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Green synthesis of Magnesium oxide nanoparticles from peel of *Carica papaya* and analysis for its anti-inflammatory and antimicrobial potential

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ABSTRACT
Carica papaya is sometimes known as paw paw which is a fast growing herbaceous plant, it belongs to the Caricaceae family and it contains the proteolytic enzymes papain and chymopapain which are active compounds in breaking down or digesting protein. The extract was mixed with distilled water, treated with magnesium nitrate, centrifuged, and dried at 50°C overnight. UV analysis showed a peak at 302.5 nm, while SEM revealed spherical, flower-like MgO NPs. IR spectroscopy confirmed the presence of OH groups, carbon, and water. The nanoparticles exhibited low anti-inflammatory activity (40% inhibition) and mild antimicrobial effects, demonstrating the potential of papaya peel in green synthesis of MgO NPs with moderate biological properties. The extract concentration influenced the size and morphology of the nanoparticles. The synthesis method was simple, cost-effective, and eco-friendly. Bioactive compounds in the extract likely facilitated nanoparticle formation by acting as reducing and stabilizing agents. Additionally, the process avoided the use of toxic chemicals, making it suitable for sustainable nanomaterial production. Despite modest biological activity, the approach shows promise for further development in biomedical or environmental applications.

INTRODUCTION

Inflammation is the body's biological response to external stimuli, such as pathogens, injured cells, or irritants. It serves as a defence mechanism to remove the original cause of damage, such as necrotic cells and tissues, further allowing the healing process to occur [1][2]. There are coordinated reactions between blood vessels, immune cells, and molecular mediators noted and play an essential part in the innate immune system. In general, inflammation can be divided into two categories: acute and chronic [3]. Acute inflammation lasts a few days, and it manifests as redness, heat, swelling, discomfort, and loss of function at the site of injury. Acute inflammation is caused due to injuries, burns and other foreign objects and disappears when the underlying stimulus is removed[4][5]. Chronic inflammation is a long-lasting infection or a chance for its recurrence. It can be asymptomatic and metastatic [6][7].
Antimicrobial and anti-inflammatory drugs play similar functions in the immune system and in the treatment of disease, especially when infections cause inflammation, but do have different ways to approach the target site [8][9]. When the host encounters a pathogen, the immune system activates the inflammatory response through mechanisms such as pattern recognition receptors on immune cells that recognize pathogen-associated molecular patterns (PAMPs). This mechanism triggers the release of inflammatory mediators like histamine and prostaglandins from damaged cells and immune cells like mast cells, which cause blood vessels to widen (vasodilation) and become more permeable, allowing fluid and immune cells to move from the blood into the surrounding tissue. The white blood cells, particularly neutrophils and macrophages, are attracted to the site of infection. They engulf and destroy the pathogens and cellular debris, a process called phagocytosis. This process causes inflammation [10–12].The anti-inflammatory and antimicrobial drugs play a distinct role in resolving inflammation. The antimicrobial medications work by directly targeting and killing or inhibiting the growth of the infectious organisms, resulting in a decrease in irritation of the gingiva. The anti-inflammatory medications, on the other hand, help to avoid excessive inflammation, which can cause tissue damage if left untreated [13][14]. This is especially crucial in cases of infections (such as sepsis or serious viral infections) because an overreaction to inflammation can be more detrimental than the infection itself. Furthermore, controlling inflammation can improve drug delivery and efficacy since inflammation frequently modifies tissue microenvironments (such as increased vascular permeability or pH changes), which can impact the effectiveness of antimicrobial medicines.Both kinds of medications are frequently used in combination in clinical practice; for example, antibiotics are used to eradicate microbes from an infected wound, while anti-inflammatory drugs are used to lessen swelling and encourage healing[15][16]. Therefore, even though they serve different purposes, anti-inflammatory and antibacterial drugs often operate in concert to restore health by treating the root cause of illness as well as its effects.
Nanotechnology plays a large role in the development of fields such as medicine, agriculture, and environmental science. The nanoscale materials have unique properties that make them more effective for various applications, which help in disease treatment, food production, and pollution control [17][18]. The Magnesium oxide nanoparticles exhibit antibacterial activity against various oral pathogens and are not toxic to living cells, due to which they are generally considered biocompatible. The Magnesium oxide nanoparticles can improve the mechanical properties of dental materials

by improving the compressive strength, diametral tensile strength, and shear bond strength. The traditional methods of nanoparticle synthesis, like physical and chemical processes, involve high energy requirements, produce toxic waste, and harmful chemicals. These methods are costly and complex due to which green synthesis is preferred, where it offers a sustainable and environmentally friendly approach to nanoparticle production, by leveraging the reducing and stabilizing properties of natural substances, particularly plant extracts, by eliminating toxic byproducts, and reduces reliance on hazardous chemicals to synthesize nanoparticles [19][20][21].

Carica papaya belongs to the Caricaceae family, is a tropical herbaceous fruit that has natural compounds, which demonstrate anti-inflammatory, antioxidant, and antimicrobial effects. They are also rich in vitamin and mineral content, which helps to maintain a balanced diet[22][23].The papaya fruit is rich in biologically active substances such as papain, chymopapain, flavonoids, alkaloids, saponins, and phenolic compounds. In general, the papaya peels are often considered agricultural waste, but the peels are effective for synthesizing nanoparticles as they have antibacterial, anti-inflammatory, and antioxidant properties, which make them suitable for environmentally friendly applications [24][25]. *Carica papaya* also counteracts oxidative stress in inflammation, skin aging and healing, chronic diseases, and cancers. The oxidative stress occurs due to excessive ROS production, which will cause oxidative damage to tissues [26][27].

The present study indicates that the aqueous extract from the *Carica papaya* peel is used to investigate the environmentally friendly production of magnesium oxide nanoparticles. The optical characteristics of the synthesised MgO NPs will be assessed using UV-visible spectroscopy. The functional groups involved in the reduction and stabilization were identified using Fourier Transform Infrared Spectroscopy (FTIR), and the surface morphology and particle shape will be examined using Scanning Electron Microscopy (SEM). Then the antimicrobial test will be done using the agar well diffusion method with a standard solution such as diclofenac, followed by anti-inflammatory assays. The main goal of this work is to evaluate the efficiency of papaya peel-mediated MgO NPs as a biologically active material with potential therapeutic uses. Hence, this study aims to synthesize magnesium oxide nanoparticles using *Carica papaya* peel extract and evaluate their anti-inflammatory and antimicrobial properties.

MATERIALS AND METHODS

2.1. Chemicals

The *Carica papaya* peel extract was collected from Chennai District (Tamil Nadu, India). The species were identified and authenticated at the Department of Centre for Advanced Study in Botany, University of Madras, Chennai, India. All chemicals and reagents used for this research work were purchased from Sigma Chemical Company St. Louis, MO, USA; Eurofins Genomics India Pvt Ltd, Bangalore, India; New England Biolabs(NEB), USA.

2.2 .Synthesis of Copper oxide nanoparticles using the peel of *Carica papaya* :

Fresh papaya peel is thoroughly washed and finely cut to increase surface area for the green synthesis of magnesium oxide nanoparticles. About 10 ml of the moist peel is mixed with 25 ml of distilled water and shaken for 45 minutes to extract bioactive compounds like flavonoids and phenolics, which act as natural reducing and stabilizing agents. On filtration,a clear extract of about 3 ml of magnesium nitrate solution is added to initiate nanoparticle formation. The mixture is stirred gently and color change occurs which indicates the reduction of copper ions and then this reaction mixture is centrifuged at 3000 rpm for 10 minutes to separate the copper oxide nanoparticles (pellet) from the supernatant, which is further discarded. Further the pellet is washed with distilled water and centrifuged again to purify the nanoparticles and unreacted compounds and impurities were removed . The final pellet is then dried overnight at 50°C , removing the moisture and stabilizing the nanoparticles and once it is dried, it is carefully collected and stored in airtight containers to prevent contamination. These dried copper oxide nanoparticles are used for characterization and are also tested for their antioxidant and antimicrobial properties which plays a major role in medical and dental application.

2.3 .Physico Chemical CharacterisationS

Scanning Electron Microscopy (SEM) provides high-resolution images to confirm the size, shape, and surface morphology of copper oxide nanoparticles (MgO NPs), which is crucial since smaller, uniform particles with larger surface areas exhibit enhanced antioxidant and antimicrobial activities. Ultraviolet–Visible (UV–Vis) spectroscopy is an analytical technique used to measure the absorbance of ultraviolet or visible light of a substance and help to confirm the formation of nanoparticles and is commonly used to monitor the reduction of copper ions and synthesize the nanoparticles by observing characteristic absorption peaks. Fourier Transform Infrared Spectroscopy (FTIR) is a technique used to identify the functional groups and molecular bonds present on the surface of nanoparticles. The FTIR helps to confirm the role of plant-derived phytochemicals in reducing and stabilizing the nanoparticles and also reveals characteristic absorption bands that correspond to functional groups such as hydroxyl (–OH), carbonyl (C=O), and amine (–NH) groups, which are commonly found in flavonoids, phenols, and proteins from the plant extract .

2.4. Anti-inflammatory assay

This anti-inflammatory assay is performed using the protein denaturation assay, mainly the Bovine Serum Albumin (BSA) Denaturation Assay, which is reliable and widely used for screening compounds that have anti-inflammatory properties. Diclofenac sodium was used as the positive control for the test. Further which a solution of Bovine Serum Albumin (BSA) was mixed with both the MgO nanoparticle solution and diclofenac solution separately and the pH was adjusted about 6.4 using dilute hydrochloric acid. The mixtures were then incubated at 37 °C for 20 min after which they were heated at 70 °C for 5 minutes to induce protein denaturation. After cooling, the solution is measured using a UV spectrophotometer at 660 nm.

2.5. Anti-microbial Assay

The antimicrobial assay was done using the agar well diffusion method. The fresh microorganism culture such as *S. aureus*, *E. coli*, *Enterococcus faecalis* which are bacterial species and *Candida albicans,* which is a fungal species, were grown and spread uniformly onto Mueller-Hinton Agar (MHA) for bacterial species and Sabouraud Dextrose Agar (SDA) for fungal species using swab method to create a lawn of microorganisms. Sterile cork borers were then used to create wells by punching into the agar surface. Two different concentrations, such as 25 µL and 100 µL of the green-synthesised magnesium oxide (MgO) nanoparticles from Carica papaya peel extract, were pipetted into the wells. Then the standard antibiotic disc was placed at the centre of each plate as a positive control and the plates were then incubated at 37 °C for 24 hours for bacterial strains and about 28–30 °C for fungal strains. After incubation, zones of inhibition were noted around the wells, which indicates antimicrobial activity. The diameters of these zones of inhibition were measured to evaluate the efficiency of the MgO nanoparticles in comparison with the standard antibiotic.

3. RESULTS AND DISCUSSION :

3.1. MgO NPs physicochemical properties:

The biosynthesis of MgO nanoparticles using *Carica papaya* peel extract is visually, clearly seen by the conversion from a turbid mixture to a clear supernatant and a dense, white pellet, culminating in a fine, uniform white powder characteristic of phase-pure MgO nanoparticles.The scanning electron microscopy (SEM) image reveals that the magnesium oxide (MgO) nanoparticles synthesized using *Carica papaya* peel extract are spherical and appear to be moderately aggregated, in the form of clusters or flower-like structures. The particles are fairly uniform in size and morphology, and the scale bar of 100 nm confirms that the particles are within the nanoscale range and the surface texture appears rough and dense, suggestive of effective formation and stabilization of nanoparticles and that confirms the successful green synthesis of MgO nanoparticles with desirable morphological characteristics suitable for further biological applications.

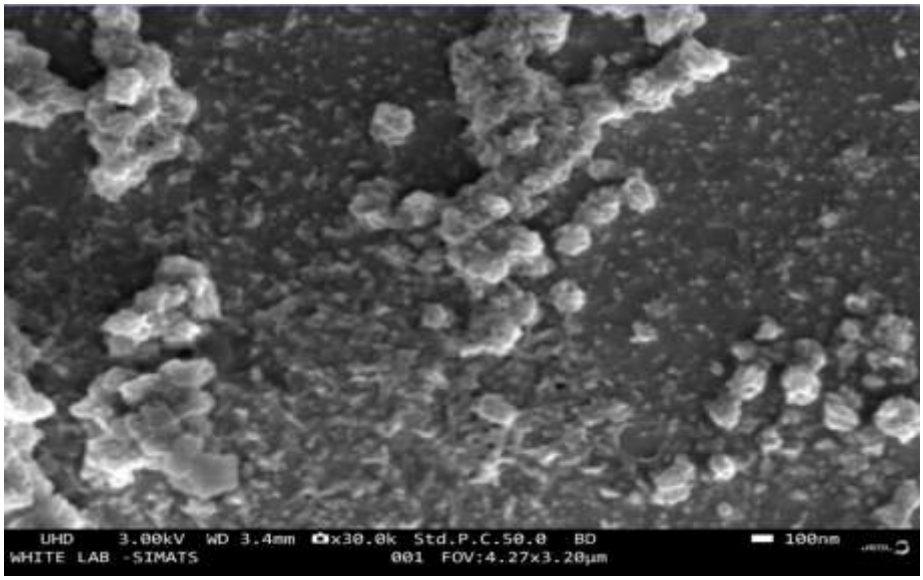
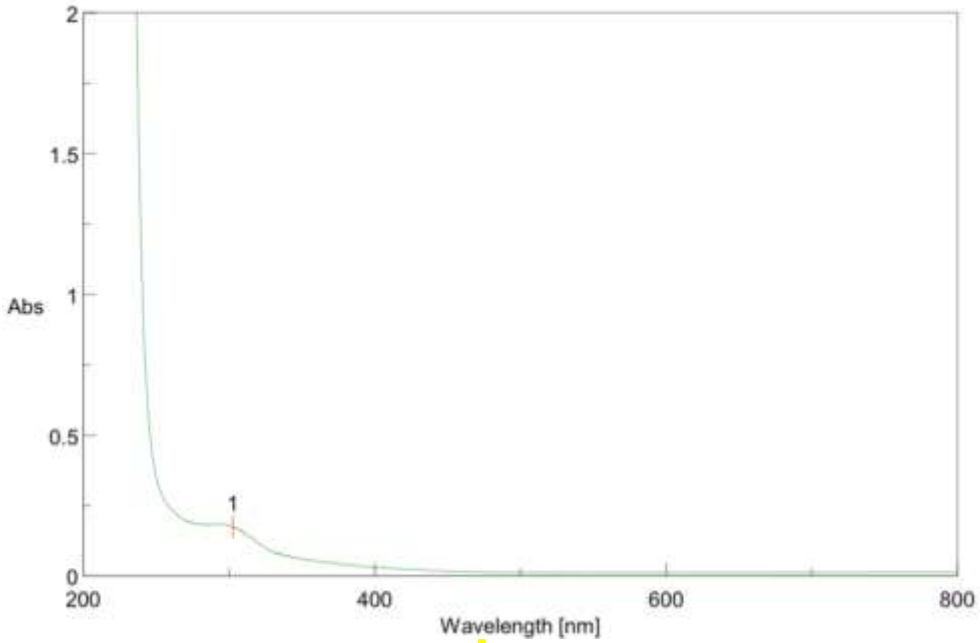


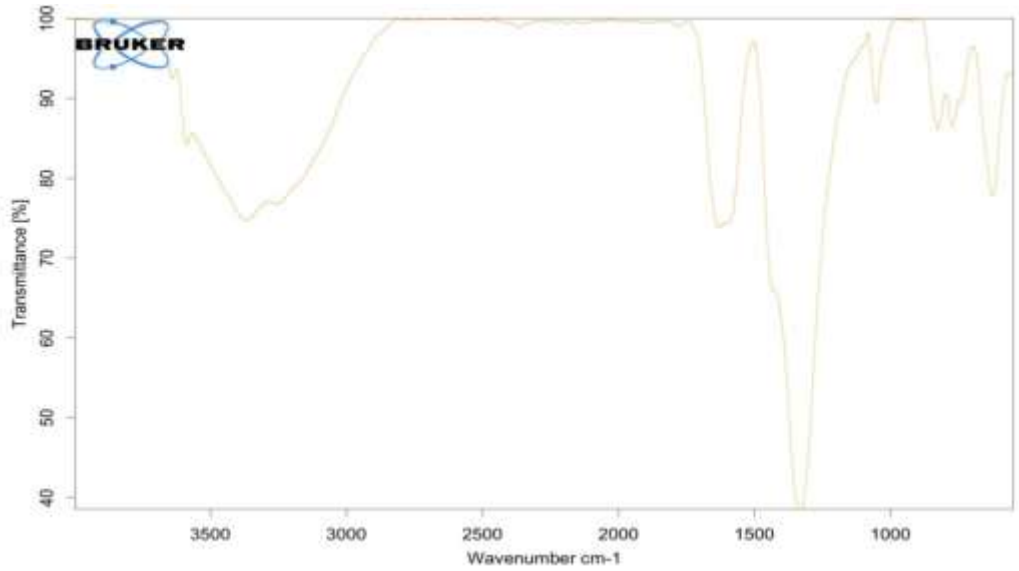
Fig. 1. Scanning electron microscopic image of MgO nanoparticles synthesized from *Carica papaya* peel extract

The UV-Vis analysis graph shows a strong peak around 302.5 nm, which confirms the presence of nanoparticles and also due to the reaction of nanoparticles when they hit light (Surface Plasmon Resonance) and they are a clear sign that the particles are at nanoscale. There is a steep decline in absorbance beyond this peak, suggesting the presence of a relatively uniform nanoparticle size with minimal aggregation and this in final supports the presence, stability and purity of biosynthesized MgO nanoparticles, validating the efficacy green synthesis process used. Further, the absorbance quickly goes down, which means the nanoparticles are not clumped together, which is a clear sign that the particles are stable and well-formed.



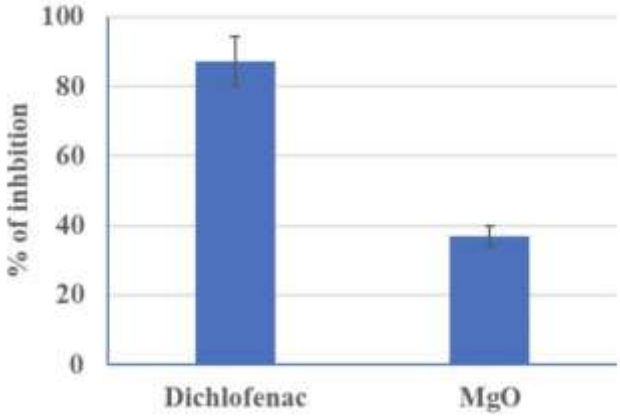
Graph 1. UV-Vis spectroscopy analysis

The FT-IR (Fourier Transform Infrared Spectroscopy) graph shows several peaks that shows the presence of key functional groups involved in the formation and stabilization of magnesium oxide (MgO) nanoparticles. A broad absorption band around 3400 cm^{-1} corresponds to O–H stretching vibrations indicating the presence of hydroxyl groups, likely from water or plant-based phenols. Peaks near 1630 cm^{-1} suggest C=O stretching from carbonyl groups, while those around 1400–1000 cm^{-1} may be attributed to C–O stretching and those below 1000 cm^{-1} belong to Mg–O bond vibrations confirming the formation of MgO nanoparticles. These functional groups originate from the Carica papaya peel extract, which acts as a natural reducing and stabilizing agent during green synthesis, helping in nanoparticle formation and preventing agglomeration.



Graph 2. Fourier transform infrared spectroscopy analysis

The anti-inflammatory graph presents a comparative analysis between the biosynthesized magnesium oxide (MgO) nanoparticles and the standard anti-inflammatory drug used is diclofenac sodium. Diclofenac shows a high inhibition rate of around 88–90%, showing its high anti-inflammatory potential. The MgO nanoparticles show a moderate inhibition of about 40% which indicates that while containing the anti-inflammatory activity, it is significantly decreased from the pharmaceutical standard, suggesting that the MgO nanoparticles do exhibit biological activity, due to the presence of phytochemicals obtained from papaya peel extract. But their effectiveness needs to be adjusted by changing the concentration or combining with other agents for therapeutic use.



Graph 3. Anti-inflammatory analysis

The antimicrobial analysis, above in the form of a tabular cold and image, denotes the efficiency of biosynthesized magnesium oxide (MgO) nanoparticles against the microorganisms Candida albicans, Staphylococcus aureus, Enterococcus faecalis, and Escherichia coli. The agar well diffusion method was used, where the wells were carefully made and about 25 µL and 100 µL concentrations of MgO nanoparticles were added into wells on inoculated agar plates to note the effectiveness of the nanoparticles, alongside standard antibiotics used as positive controls.

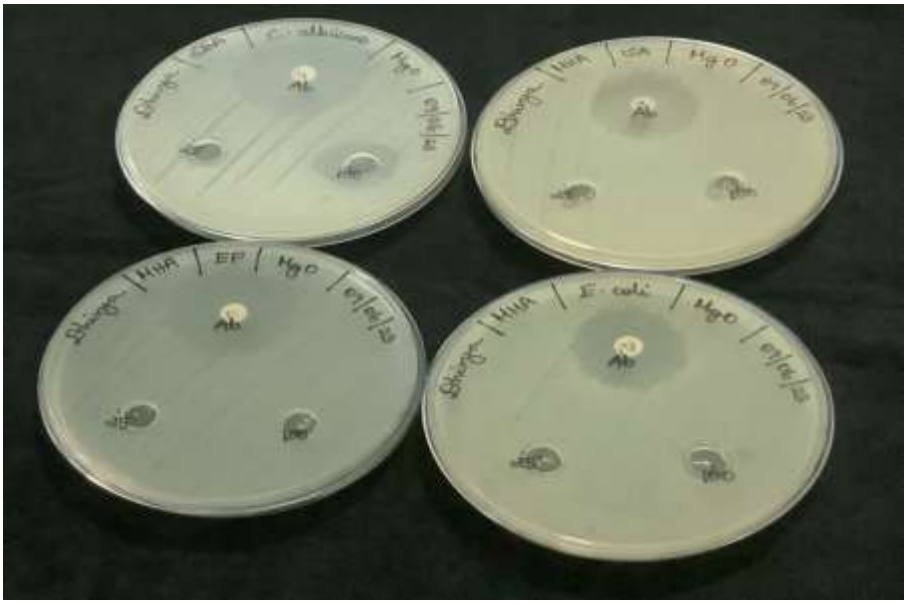


Fig. 2. Antimicrobial analysis

Table 1. Antimicrobial analysis

Micro organism	**Antibiotic**	**25**	**100**
Candida albicans	35mm	12mm	17mm
Staphylococcus aureus	25mm	-	11mm
Enterococcus faecalis	28mm	-	-
Escherichia coli	28mm	11mm	12 mm

The table denotes the measure of the diameter of zones of inhibition. The standard antibiotics control used showed significantly larger zones of inhibition across all organisms, ranging from 25 mm to 35 mm in the four discs and which confirmed their strong antimicrobial activity. The MgO nanoparticles exhibited mild antimicrobial activity that increased slightly with concentration. As we see in the above table, in *C. albicans*, the inhibition zones were 12 mm (25 µL) and 17 mm (100 µL), for *E. coli*, they were 11 mm and 12 mm, respectively. However, for *S. aureus* and *E. faecalis*, inhibition

was only noticeable at 100 µL (11 mm), with no observable effect at 25 µL. This proves that the magnesium nanoparticles have mild antimicrobial properties, but this increases with an increase in concentration.

DISCUSSION

The present study successfully demonstrates the green synthesis of magnesium oxide (MgO) nanoparticles using *Carica papaya* peel extract, through UV–Vis spectroscopy, FTIR analysis, and SEM imaging and the characterization results showed typical features of well-formed MgO nanoparticles, such as a sharp absorption peak at 302.5 nm, suggesting of surface plasmon resonance, and also FT-IR bands representing key functional groups responsible for reduction and stabilization. The SEM images revealed spherical, moderately dispersed nanoparticles. Henceforth, these structural and chemical characteristics support the efficacy of papaya peel extract as both a reducing and capping agent, confirming its potential for sustainable nanoparticle synthesis. However, when compared with other green synthesis approaches, there is moderate anti-inflammatory activity observed (~40% inhibition), suggesting that the phytochemical composition of *Carica papaya* is less compared to other medicinal plants rich in flavonoids, alkaloids, and polyphenols, which have shown stronger biological responses in other similar assays [28].

The present study FTIR analysis shows O–H, C=O, and metal–oxygen bands, which collaborate and develop a mechanism in which papaya phytochemicals act as both reducing and capping agents. This mechanism is similar to previous studies regarding ZnO and CuO, where similar functional groups were identified, which confirms the involvement of natural biomolecules in nanoparticle formation and stabilisation. These components reduce the ions to nanoparticles while also capping the nanoparticles to prevent aggregation. These findings support the idea that the phytochemical components of papaya peel have precise control over nanoparticle size, shape and surface characteristics during the synthesis[29][30].

Regarding the antimicrobial activity, the MgO nanoparticles displayed effective antibacterial activity, though the results were slightly lower when compared to other plants in other antimicrobial studies. This suggests that the efficacy of MgO nanoparticles depends heavily on the nature and concentration of bioactive compounds present in the source material. To improve the efficiency of nanoparticles, conditions such as temperature, pH and reaction time need to be maintained properly to get better quality nanoparticles and an increase in the concentration of nanoparticles for stronger effects to be achieved. Also, in combination with MgO nanoparticles with other natural compounds to improve the antimicrobial and anti-inflammatory activity [31].

The anti-inflammatory activity, MgO nanoparticles, shows about 40% inhibition of protein denaturation when compared with diclofenac, which shows 90% inhibition. Though it shows lower efficiency, the result is still significant as it shows that the bioactivity is influenced by phytochemicals from papaya peel. Previous studies regarding papaya peel mediates gold nanoparticles, which have been shown to have better anti-inflammatory and antioxidant effects, which supports the idea that the plant-derived components are effective. Thus, while MgO NPs are less effective than metallic counterparts, they retain meaningful bioactivity attributable to phytochemical capping. Incremental optimisation, such as controlled particle size adjustment, surface modification, or synergistic formulation, could enhance these therapeutic effects[32].

CONCLUSION

This study underlines the successful green synthesis of magnesium oxide (MgO) nanoparticles using *Carica papaya* peel extract, showing a sustainable and cost-effective alternative to conventional chemical synthesis methods. The characterization techniques, such as UV–Vis spectroscopy, FTIR, and SEM, confirmed the formation, structural integrity, and morphology of the nanoparticles. The presence of bioactive chemical compounds in the peel extract, such as proteolytic enzymes (papain and chymopapain), plays an important role in reducing the magnesium oxide NPS and keep it under control . These particles exhibited spherical morphology in the form of clustered aggregation or flower structure, with nanoscale dimensions confirmed through SEM analysis. FT-IR further validated the involvement of identifying the functional groups present in a sample, such as hydroxyl, carbonyl, and Mg–O functional groups, by measuring them by absorbing the infrared light at different wavelengths in the synthesis process.The synthesized MgO nanoparticles showed moderate anti-inflammatory activity and mild antimicrobial effects against both bacterial and fungal strains and their efficacy was less compared to standard control group compounds such as diclofenac. The results establish their potential as natural, biocompatible alternatives in biomedical applications. Further study needs to be done to optimize the nanoparticle formulation, such as varying extract concentrations, improving dispersion, and combining with other therapeutic agents to enhance their biological performance and expand their potential use in antimicrobial and anti-inflammatory treatments.

Credit authorship contribution statement

Dhivya Sarathi : Writing – original draft, Data curation, Conceptualization. Rajalakshmanan Eswaramoorthy: Conceptualization, Investigation, Data curation. Saranya: Investigation, Data curation. Palanivel Sathishkumar: Writing–review & editing, Validation, Supervision, Resources, Project administration, Methodology, Conceptualization.

Declaration of competing interest
The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability
Data will be made available on request.

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