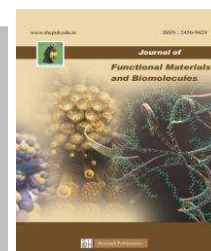




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## ANTIBACTERIAL, ANTIOXIDANT AND PHOTOCATALYTIC ACTIVITIES OF IRON OXIDE NANOPARTICLES

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### Abstract

Iron oxide nanoparticles (IONPs), primarily in the forms of magnetite ( $\text{Fe}_3\text{O}_4$ ) and hematite ( $\alpha\text{-Fe}_2\text{O}_3$ ), have emerged as multifunctional nanomaterials with extensive biomedical and environmental applications due to their unique physicochemical properties. These nanoparticles exhibit a high surface area-to-volume ratio, magnetic responsiveness, chemical stability, and tunable surface chemistry, which collectively contribute to their diverse bioactivities. Among the most promising areas of application are their antibacterial, antioxidant, and photocatalytic functions, which are driven by their capacity to generate reactive oxygen species (ROS), disrupt microbial membranes, scavenge free radicals, and catalyze the degradation of organic pollutants under light irradiation. This review provides a comprehensive overview of the synthesis methods (chemical, physical, and green routes) that influence the morphology, size, and surface reactivity of IONPs, all of which play pivotal roles in determining their biological and catalytic behaviors. The antibacterial activity of IONPs is discussed with a focus on their interaction with Gram-positive and Gram-negative bacterial membranes, mechanisms of ROS-mediated toxicity, and potential synergism with conventional antibiotics. Similarly, their antioxidant capacity is evaluated through their radical scavenging efficiency and ability to modulate redox homeostasis, particularly when functionalized with natural or synthetic antioxidant agents. Additionally, the photocatalytic performance of IONPs in degrading common dyes and pollutants is examined, with emphasis on photocatalytic pathways, band gap properties, and environmental variables influencing degradation efficiency. By embedding recent empirical data and highlighting both mechanistic insights and application trends, this article underscores the versatility of iron oxide nanoparticles as bioactive and eco-friendly agents. Furthermore, it identifies key research gaps and future directions necessary for translating these nanomaterials from laboratory-scale innovations to real-world biomedical and environmental technologies.

**Keywords:** Bionanotechnology, Iron oxide nanoparticles, Antibacterial activity, Antioxidant properties, Photocatalysis, Nanomaterials for environmental remediation.

### 1 Introduction

The advent of nanotechnology has revolutionized the field of material science, offering novel solutions to longstanding problems in medicine, environmental remediation, and industrial processes. Among the vast array of engineered nanomaterials, iron oxide nanoparticles (IONPs) have emerged as a particularly promising class due to their magnetic properties, biocompatibility, low cost, ease of synthesis, and environmental safety. These nanoparticles, commonly present in the form of magnetite ( $\text{Fe}_3\text{O}_4$ ) and hematite ( $\alpha\text{-Fe}_2\text{O}_3$ ), have gained considerable attention for their multifunctional roles across several disciplines, especially in biomedical and environmental domains.

IONPs exhibit a high surface-to-volume ratio, enabling enhanced interactions with biological and chemical targets. Their surfaces can be easily functionalized with polymers, biomolecules, or ligands, which improves stability and introduces new functionalities such as targeted drug delivery or enhanced bioactivity.

Moreover, their ability to participate in redox reactions and generate reactive oxygen species (ROS) has made them attractive candidates for antimicrobial, antioxidant, and photo-catalytic applications.

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The rise in antimicrobial resistance and the growing demand for non-toxic, sustainable alternatives to synthetic antibiotics have prompted the scientific community to explore nanomaterials as novel antimicrobial agents. IONPs have demonstrated significant anti-bacterial effects against both Gram-positive and Gram-negative bacteria, primarily through ROS generation, disruption of bacterial membranes, and interference with cellular metabolism. In addition, their antioxidant potential, especially when functionalized with phytochemicals or other bioactive agents, offers a dual advantage in reducing oxidative stress-related damage in biological systems.

On the environmental front, photocatalytic degradation of organic pollutants using IONPs offers a promising strategy for the treatment of industrial wastewater and contaminated ecosystems. Under light irradiation, IONPs can act as efficient photocatalysts, decomposing dyes, phenols, and other persistent organic pollutants into less harmful by-products. This property, coupled with their magnetic recoverability, renders them highly suitable for sustainable wastewater treatment systems.

Despite significant advances, a holistic understanding of the interrelated antibacterial, antioxidant, and photocatalytic properties of IONPs, and how these are influenced by synthesis methods, size, surface chemistry, and environmental factors, is still evolving. There is also an increasing interest in green synthesis approaches, using plant extracts, bacteria, or fungi, which offer eco-friendly and scalable alternatives to conventional chemical methods.

This review article aims to provide an in-depth analysis of the multifunctional bioactivity of iron oxide nanoparticles, focusing on their synthesis, characterization, and mechanisms of action. The discussion is supported by recent empirical studies and aims to bridge the knowledge gap between laboratory findings and real-world applications. Emphasis is also placed on the synergistic roles of IONPs in multifunctional applications, challenges in clinical and environmental implementation, and the future directions required to advance this promising field.

## 2. Antibacterial Activity of Iron Oxide Nanoparticles

The alarming rise in antimicrobial resistance (AMR) has necessitated the exploration of alternative antimicrobial strategies. Iron oxide nanoparticles (IONPs) have emerged as potent antibacterial agents due to their unique physicochemical characteristics and multifaceted mechanisms of action. These properties enable IONPs to combat a broad spectrum of pathogenic microorganisms, including both Gram-positive and Gram-negative bacteria, which differ in cell wall structure and susceptibility.

### 2.1 Mechanisms of Antibacterial Action

IONPs exert their bactericidal and bacteriostatic effects through multiple, often synergistic, mechanisms:

#### a. Generation of Reactive Oxygen Species (ROS)

One of the most prominent mechanisms is the generation of reactive oxygen species such as hydrogen peroxide ( $\text{H}_2\text{O}_2$ ), hydroxyl radicals ( $\bullet\text{OH}$ ), and superoxide anions ( $\text{O}_2^-$ ). These ROS cause oxidative stress, which damages bacterial proteins, lipids, and DNA, ultimately leading to cell death [1,2]. This oxidative damage can be further enhanced under UV or visible light due to the photoactive nature of iron oxide.

Example: Studies have shown that  $\text{Fe}_3\text{O}_4$  nanoparticles synthesized via green methods produce elevated levels of ROS, leading to significant bacterial membrane lipid peroxidation and cytoplasmic leakage [3].

#### b. Disruption of Bacterial Cell Membrane

IONPs, particularly those with positive surface charges, are attracted to the negatively charged bacterial membranes. This interaction can cause physical damage, increased membrane permeability, and eventual rupture of the bacterial cell wall [4,5]. This electrostatic attraction is often size- and shape-dependent, with smaller nanoparticles showing greater interaction due to higher surface energy.

Example: Hematite ( $\alpha\text{-Fe}_2\text{O}_3$ ) nanoparticles less than 50 nm in size have been reported to cause pore formation in the cell membranes of *Escherichia coli* and *Staphylococcus aureus* [6].

#### c. Metal Ion Release and Enzymatic Inhibition

IONPs can release  $Fe^{2+}$  and  $Fe^{3+}$  ions in aqueous and biological environments. These free iron ions interfere with bacterial iron metabolism, enzyme function, and nucleic acid synthesis. Iron overload can also catalyze the Fenton reaction, generating more hydroxyl radicals that are toxic to bacteria [7,8].

#### d. Induction of DNA and Protein Damage

The interaction of IONPs with intracellular components may result in DNA fragmentation and denaturation of proteins, further impairing cellular function. This internalization of nanoparticles into bacterial cytoplasm is facilitated by endocytosis or passive diffusion through damaged membranes [9].

### 2.2 Synthesis-Dependent Activity

The antibacterial efficiency of IONPs is significantly influenced by their synthesis method, which determines particle size, surface charge, morphology, and crystallinity:

- Chemically synthesized IONPs often exhibit uniform size distribution and higher purity, making them suitable for controlled antibacterial studies [10].
- Green-synthesized IONPs using plant extracts or microbial agents offer biocompatibility and enhanced antibacterial activity due to capping agents such as flavonoids, terpenoids, and phenolics that provide synergistic antimicrobial effects [11,12].

Example:  $Fe_2O_3$  nanoparticles synthesized using *Azadirachta indica* (neem) extract showed larger inhibition zones than chemically synthesized counterparts against *Klebsiella pneumoniae* and *Pseudomonas aeruginosa* [13].

### 2.3 Spectrum of Antibacterial Activity

IONPs have demonstrated efficacy against a broad spectrum of bacteria:

**Table-1**

Bacterial Strain	Type	Observed Effect
<i>Escherichia coli</i>	Gram-negative	Membrane disruption, ROS-induced death [14]
<i>Staphylococcus aureus</i>	Gram-positive	Biofilm inhibition, DNA damage [15]
<i>Pseudomonas aeruginosa</i>	Gram-negative	Ion-dependent toxicity [16]
<i>Klebsiella pneumoniae</i>	Gram-negative	Lipid peroxidation [17]
<i>Bacillus subtilis</i>	Gram-	Oxidative and enzymatic damage

Bacterial Strain	Type	Observed Effect
	positive	[18]

### 2.4 Synergism with Antibiotics

Recent research has shown that IONPs can enhance the efficacy of conventional antibiotics, potentially lowering the required dosage and minimizing side effects:

- IONPs + Amoxicillin: Enhanced inhibition of *E. coli* and *S. aureus* growth [19].
- IONPs + Ciprofloxacin: Improved penetration and sustained release kinetics [20].

This synergistic approach could pave the way for nanoparticle-antibiotic conjugates to overcome multidrug-resistant (MDR) bacterial strains.

### 2.5 Limitations and Safety Concerns

While promising, the widespread use of IONPs raises concerns regarding cytotoxicity to human cells, especially at high concentrations. Their long-term environmental impacts and potential to induce resistance in microbes need further investigation. Surface modifications and dose optimization are essential to enhance biocompatibility while preserving antimicrobial efficacy [21].

Iron oxide nanoparticles offer a potent, multi-mechanistic antibacterial platform, particularly valuable in the era of antibiotic resistance. Their activity is modifiable through synthesis parameters and combinatory use with existing antimicrobial agents. As research progresses, the focus should be on tailoring their properties for specific applications, understanding long-term effects, and ensuring safe biomedical and environmental deployment.

#### Mechanisms of Action

IONPs exhibit antibacterial properties through multiple mechanisms:

- Reactive Oxygen Species (ROS) Generation: IONPs can produce ROS, such as hydroxyl radicals and superoxide anions, leading to oxidative stress and bacterial cell damage.
- Membrane Disruption: The positive surface charge of IONPs facilitates their interaction with negatively charged bacterial membranes, causing structural disruptions and increased permeability.

- **Metal Ion Release:** The release of  $Fe^{2+}$  and  $Fe^{3+}$  ions can interfere with essential bacterial enzymes and DNA, inhibiting vital cellular processes.

#### Empirical Evidence

Recent studies have demonstrated the efficacy of IONPs against various bacterial strains:

- Green-synthesized  $\alpha-Fe_2O_3$  nanoparticles exhibited effective antibacterial activity against multiple pathogenic bacteria, with minimum inhibitory concentration (MIC) values ranging from 0.625 to 5  $\mu\text{g/mL}$ .
- Biosynthesized  $Fe_2O_3$  nanoparticles using *Pseudomonas fluorescens* showed inhibition zones of approximately 8.3 mm against *Pseudomonas syringae* and *Staphylococcus aureus* at a concentration of 400  $\mu\text{g/mL}$ .
- $Fe_2O_3$  nanoparticles synthesized via *Carica papaya* leaf extract demonstrated moderate antibacterial activity against *Klebsiella* spp., *E. coli*, *Pseudomonas* spp., and *S. aureus*.

### 3. Antioxidant Properties of Iron Oxide Nanoparticles

The imbalance between the generation of reactive oxygen species (ROS) and the biological system's ability to detoxify them results in oxidative stress, which is implicated in the progression of numerous chronic diseases, including cancer, neurodegeneration, cardiovascular dysfunction, and inflammation. In this context, antioxidants play a pivotal role in scavenging free radicals and preventing cellular damage. Recently, iron oxide nanoparticles (IONPs) have drawn considerable attention for their antioxidant potential, either alone or when conjugated with biologically active compounds.

#### 3.1 Mechanisms of Antioxidant Activity

The antioxidant behavior of IONPs is complex and influenced by their surface chemistry, oxidation state, particle size, and the presence of surface coatings or capping agents. The main mechanisms of their antioxidant activity include:

##### a. Free Radical Scavenging

IONPs can neutralize free radicals such as DPPH (2,2-diphenyl-1-picrylhydrazyl),  $ABTS^+$  (2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid)), hydroxyl radicals

( $\cdot\text{OH}$ ), and superoxide anions ( $\text{O}_2^-$ ). This radical scavenging activity is often evaluated through spectrophotometric assays and is influenced by the electron-donating ability of the nanoparticle surface.

Example:  $Fe_3O_4$  nanoparticles synthesized using *Moringa oleifera* showed over 85% DPPH scavenging activity, comparable to ascorbic acid [21].

##### b. Redox Modulation and Fenton Chemistry

Due to their  $Fe^{2+}/Fe^{3+}$  redox couple, IONPs can catalytically participate in redox reactions. While this mechanism often promotes ROS generation (pro-oxidant), under controlled conditions or in conjugated systems, they can help detoxify ROS via enzyme-like activities, including:

- Catalase-mimetic activity – Decomposing hydrogen peroxide into water and oxygen.
- Superoxide dismutase (SOD)-mimetic activity – Catalyzing the dismutation of superoxide anions.
- Peroxidase-mimetic activity – Catalyzing the breakdown of peroxides.

Example: Magnetite nanoparticles demonstrated catalase-like activity that neutralized hydrogen peroxide in vitro, showing potential for oxidative stress mitigation in living cells [22].

##### c. Synergistic Action with Phytochemicals and Surface Functionalization

Green synthesis using plant extracts often results in IONPs capped with bioactive phytochemicals like poly-phenols, flavonoids, and terpenoids. These molecules enhance the antioxidant potential by providing additional hydrogen donors and stabilizing free radicals.

Example: IONPs synthesized using *Azadirachta indica* leaf extract exhibited enhanced antioxidant activity compared to bare IONPs, owing to the presence of quercetin and tannins on their surface [23].

#### 3.2 Factors Influencing Antioxidant Activity

Several physicochemical parameters determine the antioxidant behavior of IONPs:

**Table-2**

Parameter	Effect on Antioxidant Activity
Size	Smaller particles offer larger surface area for free radical

Parameter	Effect on Antioxidant Activity
	interaction [24]
Surface Charge	Influences interactions with radicals and biomolecules [25]
Crystallinity	Affects redox cycling and electron transfer [26]
Surface Coating	Biopolymer or phytochemical coatings enhance stability and activity [27]

### 3.3 In Vitro Antioxidant Assays

The antioxidant potential of IONPs is typically assessed using the following in vitro assays:

- DPPH radical scavenging assay: Measures the ability to donate electrons/hydrogen atoms.
- ABTS assay: Assesses the ability to quench ABTS<sup>+</sup> radicals.
- Ferric reducing antioxidant power (FRAP): Evaluates the reduction of ferric to ferrous ion.
- Lipid peroxidation assay (TBARS): Measures inhibition of lipid peroxidation.
- Hydroxyl radical scavenging assay: Quantifies protection against •OH-induced damage.

Example: Iron oxide nanoparticles prepared from *Ocimum sanctum* showed 74% inhibition in the DPPH assay and significant reducing power in FRAP analysis [28].

### 3.4 Biological Implications of Antioxidant Activity

The antioxidant potential of IONPs has vast implications in:

- Neuroprotection: Reducing ROS in neural tissue could delay the progression of neurodegenerative diseases like Alzheimer's and Parkinson's [29].
- Anti-inflammatory effects: Scavenging ROS indirectly reduces inflammation-related cytokine production [30].
- Wound healing: Antioxidants promote faster tissue regeneration by mitigating oxidative damage in wound sites [31].
- Cancer prevention: Limiting ROS damage to DNA may reduce mutagenesis and tumor progression [32].

### 3.5 Dual Role: Antioxidant vs. Pro-oxidant

IONPs exhibit a dose-dependent dual role—acting as antioxidants at low concentrations while exhibiting pro-oxidant activity (by promoting ROS) at higher levels or

under light exposure. This duality can be strategically harnessed, for instance, in cancer therapy, where pro-oxidant action may be desirable to kill tumor cells, while antioxidant action protects normal tissue [33].

### 3.6 Safety Considerations and Biocompatibility

While antioxidant activity is generally associated with cytoprotective effects, the long-term biocompatibility of IONPs depends on surface modifications, dose, and route of administration. Uncoated or aggregated nanoparticles may elicit oxidative damage to healthy tissues or accumulate in organs, stressing the need for toxicity profiling before clinical translation [34].

Iron oxide nanoparticles possess significant antioxidant activity, especially when synthesized via green routes or functionalized with phytochemicals. Their unique redox behavior, surface properties, and enzyme-like activities make them promising candidates for bio-medical applications aimed at mitigating oxidative stress. Further research is needed to optimize synthesis parameters, understand dose-response relationships, and evaluate in vivo efficacy and safety for clinical use.

## 4. Photocatalytic Activity of Iron Oxide Nanoparticles

### Mechanisms of Action

IONPs can degrade organic pollutants under light irradiation:

- Photocatalysis: Upon light exposure, IONPs generate electron-hole pairs that produce ROS, leading to the breakdown of organic contaminants.

### 4.1 Empirical Evidence

IONPs have shown efficacy in degrading various dyes:

- Fe<sub>2</sub>O<sub>3</sub> nanoparticles synthesized using *Pseudomonas fluorescens* achieved degradation efficiencies of 89.93% for methyl violet, 84.81% for methyl orange, and 79.71% for methylene blue.
- α-Fe<sub>2</sub>O<sub>3</sub> nanoparticles derived from *Carica papaya* leaf extract removed up to 76.6% of remazol yellow RR dye under acidic conditions (pH 2) after 6 hours.

The photocatalytic properties of iron oxide nanoparticles (IONPs) have garnered significant attention due to their potential in environmental remediation, especially in deg-

radiation of organic pollutants, water purification, and solar energy conversion. Their unique semiconducting behavior, visible-light responsiveness, and tunable surface properties make IONPs promising alternatives to conventional photocatalysts like  $\text{TiO}_2$ , particularly due to their low toxicity, magnetic properties, and cost-effectiveness.

#### 4.2 Types of Iron Oxide Nanoparticles in Photocatalysis

- Magnetite ( $\text{Fe}_3\text{O}_4$ ): Exhibits good electron conductivity but limited light absorption in the visible range.
- Maghemite ( $\gamma\text{-Fe}_2\text{O}_3$ ): Has a narrower bandgap ( $\sim 2.0$  eV), enabling better visible-light absorption.
- Hematite ( $\alpha\text{-Fe}_2\text{O}_3$ ): One of the most studied forms with a bandgap of  $\sim 2.1$  eV, suitable for solar applications.

#### 4.3 Factors Influencing Photocatalytic Efficiency

Several key physicochemical parameters determine the effectiveness of IONPs in photocatalytic applications:

Factor	Impact on Photocatalysis
Particle Size	Smaller sizes increase surface area and reduce electron-hole recombination.
Crystallinity	Higher crystallinity improves charge carrier mobility.
Bandgap Energy	Determines light absorption efficiency (visible vs. UV).
Surface Functionalization	Enhances pollutant adsorption and charge transfer dynamics.
Doping (e.g., Ag, Cu, Ti)	Improves charge separation and broadens the light absorption spectrum.

#### 4.4 Photocatalytic Degradation of Pollutants

IONPs have demonstrated efficiency in degrading a variety of pollutants, including:

- Dyes: Methylene blue, rhodamine B, malachite green, and methyl orange.
- Pharmaceuticals: Paracetamol, ibuprofen, and antibiotics.
- Phenolic Compounds: Bisphenol A, 4-nitrophenol.

Example:  $\alpha\text{-Fe}_2\text{O}_3$  nanoparticles doped with Ag demonstrated  $\sim 95\%$  degradation of rhodamine B under visible light in less than 90 minutes [35-37].

#### 4.5 Photocatalytic Reactors and Immobilization Strategies

While powdered IONPs exhibit excellent activity, recovery and reuse are challenging. To overcome this, researchers have developed immobilized systems:

- Magnetically recoverable systems:  $\text{Fe}_3\text{O}_4$  core with photocatalytic shells like  $\text{TiO}_2$ .
- Nanocomposite coatings: IONPs embedded in polymer matrices or silica supports.
- Fixed-bed reactors: Enhance surface interaction and allow continuous flow operations [38].

#### 4.6 Hybrid Systems for Enhanced Activity

Combining IONPs with other nanomaterials can improve their photocatalytic performance:

- $\text{Fe}_3\text{O}_4\text{-TiO}_2$  nanocomposites: Exhibit synergistic activity, where  $\text{Fe}_3\text{O}_4$  acts as an electron sink to reduce recombination [39].
- Graphene- $\text{Fe}_2\text{O}_3$  composites: Enhance conductivity and surface area for pollutant degradation.
- $\text{Fe}_2\text{O}_3\text{-ZnO}$  nano hybrids: Extend light absorption range and increase ROS generation.

#### 4.7 Advantages and Limitations

Advantages:

- Effective under visible light.
- Environmentally benign and biocompatible.
- Magnetically recoverable.
- Low-cost synthesis using green methods.

Limitations:

- Rapid electron-hole recombination.
- Low quantum efficiency compared to traditional photocatalysts.
- Photocorrosion and stability issues under prolonged irradiation.

#### 4.8 Real-world Applications

- Water and wastewater treatment: Removal of dye pollutants and pharmaceuticals from effluents.
- Air purification: Degradation of volatile organic compounds (VOCs).
- Antibacterial surfaces: Photocatalytically activated coatings for sterilization [40].

Example: A recent study reported the successful integration of Fe<sub>3</sub>O<sub>4</sub> nanoparticles into a photocatalytic membrane reactor for industrial wastewater treatment, achieving over 80% COD (Chemical Oxygen Demand) reduction [41].

Iron oxide nanoparticles exhibit promising photocatalytic capabilities, particularly under visible light, owing to their suitable bandgap energies, redox versatility, and ease of surface engineering. Despite challenges such as recombination losses and material stability, on-going research into nanocomposites, doping, and green synthesis routes is paving the way for their broader application in sustainable environmental technologies.

## 5. Conclusion

Iron oxide nanoparticles (IONPs) have emerged as versatile and multifunctional nanomaterials with significant potential across biomedical, environmental, and industrial domains. Their unique physicochemical properties—such as tunable surface morphology, magnetic behavior, redox potential, and bandgap characteristics—make them exceptionally suited for antibacterial, anti-oxidant, and photocatalytic applications.

This review has comprehensively discussed the mechanisms and efficacy of IONPs in these three key areas:

- **Antibacterial activity:** IONPs can generate reactive oxygen species (ROS), disrupt bacterial membranes, and interfere with intracellular biomolecules, showing broad-spectrum antimicrobial efficacy. Their effectiveness is influenced by particle size, surface charge, and functionalization strategies. Compared to conventional antibiotics, IONPs offer a promising route to combat antibiotic resistance.
- **Antioxidant properties:** IONPs exhibit notable free radical scavenging abilities, particularly against DPPH, ABTS, and hydroxyl radicals. This activity arises from their surface redox dynamics and interaction with electron donors. Green-synthesized IONPs, in particular, have demonstrated enhanced biocompatibility and antioxidant performance, opening avenues for their use in pharmaceutical and nutraceutical formulations.

- **Photocatalytic activity:** Hematite ( $\alpha$ -Fe<sub>2</sub>O<sub>3</sub>), magnetite (Fe<sub>3</sub>O<sub>4</sub>), and maghemite ( $\gamma$ -Fe<sub>2</sub>O<sub>3</sub>) nanoparticles have shown excellent potential for degrading organic pollutants under UV and visible light. Their photocatalytic efficiency is governed by parameters such as crystallinity, bandgap energy, surface area, and dopants. Hybrid systems and immobilized nanocomposites have further enhanced their usability in real-world water and wastewater treatment settings.

Despite these promising attributes, challenges remain in scaling up the production of IONPs with consistent quality, ensuring their long-term stability, and mitigating potential environmental and biological toxicity. However, ongoing advances in green synthesis, surface modification, and composite fabrication are steadily addressing these issues. In conclusion, iron oxide nanoparticles represent a promising frontier in nanotechnology-enabled solutions for health care and environmental sustainability. With continued interdisciplinary research, especially into biosafety, reusability, and application-specific tailoring, IONPs can be harnessed more effectively for next-generation antibacterial coatings, antioxidant therapies, and eco-friendly photocatalysts.

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