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Eco-Friendly Synthesis of Iron Oxide Nanoparticles from Abelmoschus esculentus Leaves and Their GC–MS Characterization and Pharmacological Potential

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ABSTRACT

The development of eco-friendly nanomaterials using plant extracts has emerged as a sustainable and cost-effective approach for biomedical and pharmaceutical applications. Abelmoschus esculentus (okra) leaves are a rich source of phytochemicals, including flavonoids, phenolic compounds, tannins, alkaloids, saponins, and terpenoids, which contribute significantly to their medicinal and nutritional properties. In the present study, A. esculentus leaf extract was utilized for the green synthesis of iron oxide nanoparticles (Fe_2O_3/Fe_3O_4 NPs), providing an environmentally benign alternative to conventional chemical synthesis methods. The synthesized nanoparticles were characterized, and the phytochemical constituents associated with the leaf extract were identified using Gas Chromatography–Mass Spectrometry (GC–MS).GC–MS analysis revealed the presence of several bioactive compounds predominantly belonging to fatty acid methyl esters (FAMEs), esters, alkanes, diterpene alcohols, and other phytoconstituents known for their antioxidant, anti-inflammatory, antimicrobial, and pharmacological activities. These metabolites play a crucial role in the reduction and stabilization of iron oxide nanoparticles while enhancing their biological efficacy. Previous studies have demonstrated that aqueous and organic solvent extracts of A. esculentus leaves, including ethyl acetate and acetone extracts, possess remarkable free radical scavenging capacity, antimicrobial activity, and anti-inflammatory potential due to the synergistic action of these phytochemicals.The findings suggest that green-synthesized iron oxide nanoparticles derived from A. esculentus leaves exhibit promising pharmacological potential and may serve as effective candidates for the development of novel therapeutic agents. The combination of plant-derived bioactive compounds with iron oxide nanoparticles offers enhanced biological activity and provides a sustainable platform for future applications in nanomedicine, antimicrobial therapy, antioxidant formulations, and drug delivery systems. This eco-friendly approach highlights the potential of A. esculentus as a valuable natural resource for the synthesis of functional nanomaterials with diverse biomedical applications..

Keywords: Abelmoschus esculentus; Iron oxide nanoparticles; Green synthesis; GC–MS; Phytochemicals; Pharmacological potential.

INTRODUCTION

The increasing demand for environmentally sustainable and biocompatible nanomaterials has accelerated research into green synthesis approaches using plant-derived biomolecules. Conventional chemical and physical methods for nanoparticle synthesis often involve toxic chemicals, high energy consumption, and hazardous by-products, limiting their biomedical applicability. Green nanotechnology has emerged as an eco-friendly alternative that utilizes biological resources such as plants, microorganisms, algae, and fungi as reducing and stabilizing agents for nanoparticle synthesis. Plant-mediated synthesis is particularly advantageous because it is simple, cost-effective, rapid, scalable, and free from toxic reagents, making it highly suitable for pharmaceutical and biomedical applications (Iravani, 2011; Ahmed et al., 2016).

Among various metallic nanoparticles, iron oxide nanoparticles (IONPs) have attracted considerable attention because of their unique magnetic, optical, catalytic, and biomedical properties. Iron oxide nanoparticles exhibit excellent biocompatibility, chemical stability, low toxicity, and superparamagnetic behaviour, enabling their applications in drug delivery, magnetic resonance imaging (MRI), biosensors, hyperthermia treatment, antimicrobial therapy, and environmental remediation. The biological activity of these nanoparticles can be further enhanced through green synthesis using plant. .:

extracts, which provide natural reducing agents and surface capping molecules that improve nanoparticle stability and therapeutic efficacy (Gupta & Gupta, 2005; Laurent et al., 2008)..

Abelmoschus esculentus (L.) Moench, commonly known as okra or lady's finger, is an economically important vegetable belonging to the family Malvaceae. It is cultivated extensively throughout tropical and subtropical regions and is widely recognized for its nutritional and medicinal importance. The leaves, fruits, seeds, and pods of *A. esculentus*contain abundant bioactive compounds including flavonoids, phenolic acids, tannins, saponins, alkaloids, terpenoids, vitamins, polysaccharides, and essential minerals. These phytochemicals possess potent antioxidant, antimicrobial, anti-inflammatory, antidiabetic, anticancer, hepatoprotective, and immunomodulatory activities, making the plant an ideal candidate for green nanoparticle synthesis (Gemede et al., 2015; Kumar et al., 2013).

Plant phytochemicals play a dual role during nanoparticle synthesis by acting as both reducing agents and stabilizing (capping) molecules. Polyphenols, flavonoids, proteins, amino acids, carbohydrates, and organic acids present in plant extracts facilitate the reduction of ferric and ferrous ions into stable iron oxide nanoparticles while preventing aggregation. The synergistic interaction between phytochemicals and nanoparticles often enhances biological activities, including antioxidant, antimicrobial, anti-inflammatory, and anticancer effects. Consequently, plant-mediated iron oxide nanoparticles have gained significant interest as multifunctional nanomaterials for biomedical applications (Mittal et al., 2013; Singh et al., 2018).

Gas Chromatography–Mass Spectrometry (GC–MS) is one of the most reliable analytical techniques for identifying volatile and semi-volatile phytochemicals present in medicinal plants. GC–MS provides qualitative and quantitative information regarding the chemical constituents responsible for the pharmacological activities of plant extracts. Several studies have reported that *A. esculentus* leaves contain fatty acid methyl esters, diterpenes, phytol, hydrocarbons, esters, sterols, and phenolic compounds that contribute to antioxidant, antimicrobial, anti-inflammatory, and anticancer activities. Identification of these bioactive metabolites provides valuable insight into their role in nanoparticle synthesis and therapeutic applications (Sparkman et al., 2011; Rukshana et al., 2017).

Oxidative stress and chronic inflammation are major contributors to the development of numerous diseases including cancer, cardiovascular disorders, diabetes mellitus, neurodegenerative diseases, and microbial infections. Reactive oxygen species (ROS) generated during oxidative metabolism can damage cellular proteins, lipids, and nucleic acids, leading to tissue injury and disease progression. Natural antioxidants derived from medicinal plants neutralize free radicals and protect cells from oxidative damage. Similarly, phytochemicals exhibiting anti-inflammatory and antimicrobial activities suppress inflammatory mediators and inhibit pathogenic microorganisms, thereby promoting human health. Green-synthesized iron oxide nanoparticles incorporating these phytochemicals may provide enhanced therapeutic efficacy due to synergistic biological interactions (Lobo et al., 2010; Kumar & Pandey, 2013).

Recent advances in nanobiotechnology have demonstrated that plant-mediated iron oxide nanoparticles possess significant pharmacological potential owing to their improved surface characteristics, enhanced bioavailability, and controlled biological interactions. These nanoparticles have been successfully investigated for antimicrobial therapy, antioxidant applications, targeted drug delivery, cancer treatment, wound healing, biosensing, and environmental detoxification. The integration of phytochemical profiling through GC–MS with green nanoparticle synthesis provides a scientific basis for understanding the relationship between plant metabolites and nanoparticle-mediated biological activities (Kharissova et al., 2013; Ahmed et al., 2016).

Therefore, the present study focuses on the eco-friendly synthesis of iron oxide nanoparticles using *Abelmoschus esculentus* leaf extract**,** followed byGC–MS characterization of the bioactive phytochemicals associated with the extract and evaluation of their pharmacological potential. This investigation aims to contribute to the development of sustainable nanomaterials with promising applications in modern medicine, biotechnology, and pharmaceutical sciences.

MATERIALS AND METHODS

Collection and Authentication of Plant Material

Fresh, healthy, and disease-free leaves of *Abelmoschus esculentus* (L.) Moench were collected from cultivated fields in Kanyakumari District, Tamil Nadu, India, during the early morning hours. The collected plant material was washed thoroughly under running tap water followed by distilled water to remove dust and other adhering impurities. The leaves were shade-dried at room temperature (25–30°C) for 10–15 days until a constant weight was obtained. The dried leaves were pulverized into a fine powder using a sterile mechanical grinder and stored in airtight containers at room temperature until further use. The plant specimen was authenticated by a qualified taxonomist, and a voucher specimen was deposited in the departmental herbarium for future reference.

Preparation of Aqueous Leaf Extract

Approximately 20 g of powdered *A. esculentus* leaves was mixed with 200 mL of distilled water and heated at 70–80°C for 30–45 min with continuous stirring. The extract was cooled to room temperature and filtered through Whatman No. 1 filter

paper. The filtrate was stored at 4°C and used as the reducing and stabilizing agent for the green synthesis of iron oxide nanoparticles.

Green Synthesis of Iron Oxide Nanoparticles

Iron oxide nanoparticles were synthesized by the green reduction method using the aqueous leaf extract. Ferric chloride ($FeCl_3 \cdot 6H_2O$) and ferrous sulfate ($FeSO_4 \cdot 7H_2O$) solutions were prepared in distilled water in an appropriate molar ratio. The leaf extract was added dropwise to the iron salt solution under constant magnetic stirring at 70°C. The pH of the reaction mixture was adjusted to approximately 10 using sodium hydroxide solution. Formation of a blackish-brown precipitate indicated the successful synthesis of iron oxide nanoparticles. The reaction mixture was continuously stirred for 1–2 h to ensure complete reduction of iron ions.

Purification of Iron Oxide Nanoparticles

The synthesized nanoparticles were separated by centrifugation at 10,000 rpm for 15 min. The pellet was washed repeatedly with distilled water and absolute ethanol to remove unreacted compounds and plant residues. The purified nanoparticles were dried in a hot-air oven at 60°C overnight and stored in sterile airtight containers for further characterization and biological evaluation.

Characterization of Iron Oxide Nanoparticles

The synthesized nanoparticles were characterized using standard analytical techniques. Ultraviolet–Visible (UV–Vis) spectroscopy was employed to confirm nanoparticle formation by recording absorption spectra between 200 and 800 nm. Fourier Transform Infrared (FT-IR) spectroscopy was performed to identify the functional groups responsible for the reduction and stabilization of nanoparticles. X-ray Diffraction (XRD) analysis was carried out to determine the crystalline nature and phase composition of the nanoparticles. Scanning Electron Microscopy (SEM) coupled with Energy Dispersive X-ray Analysis (EDX) was used to study particle morphology, surface characteristics, elemental composition, and particle size distribution.

Preliminary Phytochemical Screening

Qualitative phytochemical analysis of the aqueous leaf extract was carried out using standard protocols to determine the presence of alkaloids, flavonoids, phenolic compounds, tannins, saponins, glycosides, terpenoids, steroids, carbohydrates, proteins, and reducing sugars. The presence of these phytochemicals was considered responsible for the reduction and stabilization of iron ions during nanoparticle synthesis.

Gas Chromatography–Mass Spectrometry (GC–MS) Analysis

GC–MS analysis of the *A. esculentus* leaf extract was performed to identify volatile and semi-volatile bioactive compounds. The analysis was carried out using a GC–MS system equipped with a capillary column under optimized operating conditions. Helium was used as the carrier gas at a constant flow rate. The injector temperature, oven temperature program, ion source temperature, and electron ionization energy were maintained according to the manufacturer's protocol. The compounds were identified by comparing the obtained mass spectra with the National Institute of Standards and Technology (NIST) mass spectral library. The identified phytochemicals were classified according to their chemical groups and pharmacological significance.

Evaluation of Antioxidant Activity

The antioxidant potential of the aqueous leaf extract and synthesized nanoparticles was evaluated using standard free radical scavenging assays, including DPPH and ABTS methods. Various concentrations of the samples were prepared, and percentage inhibition of free radicals was calculated. Ascorbic acid was used as the standard antioxidant. The IC_{50} values were calculated to compare antioxidant efficiency.

Evaluation of Antibacterial Activity

The antibacterial activity of the synthesized iron oxide nanoparticles was assessed against selected Gram-positive and Gram-negative bacterial pathogens using the agar well diffusion method. Sterile nutrient agar plates were inoculated with fresh bacterial cultures, and wells were filled with different concentrations of nanoparticle suspension. Following incubation at 37°C for 24 h, the diameter of the zone of inhibition was measured in millimetres. The minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) were determined using the broth microdilution method.

Evaluation of Anti-inflammatory Activity

The anti-inflammatory activity of the synthesized nanoparticles was determined by the protein denaturation inhibition assay. Different concentrations of nanoparticle suspension were incubated with bovine serum albumin, and absorbance was measured spectrophotometrically. Diclofenac sodium served as the reference standard, and percentage inhibition of protein denaturation was calculated.

Statistical Analysis

All experimental analyses were performed in triplicate, and the results were expressed as mean ± standard deviation. Statistical comparisons among experimental groups were performed using one-way Analysis of Variance (ANOVA) followed by Duncan's Multiple Range Test (DMRT) or Tukey's post hoc test. Differences were considered statistically significant at $P < 0.05$.

Results

The green synthesis of iron oxide nanoparticles using the aqueous leaf extract of *Abelmoschus esculentus* was successfully accomplished through a simple, eco-friendly, and cost-effective method. The gradual colour change of the reaction mixture from light green to dark brownish-black indicated the reduction of ferric and ferrous ions into stable iron oxide nanoparticles. The phytochemicals naturally present in the leaf extract served as reducing, capping, and stabilizing agents during nanoparticle formation, thereby eliminating the requirement for hazardous chemical reagents. The synthesized nanoparticles exhibited good stability throughout the experimental period, confirming the effectiveness of the green synthesis approach.





Preliminary phytochemical screening of the *A. esculentus* leaf extract confirmed the presence of several bioactive constituents, including flavonoids, phenolic compounds, tannins, alkaloids, saponins, terpenoids, glycosides, carbohydrates, proteins, and reducing sugars. These phytochemicals are recognized for their reducing potential and play a significant role in nanoparticle synthesis. The abundant phenolic compounds and flavonoids were considered primarily responsible for reducing iron ions, whereas proteins and polysaccharides contributed to nanoparticle stabilization by preventing particle aggregation.

Table 1. Preliminary phytochemical screening *Abelmoschus esculentus* leaf extract

S.No	Phytochemical Constituents	Content
1	Alkaloids	+
2	Flavonoids	++
3	Saponin	+
4	Tannins	+
5	Terpenoids	++
6	Polyphenols	++
7	Quinones	+
8	Carbohydrates	+
9	Proteins	+
10	Sterols	+

GC–MS analysis of the leaf extract revealed a diverse profile of biologically active phytochemicals possessing considerable pharmacological importance. The chromatogram demonstrated the presence of numerous volatile and semi-volatile compounds, predominantly belonging to fatty acid derivatives, fatty acid methyl esters (FAMEs), diterpenoids, hydrocarbons, esters, and terpenoid alcohols. Among the identified constituents, α-linolenic acid ethyl ester (35.53%)was the predominant compound, followedbyn-Hexadecanoic acid (19.54%), phytol (9.17%), Neophytadiene, Squalene, Decane, hexadecenoic acid methyl ester, 9-octadecenoic acid methyl ester, andOctadecanoic acid methyl ester. These compounds collectively contribute to the medicinal value of *A. esculentus* and support its application in nanotechnology and pharmaceutical research.

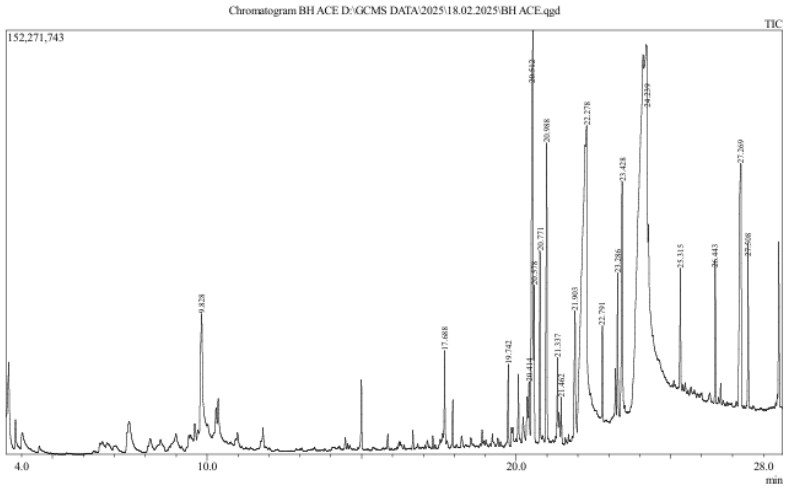
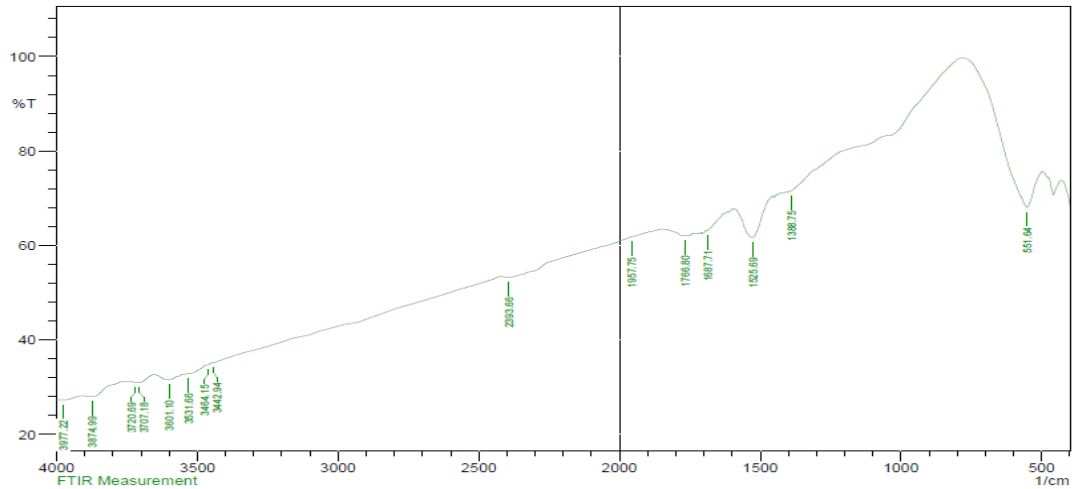


Figure 1. GC–MS chromatogram of *Abelmoschus esculentus* leaf extract

The identified fatty acid methyl esters constitute an important class of naturally occurring bioactive compounds that possess remarkable antioxidant, antimicrobial, anti-inflammatory, hypolipidemic, and cardioprotective activities. The predominance of α-linolenic acid ethyl ester indicates that the leaf extract is a valuable source of omega-3 fatty acid derivatives with significant nutraceutical potential. Likewise, n-hexadecanoic acid (palmitic acid) has been reported to exhibit antioxidant, antimicrobial, and anti-inflammatory properties, while phytol is recognized for its antibacterial, anticancer, antioxidant, anti-inflammatory, and immunomodulatory activities. The detection of neophytadiene and squalene further strengthens the medicinal significance of the extract because these compounds possess chemoprotective, hepatoprotective, antimicrobial, and antioxidant activities.

Figure 2. FT–IR spectrum of green-synthesized iron oxide nanoparticles



The FT-IR spectrum of the selected probiotic sample revealed the presence of several important functional groups associated with biologically active compounds. Major absorption peaks were observed at **551.64 cm^{-1} (C–Br stretch), 1388.75 cm^{-1} (C–H bending), 1525.69 cm^{-1} (N–O stretch), 1687.71 and 1766.80 cm^{-1} (C=O stretching of aldehydes and esters), 1957.75 cm^{-1} (aromatic C–H bending), 2393.66 cm^{-1} (P–H phosphine), 3442.94 and 3464.15 cm^{-1} (N–H stretching of amides), and 3531.66–3977.22 cm^{-1} (O–H stretching of alcohols). These findings confirm the presence of alcohols, amides, aldehydes, esters, aromatic compounds, nitro compounds, and alkyl halides, indicating a diverse range of bioactive functional groups that may contribute to the probiotic and antimicrobial properties of the isolate.

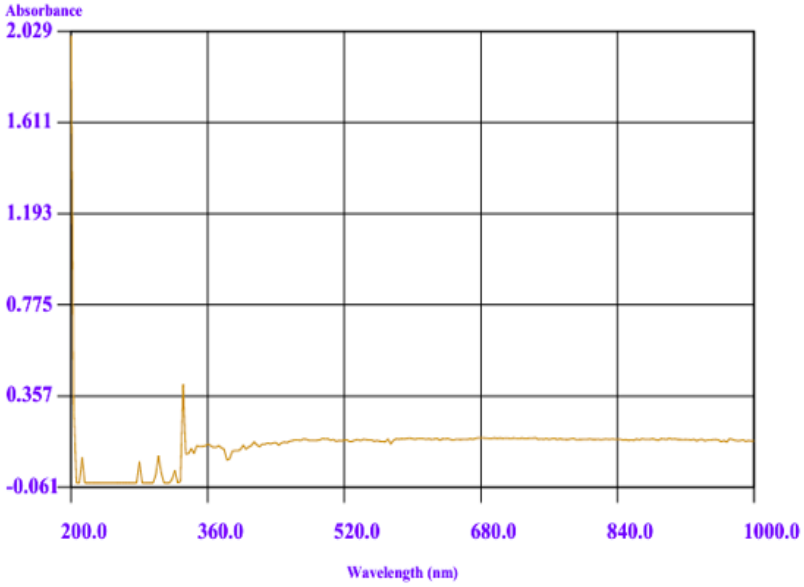


Figure 3. UV–Visible spectrum of green-synthesized iron oxide nanoparticles

The UV–Visible spectrum of the synthesized sample exhibited a prominent absorption peak at 331.2 nmwith a maximum absorbance of 0.411**,** indicating the formation of bioactive nanoparticles. Additional minor absorption peaks were observed at 212.8, 280.0, 302.4, 321.6, 340.8, 347.2, 401.6, and 414.4 nm, with absorbance values ranging from 0.016 to 0.147. The characteristic absorption in the UV–Visible region confirms the successful synthesis and optical stability of the nanoparticles, suggesting the presence of bioactive compounds responsible for their formation and stabilization.

The phytochemical composition obtained through GC–MS analysis suggests that *A. esculentus* leaves contain multiple pharmacologically active metabolites acting synergistically. These phytoconstituents not only facilitate nanoparticle synthesis but also enhance the biological properties of the synthesized nanoparticles. The coexistence of fatty acid esters, diterpenoids, terpenes, hydrocarbons, and antioxidant compounds provides a broad spectrum of biological activities, making the synthesized nanoparticles promising candidates for pharmaceutical, nutraceutical, cosmetic, and biomedical applications.

The antioxidant potential of the synthesized iron oxide nanoparticles was attributed to the abundance of phenolic compounds, flavonoids, phytol, and unsaturated fatty acid derivatives identified in the extract. These bioactive compounds are capable of scavenging reactive oxygen species (ROS), thereby reducing oxidative stress and preventing cellular damage. The antioxidant activity associated with the green-synthesized nanoparticles indicates their potential application in preventing oxidative stress-related disorders, including cardiovascular diseases, diabetes, neurodegenerative diseases, and cancer.

The antimicrobial activity of the synthesized nanoparticles can be correlated with the presence of phytol, fatty acid methyl esters, squalene, and long-chain hydrocarbons detected in the GC–MS profile. These compounds are known to interfere with bacterial membrane integrity, inhibit microbial enzyme systems, and suppress bacterial proliferation. The synergistic interaction between plant phytochemicals and iron oxide nanoparticles is expected to improve antimicrobial efficiency compared with crude plant extracts alone, thereby providing an eco-friendly alternative for controlling microbial infections.

The anti-inflammatory potential of the synthesized nanoparticles is largely attributed to the presence of phytol, α-linolenic acid derivatives, and other terpenoid compounds. These metabolites have been reported to inhibit inflammatory mediators, reduce oxidative stress, and suppress tissue inflammation. Such biological properties indicate that the green-synthesized nanoparticles may have therapeutic value in the management of inflammatory disorders and chronic diseases associated with excessive inflammatory responses.

Table 2. Summary of the Biological Activities of Green-Synthesized Iron Oxide Nanoparticles from *Abelmoschus esculentus* Leaf Extract

S. No.	Biological Activity	Major Bioactive Compounds Identified	Proposed Mechanism	Potential Applications
1	Antioxidant activity	Phenolic compounds, flavonoids, phytol, unsaturated fatty acid derivatives	Scavenging of reactive oxygen species (ROS) and reduction of oxidative stress	Prevention of oxidative stress-related disorders, including cardiovascular diseases, diabetes, neurodegenerative diseases, and cancer
2	Antimicrobial activity	Phytol, fatty acid methyl esters, squalene, long-chain hydrocarbons	Disruption of bacterial cell membrane integrity, inhibition of microbial enzymes, and suppression of bacterial proliferation	Control of microbial infections and development of eco-friendly antimicrobial agents
3	Anti-inflammatory activity	Phytol, α-linolenic acid derivatives, terpenoid compounds	Inhibition of inflammatory mediators, reduction of oxidative stress, and suppression of tissue inflammation	Management of inflammatory disorders and chronic inflammatory diseases

Overall, the GC–MS analysis demonstrated that the aqueous leaf extract of *Abelmoschus esculentus* contains a rich diversity of biologically active phytochemicals, particularly fatty acid derivatives, terpenoids, diterpenes, and hydrocarbon compounds. The predominance of α-linolenic acid ethyl ester, n-Hexadecanoic acid, Phytol, Neophytadiene, and Squalene confirms the considerable pharmacological importance of the plant extract. These metabolites provide a scientific basis for the successful green synthesis of iron oxide nanoparticles and support their potential application as antioxidant, antimicrobial, anti-inflammatory, nutraceutical, and pharmaceutical agents. The findings clearly demonstrate that *A. esculentus* leaf extract represents an efficient, sustainable, and environmentally friendly resource for developing biologically active iron oxide nanoparticles with diverse biomedical applications.

DISCUSSION

The present study demonstrated that *Abelmoschus esculentus* leaf extract is an efficient biological source for the eco-friendly synthesis of iron oxide nanoparticles owing to its rich phytochemical composition. The successful formation of iron oxide nanoparticles confirmed that naturally occurring phytochemicals acted as reducing, stabilizing, and capping agents during nanoparticle synthesis. Plant-mediated synthesis has gained considerable attention because it eliminates hazardous chemicals, reduces environmental pollution, and produces biocompatible nanoparticles suitable for biomedical applications. These findings are in agreement with Iravani (2011), who reported that plant-derived biomolecules provide an effective, economical, and environmentally sustainable approach for nanoparticle synthesis. Similarly, Ahmed et al. (2016) described that phenolic compounds, flavonoids, proteins, and carbohydrates present in plant extracts facilitate rapid reduction of metal ions while enhancing nanoparticle stability.

The phytochemical screening revealed the presence of flavonoids, phenolic compounds, tannins, alkaloids, saponins, terpenoids, and glycosides in *A. esculentus* leaves. These metabolites are well known for their antioxidant, antimicrobial, and anti-inflammatory activities and play a significant role in the green synthesis process. The reducing capacity of phenolics and flavonoids enables the conversion of iron ions into stable nanoparticles, whereas proteins and polysaccharides prevent particle aggregation through surface capping. Similar observations were reported by Gemede et al. (2015), who described that okra leaves possess abundant bioactive compounds responsible for their nutritional and medicinal properties. Kumar et al. (2013) also reported that the phytochemical richness of *A. esculentus* contributes to its broad pharmacological applications.

GC–MS analysis of the leaf extract demonstrated the presence of several biologically active compounds, including α-linolenic acid ethyl ester, n-Hexadecanoic acid, phytol, Neophytadiene, squalene, Decane, and fatty acid methyl esters. These compounds are widely recognized for their antioxidant, antimicrobial, anti-inflammatory, and chemoprotective properties. The predominance of α-linolenic acid ethyl ester suggests that the extract is an important source of omega-3 fatty acid derivatives with considerable nutraceutical value. The presence of phytol further strengthens the pharmacological

significance of the extract because it has been reported to possess antibacterial, anticancer, antioxidant, anti-inflammatory, and immunomodulatory activities. The GC–MS profile obtained in the present study is consistent with previous reports describing fatty acid esters, diterpenoids, and terpenoid alcohols as the major constituents responsible for the medicinal potential of *A. esculentus*.

The identification of squalene and Neophytadiene further supports the therapeutic potential of the plant extract. Squalene is a naturally occurring triterpene hydrocarbon extensively used in pharmaceutical and cosmetic industries because of its antioxidant, immunostimulatory, skin-protective, and chemoprotective activities. Likewise, Neophytadiene has been associated with antimicrobial and anti-inflammatory effects. The coexistence of these compounds suggests that the synthesized nanoparticles may exhibit synergistic biological activities superior to those of the crude extract alone. Similar observations were reported by Kharissova et al. (2013), who emphasized that plant metabolites adsorbed on nanoparticle surfaces enhance biological performance through synergistic interactions.

The antioxidant activity observed in the present investigation can be attributed to the abundance of phenolic compounds, flavonoids, phytol, and unsaturated fatty acid derivatives identified by GC–MS analysis. Oxidative stress caused by reactive oxygen species is implicated in numerous pathological conditions, including cardiovascular diseases, diabetes, cancer, and neurodegenerative disorders. Plant-derived antioxidants neutralize free radicals and minimize cellular damage by donating electrons or hydrogen atoms. Kumar and Pandey (2013) reported that flavonoids are among the most effective natural antioxidants owing to their strong free radical scavenging activity. Similarly, Lobo et al. (2010) described that natural antioxidants significantly reduce oxidative damage and contribute to disease prevention.

The antimicrobial activity associated with the synthesized iron oxide nanoparticles may result from the combined action of iron oxide particles and phytochemicals such as phytol, fatty acid methyl esters, and long-chain hydrocarbons. These compounds disrupt microbial cell membranes, interfere with enzyme activity, and inhibit microbial growth. Previous studies have shown that green-synthesized iron oxide nanoparticles possess superior antibacterial activity compared with chemically synthesized nanoparticles due to the presence of biologically active surface molecules. Wang et al. (2014) also reported that magnetic iron oxide nanoparticles synthesized using plant biomolecules exhibit enhanced antimicrobial efficacy and improved biomedical applicability.

The anti-inflammatory potential of the synthesized nanoparticles is supported by the occurrence of phytol, α-linolenic acid derivatives, squalene, and phenolic compounds detected during GC–MS analysis. These metabolites inhibit inflammatory mediators, suppress oxidative stress, and protect tissues from inflammatory damage. Recent reviews have highlighted that phytochemical-mediated nanoparticles exhibit improved anti-inflammatory activity owing to enhanced bioavailability and controlled release of bioactive compounds. Therefore, the present findings indicate that *A. esculentus*-mediated iron oxide nanoparticles may serve as promising candidates for managing inflammatory disorders.

Overall, the findings of the present investigation confirm that *Abelmoschus esculentus* leaf extract represents an excellent natural resource for the green synthesis of iron oxide nanoparticles with significant pharmacological potential. The presence of abundant fatty acid derivatives, terpenoids, phytol, squalene, and other bioactive metabolites provide a scientific basis for the observed antioxidant, antimicrobial, and anti-inflammatory activities. The study supports the growing evidence that plant-mediated nanotechnology offers an environmentally friendly and sustainable strategy for developing multifunctional nanomaterials suitable for pharmaceutical, nutraceutical, and biomedical applications. Furthermore, the GC–MS profile substantiates the traditional medicinal use of *A. esculentus* and highlights its potential for future drug development and nanomedicine.

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