conjugated with antibiotics, peptides, photosensitizers, metallic NPs to enhance or synergize antimicrobial efficacy.

In studies by Chernysheva et al, NDs were conjugated with antimicrobial agents including amikacin, levofloxacin, lysozyme, and miramistin. These ND conjugates exhibited prolonged retention on biological surfaces, leading to reduced bacterial infections and enhanced biocompatibility.[27,66]

Shen et al also investigated the adsorption of antibiotics onto NDs, demonstrating strong binding of vancomycin, levofloxacin, and amikacin to ND surfaces. Antimicrobial assays showed that ND-antibiotic composites effectively inhibited *S. aureus* adhesion and survival, supporting their potential use in bioprosthetic materials and prosthetic heart valves. Additionally, self-assembled C-NDs films exhibited anti-adhesive properties, preventing bacterial colonization.[25,26,28,29]

Daskalova et al investigated the antibacterial properties of NDs combined with peptides extracted from snail *Cornu aspersum* mucus. NDs significantly enhanced the bactericidal effects of these peptides against *B. laterosporus*, with the strongest effect observed in protein fractions above 30 kDa.[31]

Gvozdev et al investigated the use of NDs as carriers for polycationic zinc Pc in PACT. NDs (approximately 7 nm) effectively bound Pc via electrostatic interactions, with each particle accommodating approximately 50 Pc molecules. The ND-Pc complexes retained photodynamic activity and exhibited improved delivery, as Pc was temporarily inactivated while bound, potentially protecting it from ROS-mediated degradation. Importantly, the non-covalent interactions responsible for complex formation were weaker than the interactions between Pc and bacterial cell wall components, enabling the release of Pc upon contact with bacterial cells.[36]

Table 1 Summary of Reported Results on the Antibacterial Properties of NDs Within Different Applications

Bacteria Species	ND Size + Form	Antibacterial Assay	Findings	Ref.
Gram-Negative				
Escherichia coli	2-4 nm Pristine ND	Disk Diffusion	Low bacteriostatic effect.	[56]
	4 nm NDs integrated into titanium implant	Disk Diffusion & Implants incubated with E. coli inoculum	No inhibition zone. Three-fold reduction of bacterial surface coverage for ND-containing implant.	[57]
	3-5 nm O-ND embedded into film	Incubation & Absorbance measurement	Near 95% growth inhibition, film with 1% w/v ND.	[13]
	4 nm O-ND integrated into Poly-lactic acid scaffold	Incubation, Live/Dead staining and confocal microscope imaging	No effect observed.	[58]
	5 nm C-ND	Incubation & CFU (colony forming unit) count	30% CFU reduction at 10 mg/mL. 60% CFU reduction at 30 mg/mL. 100% CFU reduction at 100 mg/mL.	[51]
	5nm Pristine ND	Dilution and plating	95% CFU reduction at 30 μg/mL. Complete inhibition at 50 μg/mL.	[47]
	5 nm Pristine ND	Dilution and plating	Near 100% inhibition at 100 μg/mL.	[11]
	5 nm O-ND	Dilution and plating	Near 80% inhibition at 1000 μg/mL.	
	5 nm Pristine ND	Incubation & CFU count	60% CFU reduction at 1 mg/mL.	[59]
	5 nm O-ND	Incubation & CFU count	45% CFU reduction at 1 mg/mL.	
	5 nm C-ND	Incubation & Absorbance measurement	86% of E. coli underwent lysis within 4h at 100 μg/mL.	[14]
	4-6 nm C-ND	Incubation & Absorbance measurement	20% reduction in bacterial growth at 100 mg/mL.	[60]
	5-10 nm C-ND	Focused ion beam (FIB)/SEM analysis	80% antibacterial rate at 100 mg/L. 99% reduction of bacterial adhesion.	[29]
	3-10 nm Aminated ND embedded into membrane	Incubation & CFU count	52.4%, 59.7% and 63.7% increase in death rate at 250 ppm, 500 ppm, and 1000 ppm ND, respectively.	[61]
	6 nm Pristine ND	Incubation & CFU count	No reduction in CFU, at 0.1, 1.0 and 10 mg/mL.	[23]
	6 nm Aminated ND	Incubation & CFU count	Reduction in CFU at 0.1, 1.0 and 10 mg/mL not statistically significant.	
	10 nm Fluorosilane modified ND	GB/T 21510 (Analogue to ISO 22196–2011)	73% decrease of CFU vs control.	[62]
	18-125 nm ND	Incubation & CFU count	Varying ND concentrations (1–500 μg/mL) and sizes either had no effect or increased colony forming ability (CFA) in water or PBS.	[45]
	200 nm C-ND	Incubation & CFU count	87% increase in death rate at both 0.01 g/mL and 0.02 g/mL.	[17]
	ND-PVA composite	Broth microdilution	MIC observed at 4 μg/mL composite containing 0.004 mg/mL ND.	[63]

(*Continued*)

Table 1 (Continued).

Bacteria Species	ND Size + Form	Antibacterial Assay	Findings	Ref.
Porphyromonas gingivalis	150 nm O-ND	XTT assay, Live/Dead staining	95% increase in death rate at 2.5 mg/mL. 70–80% biofilm formation reduction at 1.25 mg/mL 73.3% increase in death rate in preformed biofilms at 10 mg/mL.	[64]
Salmonella	3-5 nm Pristine ND	Incubation and plating	20% bactericidal rate at 1 mg/mL vs control.	[39]
	3-5 nm cND suspension and embedded into epoxy	Incubation and plating	99.99% bactericidal rate, 80% biofilm inhibition at 1 mg/mL vs control. 99.99% bactericidal effect of epoxy embedded with NDs vs control.	
Gram-Positive				
Bacillus subtilis	5 nm Pristine ND	Dilution and plating	No effect in the entire range of tested concentrations	[11]
	5 nm O-ND	Dilution and plating	50% reduction in CFA at 1000 μg/mL	
	18 nm Pristine ND	Dilution and plating	70% reduction in CFA at 500 μg/mL.	
Bacillus sp.	5-10 nm C-ND	FIB/SEM analysis	90% increase in antibacterial rate at 100 mg/L.99% reduction in bacterial adhesion.	[29]
Brevibacillus laterosporus	2-10 nm ND	Surface inoculation	140% antibacterial rate at 100 mg/L NDs with peptides vs peptides alone. No control data.	[31]

https://doi.org/10.2147/NSA.S575116

Staphylococcus aureus	2-4 nm Pristine ND	Disk Diffusion	Low bacteriostatic effect.	[56]
	6 nm Pristine ND	Incubation & CFU count	Statistically insignificant reduction in CFU at 0.1 and 1.0 mg/mL. 32.66% increase in CFU at 10 mg/mL.	[23]
	6 nmAminated ND	Incubation & CFU count	18.74% reduction in CFU at 0.1 mg/mL. 42.44% reduction in CFU at 1.0 mg/mL. 91.24% reduction in CFU at 10 mg/mL.	
	<10 nm NDs integrated into PCL scaffold	Incubation with scaffold, after washing residing bacteria stained and imaged	Statistically significant reduction (50%) of bacteria adhered on 5% ND scaffold vs control.	[65]
	3-5 nm Pristine ND	Incubation and plating	20% bactericidal rate at 1 mg/mL vs control.	[39]
	3-5 nm cND suspension and embedded into epoxy	Incubation and plating	99.99% bactericidal rate, 80% bacterial biofilm inhibition at 1 mg/mL. 99.99% bactericidal rate of epoxy embedded with NDs vs control.	
	4 nm O-ND integrated into Poly-lactic acid scaffold	Incubation, Live/Dead staining and confocal microscope imaging	40% reduction in bacterial viability vs control, statistically significant.	[58]
	18-125 nm ND	Incubation & CFU count	Varying ND concentrations (1–500 μg/mL) and sizes either had no effect or increased CFA in water. Up to 90% reduction in CFA in PBS at higher concentrations (≤10μg/mL).	[45]
	Pristine ND	Incubation & CFU count	Three orders of magnitude (log CFU) reduction when coated with 1.6 mg ND/g matrix concentration, compared to control.	[66]
	ND-PVA composite	Broth microdilution	0.008 mg/mL NDs composite displayed lowest MIC at 4 μg/mL	[63]
Streptococcus mutans	4.2 ± 0.5 nm C-ND	Incubation & optical density measurement / CFU count	MIC: 4 μg/mL. Minimum Bactericidal concentration (MBC): 16 μg/mL.	[50]
	150nm O- ND	XTT assay, Live/Dead staining	Near 100% bactericidal rate at 2.5 mg/mL. 60–70% biofilm formation reduction at 0.15 mg/mL. 83.92% bactericidal rate in preformed biofilms at 2.5 mg/mL.	[64]

https://doi.org/10.2147/NSA.S575116

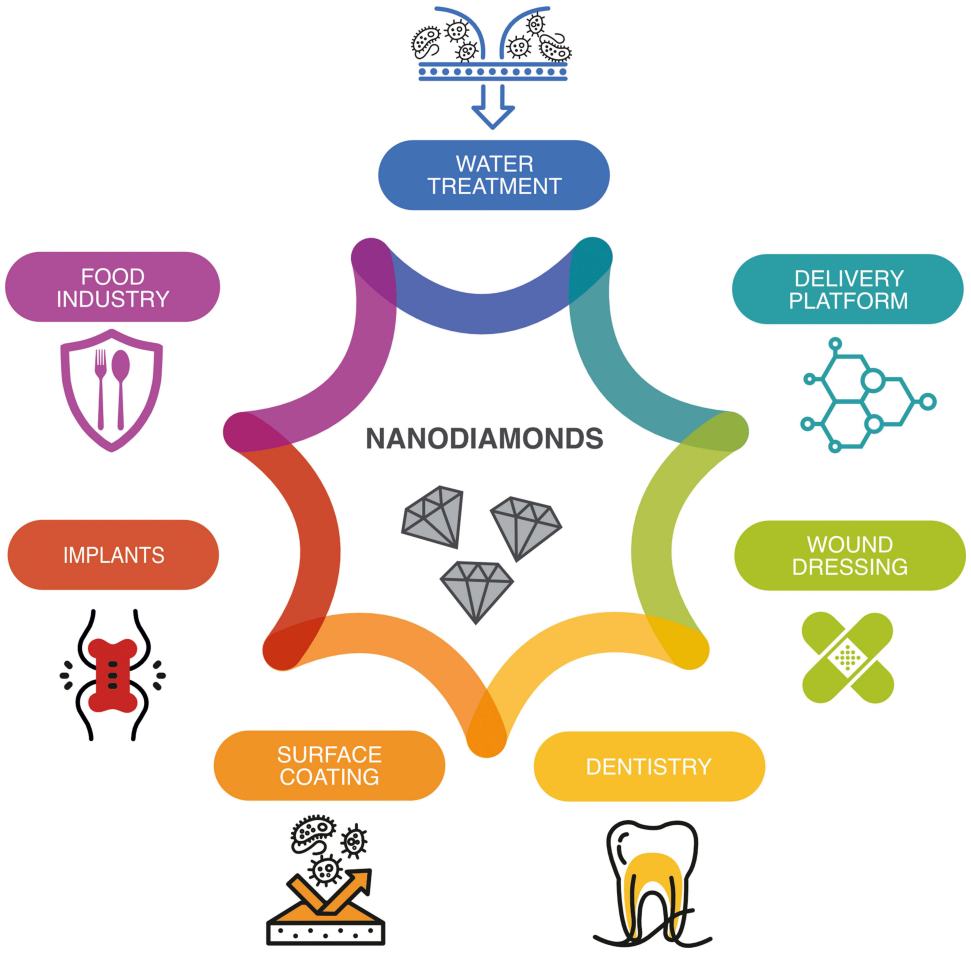


Figure 3 Antimicrobial applications of nanodiamonds.

Openda et al also explored various Pc-ND and Ag nanoconjugates to enhance PACT against *S. aureus* and *E. coli* biofilms. Across their studies, they demonstrated that positively charged Pc derivatives, functionalized with ND and AgNP, exhibited high singlet oxygen quantum yields and significantly suppressed bacterial biofilms.[35,37,38]

Chang et al explored the biomedical potential of NDs by demonstrating their ability to enhance the antibacterial properties of AgNP while improving biocompatibility. By supporting AgNP with NDs and conjugating them with albumin, the researchers developed a nanohybrid that exhibited sustained bactericidal effects against *E. coli* for over a month. Additionally, the nanohybrids were successfully integrated into natural polysaccharide-based hydrogels, showcasing their potential for wound dressings and medical devices.[41]

Iqbal et al also investigated the antibacterial properties of AgNP-ND composites, demonstrating their enhanced efficacy against *E. coli* and *S. aureus*. Using a microplasma technique to stabilize AgNP on NDs, the composite exhibited improved antimicrobial activity at lower silver concentrations, reducing toxicity concerns. NDs contributed structural stability while supporting the bactericidal effects of silver.[67]

Baron et al investigated the adsorption of viral particles from the blood plasma of patients with hepatitis B or C using NDs. Their study demonstrated that NDs significantly reduced viral load levels by 2–3 orders of magnitude through selective adsorption, suggesting their potential use in haemodialysis and plasmapheresis. This represents an atypical example compared to those described earlier, as it highlights that the same NDs properties that allow it to act as a carrier, allow NDs to bind and remove viral particles, indicating their potential interaction with viruses and opening avenues for antiviral treatment technologies.[68]

Wound Dressing

The antimicrobial properties of NDs are also being harnessed in wound care. Their incorporation into hydrogels, membranes, scaffolds and dressings could enable the prevention of bacterial infections in wounds, supporting tissue regeneration.

Khalid et al developed a multifunctional ND-silk fibroin membrane via electrospinning, for wound healing and biosensing. The inclusion of NDs (~5 nm) with negatively charged nitrogen vacancy centres enabled optically detected magnetic resonance-based temperature sensing for non-invasive infection monitoring. The membrane showed enhanced thermal stability, preserved fluorescence, and was biocompatible in a murine model, with wound healing comparable to controls. Notably, it exhibited selective antibacterial activity against Gram-negative *Pseudomonas aeruginosa* and *E. coli*, while leaving Gram-positive *S. aureus* unaffected.[69]

Houshyar et al investigated the incorporation of NDs into both poly ε-caprolactone (PCL) nanofibrous scaffolds for wound management and polypropylene (PP) surgical meshes for enhanced functionality. The integration improved thermal stability, surface energy, and hydrophilicity, thereby promoting epithelial cell proliferation and reducing *S. aureus* adhesion. NDs embedded in PP meshes introduced temperature sensing capabilities via nitrogen-vacancy centres, enabling optical assessment of infection or inflammation.[65,70]

Park et al developed ND-polydopamine (ND@PDA) NPs to enhance antibacterial and wound healing properties in electrospun nanofiber scaffolds. ND@PDA improved photothermal and oxidative stress responses under NIR light, disrupting bacterial biofilms and membranes. Catechol groups in PDA also promoted cell adhesion and biocompatibility, supporting their potential in targeted wound therapy.[71]

Wu et al developed melt electro-written poly-lactic acid/ND (PLA/ND) scaffolds for wound healing with cell delivery capability. ND incorporation enhanced PLA's thermal stability, fibroblast proliferation, and extracellular matrix remodelling. The scaffolds were effectively coated with quaternized β-chitin (QβC), boosting antibacterial activity and human cell adhesion. The final PLA/ND-QβC constructs showed strong potential for infection prevention and accelerated healing in chronic wounds.[58]

Dentistry

In dentistry, NDs are increasingly recognized for their dual functions in mechanical reinforcement and antimicrobial activity.

Quan et al demonstrated that C-NDs effectively inhibit *S. mutans* growth, while Zhang et al showed that NDs suppress biofilm formation and disrupt established biofilms of *S. mutans, Porphyromonas gingivalis, Candida albicans*, and *Candida glabrata*, common oral pathogens.[50,64]

Fang et al demonstrated that O-NDs act as peroxidase mimics, catalyzing hydrogen peroxide to produce free radicals that disrupt bacterial membranes and biofilms. In vivo, topical O-ND rinses significantly reduced bacterial infection and promoted wound healing in a rodent periodontal model.[18]

Cao et al developed silver-loaded, polycation-functionalized NDs (Ag/QNDs) to enhance the mechanical and antibacterial properties of resin-based dental materials. Ag/QNDs improved hardness, flexural strength, and modulus through homogeneous dispersion and strong resin bonding. Their dual antibacterial action, from Ag^+ and cationic polymers, effectively inhibited *S. mutans* at 1.0 wt% without notable macrophage cytotoxicity.[42]

Rifai et al investigated ND coatings on selective laser melted titanium (SLM-Ti) scaffolds. Their study found that ND-coated surfaces increased human dermal fibroblast and osteoblast cell densities by 32% and 29%, respectively, after three days of incubation. Additionally, ND coatings reduced *S. aureus* adhesion by 88%.[72]

Surface Coating

NDs are also being considered for surface coatings that resist microbial colonization. Their use in sol-gel coatings and composite films has shown promise for inhibiting microbial growth on medical devices, hospital surfaces, and other high-contact environments. The effectiveness of such coatings is largely attributed to electrostatic interactions and surface hydrophobicity, which reduce bacterial attachment and biofilm formation.

Budil et al investigated the anti-adhesive properties of 5 nm NDs films against *E. coli*. Their findings indicate that H- and F-terminated ND films reduced bacterial adhesion by approximately 50% in mineral medium (M9).[55]

Streletskiy et al developed amorphous carbon films embedded with well-dispersed NDs using magnetron sputtering, aiming to enhance antimicrobial properties. The films inhibited growth of *E. coli, S. aureus, B. subtilis, C. albicans, Aspergillus niger* and *Aspergillus fumigatus*. The incorporation of plasmonic AgNP further improved surface-enhanced Raman scattering (SERS) and luminescence properties, enabling advanced structural analysis.[73]

Gutierrez et al examined the antibacterial properties of diamond-like carbon (DLC) films doped with NDs and demonstrated that these films achieved up to 95% bacterial inhibition under direct contact, comparable to the antibiotic streptomycin.[13]

Ruzek et al investigated ND additives in sol-gel coatings, emphasizing their chemical inertness, biocompatibility, and environmental safety. The study demonstrated that NDs dispersed well within the coating matrix and enhanced antimicrobial activity, particularly through synergy with metal ions, while maintaining cytocompatibility and non-toxicity toward human fibroblast cultures.[74]

Li et al developed cNDs combined with epoxy to create antibacterial, wear-resistant surfaces. The positively charged cNDs disrupt bacterial membranes via electrostatic interactions, achieving 99.99% bactericidal efficacy and over 80% biofilm inhibition at 1 mg/mL. The antibacterial activity remained stable after heat, acid, and abrasion treatments.[39]

Uzoma et al developed a superhydrophobic ND composite coating with antibacterial, icephobic, and corrosion-resistant properties. The coating reduced *E. coli* adhesion by 73% after 24 hours, demonstrating its effectiveness in preventing bacterial contamination.[62]

Zhang et al investigated the antibacterial properties of diamond coatings with different surface morphologies: ultrananocrystalline (UNCD), nanocrystalline (NCD), and microcrystalline diamond (MCD). MCD coatings showed the highest bactericidal activity, particularly against *E. coli* and *B. subtilis*, attributed to their rough texture and oxygen-containing functional groups.[75]

Nunes-Pereira et al explore the antimicrobial potential of NDs incorporated into polyvinylidene fluoride (PVDF) composites, highlighting their ability to enhance optical, thermal, and electrical properties while maintaining biocompatibility. Their findings suggest that functionalized NDs, particularly amine-modified variants, exhibit strong bacteriostatic effects against *E. coli*.[76]

Dunseath et al applied black diamond (bD) coatings, seeded with 4 nm NDs, to 5 μm silicon nanoneedles to investigate dual-mode antibacterial mechanisms. The sharp nanoscale topography mechanically disrupted motile bacteria like *E. coli*, while surface functionalization (–H, –O, $–NH_2$, –F) enhanced bactericidal activity by around 20–30%.[54]

Implants

In the context of biomedical implants, NDs are valued not only for their antimicrobial action but also for their mechanical stability and tissue compatibility.

Chernysheva et al investigated NDs in antimicrobial coatings for prosthetic heart valves, highlighting their role in improving infection resistance and structural integrity. Incorporation of lysozyme and miramistin yielded composites with strong activity against *S. aureus*. In a follow-up study, ND-drug conjugates with amikacin or levofloxacin formed stable coatings on collagen-rich matrices, with amikacin showing superior retention and antibacterial efficacy.[27,66]

Krok et al investigated the modification of titanium implants using biofunctional NDs to enhance antimicrobial properties. NDs conjugated with antibiotics such as amoxicillin or ampicillin, effectively inhibited bacterial growth.[57]

Saha et al functionalized NDs to enhance the antimicrobial and biocompatibility properties of polypropylene hernia mesh. Hydroxyl-functionalized NDs in a chitosan coating improved hydrophilicity, fibroblast attachment, and antibacterial activity against *E. coli*. Optimized curing ensured stable ND integration, preserving mechanical strength while promoting healing and reducing infection risk.[77]

Wang et al developed a biodegradable polyurethane scaffold reinforced with silver-loaded polycationic NDs for cartilage tissue repair. ND incorporation enhanced crystallinity, which improved mechanical strength while preserving flexibility. The scaffold exhibited dual antibacterial activity, with contact-killing mediated by cationic polymers and release-killing provided by silver NPs, effectively inhibiting *S. aureus*. Its controlled degradation rate and low cytotoxicity further support its potential for biomedical applications.[40]

Food Industry

The potential of NDs extends into the food industry, where their antimicrobial characteristics could be used to enhance food safety, as active components in food packaging materials.

Deflorio et al investigated the incorporation of a coating of polydopamine, NDs, and an alkyl silane onto a polyvinyl chloride (PVC) food-contact surface, which is commonly used in the food industry for processing and storage purposes. The ND-containing coating significantly reduced *E. coli* and *Salmonella enterica* adhesion, mainly due to the coating's surface-wetting properties.[78]

Iqbal et al incorporated NDs into polyvinyl alcohol (PVA) composites to enhance its thermal, mechanical, optical, and antibacterial properties for food packaging applications. NDs improved hydrogen bonding within the matrix, resulting in increased tensile strength, ductility, thermal stability, and UV resistance. The composite also exhibited antibacterial activity against *E. coli* and *S. aureus* through membrane disruption.[63]

Liu et al developed a durable, superhydrophobic ND coating for aluminum surfaces to improve food hygiene. Sequential deposition of crystalline NDs, self-assembled L-dopa, and organofluorosilane modification led to over 99% reduction in *E. coli* and *S. aureus* adhesion. The coating also demonstrated strong mechanical durability, highlighting its potential for food-contact surface applications.[79]

Water Treatment

Environmental and water treatment applications of NDs are also under growing investigation. Their antimicrobial and adsorptive properties suggest potential use in filtration systems to remove microbial contaminants while enhancing membrane durability and efficiency. In water treatment, it is critical to engineer surfaces that attract and inactivate bacteria upon contact. This antibacterial effect is primarily governed by electrostatic interactions and surface hydrophobicity, which facilitate bacterial capture and membrane disruption.

Colon et al incorporated NDs into filtration membranes, resulting in improved plastic and elastic properties. The modified membranes also exhibited enhanced microbial removal and bactericidal performance, highlighting the potential of NDs to advance current filtration technologies.[17]

Karami et al grafted amine-functionalized NDs onto polyamide thin-film composite membranes to mitigate fouling, a major challenge in water treatment. The ND modification enhanced membrane flux and significantly increased antibacterial activity, resulting in higher *E. coli* inactivation and mortality rates.[61]

Biological Considerations

This section reviews the cellular and organism-level responses to NDs, focusing on how their physicochemical properties, including size, surface chemistry, and charge, affect biocompatibility and cytotoxicity in different biological systems. NDs, varying in size and surface chemistry, exhibit diverse biological effects depending on their physical properties and the cell types studied. NDs sized 2–10 nm, functionalized with carboxyl, sodium carboxylate, or sulfonate groups, demonstrate excellent in vitro biocompatibility, showing low cytotoxicity and high cell viability across multiple cell types (neuroblastoma, macrophages, keratinocytes, PC-12), even at concentrations up to 100 μg/mL. Tested NDs did not induce higher levels of ROS than those observed in untreated controls and support normal cellular morphology, adhesion, and growth.[80] Broz et al assessed the response of SAOS-2 osteoblast-like cells to NDs ranging from 18 to 210 nm. While low concentrations (10–100 μg/mL) had minimal effects, high concentrations (1000 μg/mL) reduced viability due to physical obstruction from agglomerates. Smaller NDs (5 nm) showed higher cellular uptake and pronounced cytotoxicity. Surface charge significantly influenced cellular response: negatively charged O-NDs (–40 mV) reduced cytotoxicity by 25–30%, whereas positively charged hydrogenated NDs (H-NDs) (around +40 mV) induced up to 85% cell death within three days.[81] These results are consistent with Tylor et al who found that O-NDs enhanced human neural stem cell (hNSC) adhesion and proliferation, whereas H-NDs inhibited attachment, causing neurosphere formation. The hydrophilic, negatively charged surface of O-NDs was more conducive to hNSC growth compared to the hydrophobic, positively charged H-NDs.[82] Woodhams et al compared O-NDs and pristine NDs, with an average size of approximately 8 nm, in breast cancer cell lines

MDA-MB-231 and MCF-7. O-NDs were better internalized and less cytotoxic, while pristine NDs induced oxidative stress and slightly reduced proliferation, particularly in MDA-MB-231 cells.[83]

Despite overall favourable biocompatibility, certain cell types display heightened sensitivity to NDs. Knötigová et al reported that 100 nm fluorescent C-NDs interact extensively with THP-1 monocytes, entering via macropinocytosis and membrane disruption. These particles accumulated in lysosomes, triggered lysosomal destabilization, cathepsin B release, and NLRP3 inflammasome activation, alongside increased ROS generation and modulation of pro-inflammatory and angiogenic mediators (eg, IL-6, ANGPT2, TIE-2).[84] Wierzbicki et al reported that pristine NDs (2–10 nm) had minimal impact on the proliferation of human mammary epithelial cells (HMECs) and fibroblast-like stromal cells (HS-5) but significantly affected human endothelial cells (HUVECs) even at the lowest concentrations, inducing cell death, morphological changes, and intracellular ND accumulation. In culture medium, NDs formed large agglomerates (up to 375 nm), with aggregation further increased in the presence of 10% FBS, likely due to the formation of an extensive protein corona. Although FBS reduced ND toxicity at lower concentrations, toxicity remained high at doses above 5 mg/L. Functionalization of NDs with an RGD-containing peptide reduced toxicity in HUVECs at low concentrations but again, was ineffective at higher doses. These findings suggest that while protein and peptide coronas can attenuate NDs toxicity, their protective effect is limited in sensitive cell types.[85]

Wehling et al demonstrated that among six types of NDs (2–10 nm), the highest antibacterial activity was observed in negatively charged, partially oxidized NDs. However, this effect was neutralized by the presence of proteins and other medium components.[52] Bacterial cells are more susceptible to NDs due to their more negatively charged membranes compared to mammalian cells, which promotes stronger electrostatic interactions with positively charged particles.[10] C-NDs disrupt bacterial membranes through superficial interactions, while mammalian cells such as SAOS-2 osteoblast-like cells internalize them with low cytotoxicity. At low to moderate concentrations, C-NDs have minimal effects on cell viability. However, high concentrations reduce viability primarily through mechanical obstruction by agglomerates rather than chemical toxicity.[46]

In vivo studies complement in vitro findings by explaining the biodistribution and systemic effects of NDs. These investigations consistently demonstrate low toxicity and favourable biocompatibility across various biological systems.[86] For instance, Yuan et al observed minimal pulmonary toxicity in mice after intratracheal instillation of 4 nm and 50 nm NDs, attributing clearance to alveolar macrophage-mediated transport via the mucociliary escalator over a 28-day period.[87] Similarly, intracranial injection of 100 nm fluorescent NDs in rats caused no behavioural or physiological impairments, with preserved cognitive function confirmed via the Novel Object Recognition Test.[88] Biodistribution studies with radio-labelled amino-functionalized NDs revealed initial accumulation in the lungs, spleen, and liver, followed by renal clearance. Notably, filtration to reduce particle size significantly decreased organ retention, whereas unfiltered NDs exhibited greater pulmonary and splenic uptake due to aggregation and size-dependent phagocytosis.[89]

Despite promising preclinical outcomes, clinical research on NDs remains limited. To date, only one registered clinical trial has evaluated ND-based applications, focusing on root canal therapy. In this study, 4–5 nm NDs were incorporated into melted commercial gutta-percha at 60 °C, forming a homogeneous nanocomposite. The resulting ND-gutta-percha exhibited enhanced antimicrobial activity, improved mechanical strength, and increased radiopacity, thereby improving the durability and radiographic detectability of the root canal filling material.[90,91]

NDs exhibit highly variable biological effects that depend on their physicochemical properties and the target organism. These findings emphasize the need for precise material engineering and continued research to bridge the gap between preclinical promise and clinical applicability.

Conclusions

Considering the evidence presented in this review, NDs exhibit multifactorial antibacterial activity, driven by their size, surface chemistry, and charge. Their mechanisms of action include membrane disruption, interference with nutrient transport, electrostatic destabilization, and structural damage to membrane-associated proteins. The antibacterial efficacy of NDs is strongly influenced by their surface functionalisation. Functional elements that promote membrane interactions, such as carboxyl moieties and other charged chemical groups, enhance their antimicrobial activity. Moreover, surface functionalisation enables conjugation with additional antibacterial agents, including antibiotics, polycations, antimicrobial

peptides, and metallic nanoparticles or ions. In addition to antibiotic-functionalised NDs, polycation-functionalised NDs exhibit some of the highest antibacterial activity reported, driven by strong electrostatic interactions that disrupt bacterial membranes. When considering bacterial defence mechanisms against NDs, the structural organisation of the bacterial envelope is a key determinant of whether a given ND type can exert antibacterial activity. Smaller NDs (around 5 nm) can penetrate the outer membrane of Gram-negative bacteria and induce cell lysis through structural alterations of membrane proteins, while also impairing other processes by blocking porins. In contrast, larger NDs (around 100 nm) interact more efficiently with the peptidoglycan layer of Gram-positive species, thereby restricting nutrient diffusion across the membrane. It is essential to account for this aspect when designing antibacterial experiments. For Gram-positive species, larger NDs should represent the primary focus, with smaller NDs included only as controls or comparative references. Conversely, for Gram-negative species, the main tests should involve smaller NDs, whereas larger NDs may serve as controls. Selecting the appropriate ND size is therefore a matter of choosing material that is fit for purpose.

The safety profile of NDs is highly context dependent. Although numerous studies report excellent biocompatibility, the outcomes vary markedly with particle size and surface chemistry. A key factor is the presence of proteins and other organic molecules in the surrounding environment, which can assemble into a biomolecular corona on the ND surface. This layer may mask reactive surface functionalities, thereby diminishing both cytotoxic and antibacterial effects. Protein adsorption can also stabilize ND suspensions or facilitate the formation of antimicrobial ND-protein complexes. These dual effects highlight the complex and dynamic nature of ND-biological interactions.

They also raise important questions about the applicability of NDs under in vivo conditions, with both host- and bacterium-derived proteins present. This highlights the need to simulate clinically relevant environments when assessing antibacterial activity or biocompatibility. The observed duality further suggests that NDs reported as biocompatible may owe this property to the protein corona acquired in biological media, which could simultaneously suppress their antibacterial activity by masking reactive surface groups. This issue also introduces concerns about potential drug interactions, as therapeutic molecules may bind to the same reactive sites on NDs, thereby altering their pharmacokinetic and pharmacodynamic profiles. Such considerations will need to be addressed in future studies if NDs are to progress toward clinical application.

The key takeaway of this article is that due to the diversity of experimental conditions, including differences in testing media composition, ND size and functionalization, and microbial species, it is challenging to directly compare results between currently available studies. Therefore, it is crucial that one should take all these factors into consideration when interpreting reported findings. This article should aid the readers in understanding the importance of such interactions when conducting their own research, and most importantly, when reporting their own findings.

Moving forward, the field would benefit from standardised protocols for evaluating NDs of different sizes under both protein-free and physiologically relevant conditions, enabling a more accurate assessment of their intrinsic cytotoxic and antimicrobial activities. Progress is further limited by the narrow range of bacterial species commonly used to characterise ND antibacterial properties, as most studies focus predominantly on *E. coli* and *S. aureus*. To fully realise the therapeutic potential of NDs, future studies should include more clinically relevant strains, such as those from the ESKAPE group or listed among the WHO priority pathogens.

Abbreviations

ND, nanodiamond; NP, nanoparticle; DND, detonation nanodiamond; HPHT, high-pressure high-temperature; O-NDs, oxidated nanodiamonds; FTIR, Fourier transform infrared spectroscopy; EDS, energy-dispersive X-ray spectroscopy; C-NDs, carboxylated nanodiamonds; PEI, polyethylenimine; SPPS, solid-phase peptide synthesis; MALDI-TOF, matrix-assisted laser desorption/ionization time-of-flight; SEM, scanning electron microscopy; BSA, bovine serum albumin; Pc, phthalocyanines; ROS, reactive oxygen species; PACT, photodynamic antimicrobial therapy; DMF, dimetyloformamid; DMSO, dimethyl sulfoxide; DCC/NHS, N,N′-dicyclohexylcarbodiimide/N-hydroxysuccinimide; AgNP, silver nanoparticle; TEM, transmission electron microscopy; cND, cationic nanodiamond; MES, 2-(N-morpholino)ethanesulfonic acid; EDC/NHS, 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide/ N-Hydroxysuccinimide; XPS, X-ray photoelectron spectroscopy; QND, quaternary ammonium-modified nanodiamond; LPS, lipopolysaccharides; DMEM, dulbecco's modified eagle medium; FBS, fetal bovine serum; PCL, poly ε-caprolactone; PP, polypropylene; PDA, polydopamine; PLA, poly-lactic acid;

QβC, quaternized β-chitin; SLM-Ti, selective laser melted titanium; M9, mineral medium; SERS, surface-enhanced raman scattering; DLC, diamond-like carbon; UNCD, ultrananocrystalline diamond; NCD, nanocrystalline diamond; MCD, microcrystalline diamond; PVDF, polyvinylidene fluoride; bD, black diamond; PVC, polyvinyl chloride; PVA, polyvinyl alcohol; CFU, colony forming unit; MH, Mueller-Hinton; LH, Luria-Bertani; FIB, focused ion beam; CFA, colony forming ability; MIC, minimum inhibitory concentration; H-ND, hydrogenated nanodiamond; hNSC, human neural stem cell; HMEC, human mammary epithelial cell; HS-5, fibroblast-like stromal cell; HUVEC, human endothelial cell.

Funding

This work was funded by the National Science Centre of Poland grant 2023/51/D/NZ7/01557.

Disclosure

The authors declare that they have no competing interests in this work.

References

1. Zhao N, Song M, Zhang X, Xu W, Liu X. Nanodiamond coating in energy and engineering fields: synthesis methods, characteristics, and applications. *Small*. 2024;20(36). doi:10.1002/smll.202401292
2. Mochalin VN, Shenderova O, Ho D, Gogotsi Y. The properties and applications of nanodiamonds. *Nat Nanotechnol*. 2011;7(1):11–23. doi:10.1038/nnano.2011.209
3. Khan MB, Khan ZH. Nanodiamonds: synthesis and Applications. In: Khan ZH editor. *Nanomaterials and Their Applications*. Springer Singapore; 2018: 1–26. doi:10.1007/978-981-10-6214-8_1
4. World Health Organization. *Global Research Agenda for Antimicrobial Resistance in Human Health*; 2025.
5. Blackman LD, Sutherland TD, De Barro PJ, Thissen H, Locock KES. Addressing a future pandemic: how can non-biological complex drugs prepare us for antimicrobial resistance threats? *Mater Horiz*. 2022;9(8):2076–2096. doi:10.1039/d2mh00254j
6. Scoffone VC, Barbieri G, Irudal S, Trespidi G, Buroni S. New antimicrobial strategies to treat multi-drug resistant infections caused by gram-negatives in cystic fibrosis. *Antibiotics*. 2024;13(1):71. doi:10.3390/antibiotics13010071
7. Park H, Patil TV, Mo C, Lim KT. Nanodiamond: a multifaceted exploration of electrospun nanofibers for antibacterial and wound healing applications. *J Nanobiotechnol*. 2025;23(1). doi:10.1186/s12951-025-03351-9
8. Rajak BL, Kumar R, Gogoi M, Patra S. Antimicrobial Activity of Nanomaterials. *Antimicro Activ Nanomater*. 2020;147–185. doi:10.1007/978-3-030-29207-2_5
9. Barzegar Amiri Olia M, Donnelly PS, Hollenberg LCL, Mulvaney P, Simpson DA. Advances in the surface functionalization of nanodiamonds for biological applications: a review. *ACS Appl Nano Mater*. 2021;4(10):9985–10005. doi:10.1021/acsanm.1c02698
10. Isakova AA, Safonov AV, Alexandrovskaya A, Galushko TB, Indenbom AV, Spitsyn BV. The effect of nanodiamond surface modification on interaction with Pseudomonas putida K12. *Prot Met Phys Chem Surf*. 2017;53(2):220–223. doi:10.1134/S2070205117020137
11. Beranová J, Seydlová G, Kozak H, et al. Sensitivity of bacteria to diamond nanoparticles of various size differs in gram-positive and gram-negative cells. *FEMS Microbiol Lett*. 2014;351(2):179–186. doi:10.1111/1574-6968.12373
12. Cumont A, Pitt AR, Lambert PA, Oggioni MR, Ye H. Properties, mechanism and applications of diamond as an antibacterial material. *Functional Diamond*. 2021;1(1):1–28. doi:10.1080/26941112.2020.1869434
13. Conceição K, de Andrade VM, Trava-Airoldi VJ, Capote G, Capote G. High antibacterial properties of DLC film doped with nanodiamond. *Surf Coat Technol*. 2019;375:395–401. doi:10.1016/j.surfcoat.2019.07.029
14. Chatterjee A, Perevedentseva E, Jani M, et al. Antibacterial effect of ultrafine nanodiamond against gram-negative bacteria Escherichia coli. *J Biomed Opt*. 2014;20(5):051014. doi:10.1117/1.jbo.20.5.051014
15. Lazovic J, Goering E, Wild AM, et al. Nanodiamond-enhanced magnetic resonance imaging. *Adv Mater*. 2024;36(11). doi:10.1002/adma.202310109
16. Mochalin V, Osswald S, Gogotsi Y. Contribution of functional groups to the Raman spectrum of nanodiamond powders. *Chem Mater*. 2009;21(2):273–279. doi:10.1021/cm802057q
17. Colon A, Avalos J, Weiner BR, Morell G, Ríos R. Water treatment membranes embedded with a stable and bactericidal nanodiamond material. *J Water Health*. 2023;21(5):601–614. doi:10.2166/wh.2023.298
18. Fang J, Wang H, Bao X, et al. Nanodiamond as efficient peroxidase mimic against periodontal bacterial infection. *Carbon N Y*. 2020;169:370–381. doi:10.1016/j.carbon.2020.07.055
19. Lei Y, Zheng ML, Zhao ZS, Duan XM. Carboxylated nanodiamond: aggregation properties in aqueous dispersion system and application in living cell fluorescence imaging. *J Nanosci Nanotechnol*. 2016;16(3):2319–2324. doi:10.1166/jnn.2016.10932
20. Astuti Y, Saputra FD, Wuning S, Bhaduri G. Enrichment of nanodiamond surfaces with carboxyl groups for doxorubicin loading and release. In: *IOP Conference Series: Materials Science and Engineering*. 172. Institute of Physics Publishing; 2017. doi:10.1088/1757-899X/172/1/012066
21. Garg S, Garg A, Sahu NK, Yadav AK. Synthesis and characterization of nanodiamond-anticancer drug conjugates for tumor targeting. *Diam Relat Mater*. 2019;94:172–185. doi:10.1016/j.diamond.2019.03.008
22. Basiuk EV, Santamaría-Bonfil A, Meza-Laguna V, et al. Solvent-free covalent functionalization of nanodiamond with amines. *Appl Surf Sci*. 2013;275:324–334. doi:10.1016/j.apsusc.2012.12.062
23. Basiuk EV, Monroy-Torres B, Carrero JC, Basiuk VA. Effects of solvent-free amine functionalization of graphene oxide and nanodiamond on bacterial growth. *Fuller Nanotub Car Nanostruct*. 2021;29(1):58–66. doi:10.1080/1536383X.2020.1811235

24. Burleson T, Yusuf N, Stanishevsky A. Surface modification of nanodiamonds for biomedical application and analysis by infrared spectroscopy. *J Achieve Mater Manuf Eng*. 2009;39:258–63.
25. Shen T, Chernysheva MG, Badun GA, et al. Levofloxacin and amikacin adsorption on nanodiamonds: mechanism and application prospects. *Colloids Interfaces*. 2022;6(2):35. doi:10.3390/colloids6020035
26. Shen T, Chernysheva MG, Popov AG, Chaschin IS, Anuchina NM, Badun GA. Adsorption complexes of vancomycin with nanodiamonds: complexation kinetics, composition, and antimicrobial properties. *Russ J Phys Chem A*. 2024;98(13):3165–3171. doi:10.1134/S0036024424702406
27. Chernysheva MG, Shen T, Chaschin IS, et al. Nanodiamond–drug conjugates for coating xenogenic heart valve prostheses. *Mendeleev Commun*. 2024;34(1):104–106. doi:10.1016/j.mencom.2024.01.031
28. Shen T, Chernysheva MG, Badun GA. Tritium labeling vancomycin and studying its adsorption on nanodiamonds. *Radiochemistry*. 2023;65 (6):690–698. doi:10.1134/S1066362223060103
29. Shen Y, Li Y, Wen Y, et al. Investigations into the antimicrobial behavior and cytotoxicity of purified detonation diamond nanoparticles and their assembled micro-/nano-structured films. *Mater Lett*. 2024:363. doi:10.1016/j.matlet.2024.136294
30. Rouhani P, Govindaraju N, Iyer JK, Kaul R, Kaul A, Singh RN. Purification and functionalization of nanodiamond to serve as a platform for amoxicillin delivery. *Mater Sci Eng C*. 2016;63:323–332. doi:10.1016/j.msec.2016.02.075
31. Daskalova E, Zheleva N, Belouhova M, et al. Antibacterial activity of combined nanodiamonds and snail fractions with biocompounds with MW below 10 KDA and above 30 KDA. *Comptes Rendus de L'Academie Bulgare des Sciences*. 2023;76(1):35–43. doi:10.7546/CRABS.2023.01.04
32. Knapinska AM, Tokmina-Roszyk D, Amar S, et al. Solid-phase synthesis, characterization, and cellular activities of collagen-model nanodiamond-peptide conjugates. *Biopolymers*. 2015;104(3):186–195. doi:10.1002/bip.22636
33. Mogil'naya OA, Bondar VS. Comparative study of antibacterial properties of Lysozyme upon its adsorption and covalent binding to nanodiamonds. *Nanotechnol Russ*. 2012;7(11–12):658–665. doi:10.1134/S1995078012060080
34. Lin CL, Lin CH, Chang HC, Su MC. Protein attachment on nanodiamonds. *J Phys Chem A*. 2015;119(28):7704–7711. doi:10.1021/acs.jpca.5b01031
35. Openda YI, Nyokong T. Enhanced photo-ablation effect of positively charged phthalocyanines-detonation nanodiamonds nanoplatforms for the suppression of Staphylococcus aureus and Escherichia coli planktonic cells and biofilms. *J Photochem Photobiol A*. 2021;411. doi:10.1016/j.jphotochem.2021.113200
36. Gvozdev DA, Gudkova VR, Moisenovich AM, Ramonova AA, Strakhovskaya MG, Maksimov EG. Detonation nanodiamonds as enhancers of E. coli photodynamic inactivation by phthalocyanines in a high molarity buffer solution. *J Photochem Photobiol B*. 2022;227. doi:10.1016/j.jphotobiol.2022.112387
37. Openda YI, Ngoy BP, Nyokong T. Photodynamic antimicrobial action of asymmetrical porphyrins functionalized silver-detonation nanodiamonds nanoplatforms for the suppression of staphylococcus aureus planktonic cells and biofilms. *Front Chem*. 2021;9. doi:10.3389/fchem.2021.628316
38. Openda YI, Matshitse R, Nyokong T. A search for enhanced photodynamic activity against: staphylococcus aureus planktonic cells and biofilms: the evaluation of phthalocyanine-detonation nanodiamond-Ag nanoconjugates. *Photochem Photobiol Sci*. 2020;19(10):1442–1454. doi:10.1039/d0pp00075b
39. Li FK, Zhao WB, Wang Y, et al. Cationic engineered nanodiamonds for efficient antibacterial surface with strong wear resistance. *Nano Res*. 2024;17(3):939–948. doi:10.1007/s12274-024-6417-8
40. Wang L, Cao W, Wang X, et al. Biodegradable silver-loaded polycation modified nanodiamonds/polyurethane scaffold with improved antibacterial and mechanical properties for cartilage tissue repairing. *J Mater Sci Mater Med*. 2019;30(4). doi:10.1007/s10856-019-6244-8
41. Chang BM, Pan L, Lin HH, Chang HC. Nanodiamond-supported silver nanoparticles as potent and safe antibacterial agents. *Sci Rep*. 2019;9(1). doi:10.1038/s41598-019-49675-z
42. Cao W, Wang X, Li Q, Ye Z, Xing X. Mechanical property and antibacterial activity of silver-loaded polycation functionalized nanodiamonds for use in resin-based dental material formulations. *Mater Lett*. 2018;220:104–107. doi:10.1016/j.matlet.2018.03.027
43. Silhavy TJ, Kahne D, Walker S. The bacterial cell envelope. *Cold Spring Harb Perspect Biol*. 2010;2(5):a000414–a000414. doi:10.1101/cshperspect.a000414
44. Norouzi N, Ong Y, Damle VG, Habibi Najafi MB, Schirhagl R. Effect of medium and aggregation on antibacterial activity of nanodiamonds. *Mater Sci Eng C*. 2020;112. doi:10.1016/j.msec.2020.110930
45. Ong SY, Van Harmelen RJJ, Norouzi N, et al. Interaction of nanodiamonds with bacteria. *Nanoscale*. 2018;10(36):17117–17124. doi:10.1039/c8nr05183f
46. Sousa GES, Martins BA, Mendes de Almeida Junior A, et al. Determining molecular-level interactions of carboxyl-functionalized nanodiamonds with bacterial membrane models as the basis for antimicrobial activity. *Langmuir*. 2025;41(9):6186–6196. doi:10.1021/acs.langmuir.4c05173
47. Beranová J, Seydlová G, Kozak H, Potocký Š, Konopásek I, Kromka A. Antibacterial behavior of diamond nanoparticles against Escherichia coli. *Phys Stat Solidi B Res*. 2012;249(12):2581–2584. doi:10.1002/pssb.201200079
48. Sawosz E, Chwalibog A, Mitura K, et al. Visualisation of morphological interaction of diamond and silver nanoparticles with salmonella enteritidis and listeria monocytogenes. *J Nanosci Nanotechnol*. 2011;11:7635–7641. doi:10.1166/jnn.2011.4735
49. Chwalibog A, Sawosz E, Hotowy A, et al. Visualization of interaction between inorganic nanoparticles and bacteria or fungi. *Int J Nanomed*. 2010;5(1):1085–1094. doi:10.2147/IJN.S13532
50. Quan C, Lin H, Xiao H, Zhao J. Inhibitory effect of carboxylated nanodiamond on oral pathogenic bacteria Streptococcus mutans. *J Clin Lab Anal*. 2021;35(8). doi:10.1002/jcla.23872
51. Moskvitina E, Kuznetsov V, Moseenkov S, Serkova A, Zavorin A. Antibacterial effect of carbon nanomaterials: nanotubes, carbon nanofibers, nanodiamonds, and onion-like carbon. *Materials*. 2023;16(3):957. doi:10.3390/ma16030957
52. Wehling J, Dringen R, Zare RN, Maas M, Rezwan K. Bactericidal activity of partially oxidized nanodiamonds. *ACS Nano*. 2014;8(6):6475–6483. doi:10.1021/nn502230m
53. Yang Y, Zhang Y, Wang L, et al. Antibacterial property of oxygen-terminated carbon bonds. *Adv Funct Mater*. 2022;32(24). doi:10.1002/adfm.202200447
54. Dunseath O, Smith EJW, Al-Jeda T, et al. Studies of black diamond as an antibacterial surface for gram negative bacteria: the interplay between chemical and mechanical bactericidal activity. *Sci Rep*. 2019;9(1). doi:10.1038/s41598-019-45280-2
55. Budil J, Matyska Lišková P, Artemenko A, et al. Anti-adhesive properties of nanocrystalline diamond films against Escherichia coli bacterium: influence of surface termination and cultivation medium. *Diam Relat Mater*. 2018;83:87–93. doi:10.1016/j.diamond.2018.02.001

56. Mitura K, Kornacka J, Niemiec-Cyganek A, et al. The influence of diamond nanoparticles on fibroblast cell line L929, cytotoxicity and bacteriostaticity of selected pathogens. *Coatings*. 2022;12(2):280. doi:10.3390/coatings12020280
57. Krok E, Balakin S, Jung J, Gross F, Opitz J, Cuniberti G. Modification of titanium implants using biofunctional nanodiamonds for enhanced antimicrobial properties. *Nanotechnology*. 2020;31(20):205603. doi:10.1088/1361-6528/ab6d9b
58. Wu X, Li W, Herlah L, et al. Melt electrowritten poly-lactic acid /nanodiamond scaffolds towards wound-healing patches. *Mater Today Bio*. 2024:26. doi:10.1016/j.mtbio.2024.101112
59. Jira J, Rezek B, Kriha V, et al. Inhibition of E. coli growth by nanodiamond and graphene oxide enhanced by luria-bertani medium. *Nanomaterials*. 2018;8(3):140. doi:10.3390/nano8030140
60. Vassallo J, Besinis A, Boden R, Handy RD. The minimum inhibitory concentration (MIC) assay with Escherichia coli: an early tier in the environmental hazard assessment of nanomaterials? *Ecotoxicol Environ Saf*. 2018;162:633–646. doi:10.1016/j.ecoenv.2018.06.085
61. Karami P, Aktij SA, Khorshidi B, et al. Nanodiamond-decorated thin film composite membranes with antifouling and antibacterial properties. *Desalination*. 2022:522. doi:10.1016/j.desal.2021.115436
62. Uzoma PC, Wang Q, Zhang W, et al. Anti-bacterial, icephobic, and corrosion protection potentials of superhydrophobic nanodiamond composite coating. *Colloids Surf a Physicochem Eng Asp*. 2021:630. doi:10.1016/j.colsurfa.2021.127532
63. Iqbal S, Rafique MS, Iqbal N, Bashir S, Malarvili MB, Anjum AA. Development of Versatile, thermally stable, flexible, UV-resistant and antibacterial polyvinyl alcohol-Nanodiamonds composite for efficient food packaging. *Heliyon*. 2024;10(12):e33270. doi:10.1016/j.heliyon.2024.e33270
64. Zhang T, Kalimuthu S, Rajasekar V, et al. Biofilm inhibition in oral pathogens by nanodiamonds. *Biomater Sci*. 2021;9(15):5127–5135. doi:10.1039/d1bm00608h
65. Houshyar S, Kumar GS, Rifai A, et al. Nanodiamond/poly-ε-caprolactone nanofibrous scaffold for wound management. *Mater Sci Eng C*. 2019;100:378–387. doi:10.1016/j.msec.2019.02.110
66. Chernysheva MG, Badun GA, Popov AG, Chashchin IS, Anuchina NM, Panchenko AV. Preparation of nanodiamond–lysozyme–miramistin composite and prospects of its application in heart valve prosthetics. *Colloid J*. 2024;86(1):120–129. doi:10.1134/S1061933X23600987
67. Iqbal S, Rafique MS, Iqbal N, Akhtar S, Anjum AA, Malarvili MB. Synergistic effect of silver-nanodiamond composite as an efficient antibacterial agent against E. coli and S. aureus. *Heliyon*. 2024;10(9):e30500. doi:10.1016/j.heliyon.2024.e30500
68. Baron AV, Osipov NV, Yashchenko SV, et al. Adsorption of viral particles from the blood plasma of patients with viral hepatitis on nanodiamonds. *Dokl Biochem Biophys*. 2016;469(1):244–246. doi:10.1134/S1607672916040037
69. Khalid A, Bai D, Abraham AN, et al. Electrospun nanodiamond-silk fibroin membranes: a multifunctional platform for biosensing and wound-healing applications. *ACS Appl Mater Interfaces*. 2020;12(43):48408–48419. doi:10.1021/acsami.0c15612
70. Houshyar S, Mirzadeh N, Pillai MM, et al. Surgical mesh coatings for infection control and temperature sensing: an in-vitro investigation. *OpenNano*. 2021:5. doi:10.1016/j.onano.2021.100032
71. Park H, Patil TV, Lee J, Kim H, Cho SJ, Lim KT. NIR-activated catechol-functionalized nanodiamond nanofibers for accelerating on-demand MRSA and E. coli biofilm eradication. *J Biol Eng*. 2025;19(1). doi:10.1186/s13036-024-00469-6
72. Rifai A, Tran N, Reineck P, et al. Engineering the interface: nanodiamond coating on 3D-printed titanium promotes mammalian cell growth and inhibits staphylococcus aureus colonization. *ACS Appl Mater Interfaces*. 2019;11(27):24588–24597. doi:10.1021/acsami.9b07064
73. Streletskiy O, Perevedentseva E, Zavidovskiy I, et al. Amorphous carbon films with embedded well-dispersed nanodiamonds: plasmon-enhanced analysis and possible antimicrobial applications. *Magnetochemistry*. 2022;8(12):171. doi:10.3390/magnetochemistry8120171
74. Růžek V, Louda P, Mitura S, et al. Antimicrobial sol-gel layers with nanodiamonds, dispergation, antimicrobial and cytotoxicity testing. *Emergent Mater*. 2025;8(3):1241–1249. doi:10.1007/s42247-024-00985-7
75. Zhang R, Zheng Y, Liu J, et al. Morphology-dependent antibacterial properties of diamond coatings. *Functional Diamond*. 2022;2(1):204–214. doi:10.1080/26941112.2022.2157225
76. Nunes-Pereira J, Costa P, Fernandes L, et al. Antimicrobial and antibiofilm properties of fluorinated polymers with embedded functionalized nanodiamonds. *ACS Appl Polym Mater*. 2020;2(11):5014–5024. doi:10.1021/acsapm.0c00869
77. Saha T, Houshyar S, Sarker SR, et al. Nanodiamond-chitosan functionalized hernia mesh for biocompatibility and antimicrobial activity. *J Biomed Mater Res A*. 2021;109(12):2449–2461. doi:10.1002/jbm.a.37237
78. DeFlorio W, Zaza A, Arcot Y, et al. Bioinspired superhydrophobic nanocoating based on polydopamine and nanodiamonds to mitigate bacterial attachment to polyvinyl chloride surfaces in food industry environments. *Ind Eng Chem Res*. 2024;63(14):6235–6248. doi:10.1021/acs.iecr.3c04230
79. Liu S, Ulugun B, DeFlorio W, et al. Development of durable and superhydrophobic nanodiamond coating on aluminum surfaces for improved hygiene of food contact surfaces. *J Food Eng*. 2021:298. doi:10.1016/j.jfoodeng.2021.110487
80. Schrand AM, Huang H, Carlson C, et al. Are diamond nanoparticles cytotoxic? *J Phys Chem B*. 2007;111(1):2–7. doi:10.1021/jp066387v
81. Brož A, Bačáková L, Štenclová P, Kromka A, Potocký Š. Uptake and intracellular accumulation of diamond nanoparticles – a metabolic and cytotoxic study. *Beilstein J Nanotechnol*. 2017;8:1649–1657. doi:10.3762/bjnano.8.165
82. Taylor AC, González CH, Miller BS, Edgington RJ, Ferretti P, Jackman RB. Surface functionalisation of nanodiamonds for human neural stem cell adhesion and proliferation. *Sci Rep*. 2017;7(1):7307. doi:10.1038/s41598-017-07361-y
83. Woodhams B, Ansel-Bollepalli L, Surmacki J, et al. Graphitic and oxidised high pressure high temperature (HPHT) nanodiamonds induce differential biological responses in breast cancer cell lines. *Nanoscale*. 2018;10(25):12169–12179. doi:10.1039/C8NR02177E
84. Knötigová PT, Mašek J, Hubatka F, et al. Application of advanced microscopic methods to study the interaction of carboxylated fluorescent nanodiamonds with membrane structures in THP-1 cells: activation of inflammasome NLRP3 as the result of lysosome destabilization. *Mol Pharm*. 2019;16(8):3441–3451. doi:10.1021/acs.molpharmaceut.9b00225
85. Wierzbicki M, Zawadzka K, Wójcik B, et al. Differences in the cell type-specific toxicity of diamond nanoparticles to endothelial cells depending on the exposure of the cells to nanoparticles. *Int J Nanomed*. 2023;18:2821–2838. doi:10.2147/IJN.S411424
86. Chauhan S, Jain N, Nagaich U. Nanodiamonds with powerful ability for drug delivery and biomedical applications: recent updates on in vivo study and patents. *J Pharm Anal*. 2020;10(1):1–12. doi:10.1016/j.jpha.2019.09.003
87. Yuan Y, Wang X, Jia G, et al. Pulmonary toxicity and translocation of nanodiamonds in mice. *Diam Relat Mater*. 2010;19(4):291–299. doi:10.1016/j.diamond.2009.11.022
88. Huang YA, Kao CW, Liu KK, et al. The effect of fluorescent nanodiamonds on neuronal survival and morphogenesis. *Sci Rep*. 2014;4(1):6919. doi:10.1038/srep06919

89. Rojas S, Gispert JD, Martín R, et al. Biodistribution of amino-functionalized diamond nanoparticles. In vivo studies based on 18F radionuclide emission. *ACS Nano*. 2011;5(7):5552–5559. doi:10.1021/nn200986z
90. Nanodiamond Modified Gutta Percha (NDGP) composite for non-surgical Root Canal Therapy (RCT) filler material. 2021. Available from: https://clinicaltrials.gov/study/NCT02698163. Accessed August 5, 2025.
91. Lee DK, Kee T, Liang Z, et al. Clinical validation of a nanodiamond-embedded thermoplastic biomaterial. *Proc Natl Acad Sci*. 2017;114(45). doi:10.1073/pnas.1711924114