

Phytochemical Profiling of Aqueous Extract of *Carica Papaya* Leaf Extract for Green Synthesis of Zinc, Magnesium and Calcium Nanoparticles

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ABSTRACT

Green synthesis of nanoparticles using plant materials has gained attention as a sustainable alternative to conventional methods involving hazardous chemicals, toxic reducing agents, and high-energy processes. Plant extracts contain phytochemicals that can function as reducing, chelating, capping, and stabilizing agents during nanoparticle synthesis. *Carica papaya* leaves are a readily available biological resource with potential for nanoparticle production. This study investigated the phytochemical profile of aqueous *Carica papaya* leaf extract for its potential application in the green synthesis of zinc, magnesium, and calcium nanoparticles. *C. papaya* leaves were collected from Agbarho, Ughelli North Local Government Area, Delta State, Nigeria, authenticated, washed, air-dried, pulverized, and extracted with distilled water at 85°C for one hour. The extract was subjected to qualitative phytochemical screening. The reddish-brown extract contained saponins, alkaloids, steroids, tannins, terpenoids, flavonoids, reducing sugars, and phenolic compounds, while anthraquinones were absent. Phenolic compounds, flavonoids, tannins, and reducing sugars possess functional groups capable of electron donation, metal-ion reduction, chelation, and stabilization. Other detected phytochemicals may contribute to nanoparticle capping and stabilization. The findings demonstrate that aqueous *C. papaya* leaf extract is a promising, simple, inexpensive, and environmentally sustainable biological resource for synthesizing zinc, magnesium, and calcium nanoparticles.

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INTRODUCTION

Nanotechnology has emerged as an important area of scientific research because of the unique physicochemical properties exhibited by materials at the nanoscale (Joudeh & Linke, 2022). Nanoparticles, generally described as particles with dimensions within the nanometre range, possess a high surface-area-to-volume ratio, enhanced reactivity, and distinctive optical, catalytic, antimicrobial, and biological properties (Obruch et al., 2019). These characteristics have encouraged their application in several fields, including medicine, agriculture, environmental remediation, food technology, cosmetics, and water treatment. Among the different types of nanoparticles, metal and metal oxide nanoparticles have received considerable attention because of their stability and diverse functional properties (Negrescu et al., 2022; Sarah et al., 2026). Zinc, magnesium, and calcium-based nanoparticles are particularly important because their constituent elements have considerable biological and industrial relevance (Odimbe et al., 2026). However, conventional approaches for synthesizing nanoparticles frequently involve high temperatures, toxic reducing agents, hazardous solvents, and substantial energy consumption, raising concerns about environmental pollution, production cost, and potential toxicity. Green synthesis has therefore gained increasing recognition as an alternative approach to

conventional nanoparticle production (Kirubakaran et al., 2026). Green synthesis employs biological materials such as plants, microorganisms, enzymes, and biomolecules as reducing, stabilizing, or capping agents during nanoparticle formation. Plant-mediated synthesis is especially attractive because plants are readily available, relatively inexpensive, and contain diverse phytochemicals capable of participating in nanoparticle formation (Nathan et al., 2025). Compounds such as flavonoids, phenolic compounds, alkaloids, tannins, terpenoids, saponins, glycosides, and proteins can donate electrons, chelate metal ions, facilitate their reduction or conversion, and subsequently stabilize the formed nanoparticles (Mikhailova, 2024). Consequently, the phytochemical composition of a plant extract is an important determinant of the efficiency, stability, morphology, and surface characteristics of nanoparticles produced through green synthesis. *Carica papaya* L., commonly known as papaya or pawpaw, is a tropical plant belonging to the family Caricaceae and is widely cultivated in many parts of the world, including Nigeria (Alara et al., 2022). Although its fruit is extensively consumed, the leaves have also attracted scientific interest because they contain a wide range of biologically active compounds. *C. papaya* leaves have been reported to contain several classes of phytochemicals, including phenolics, flavonoids, alkaloids, tannins, saponins,

terpenoids, glycosides, and other secondary metabolites. These compounds contribute to the biological activities associated with the plant and may also provide the functional groups required for the green synthesis of nanoparticles (Okpanachi et al., 2025). The aqueous extraction of *C. papaya* leaves is particularly relevant because water is inexpensive, readily available, environmentally benign, and avoids many of the hazards associated with organic extraction solvents. Furthermore, aqueous extraction may recover water-soluble phytochemicals capable of interacting with metal ions during nanoparticle synthesis (Etus and Erienu, 2026).

The use of *C. papaya* leaf extract as a biological reagent for nanoparticle synthesis is potentially advantageous because the same plant material may provide both reducing and stabilizing components (Joshi et al., 2023; Micheal et al., 2026). During synthesis, phytochemicals present in the extract can interact with metal precursor ions and promote nucleation and growth of nanoparticles. They may subsequently adsorb onto the nanoparticle surface and act as capping agents, limiting excessive aggregation and contributing to particle stability. The chemical composition of the extract is therefore closely connected to nanoparticle formation (Festus-Amadi et al., 2021). A detailed phytochemical profile can provide valuable information about the compounds potentially responsible for these activities and can improve understanding of the mechanism underlying plant-mediated nanoparticle synthesis (Edo et al., 2025). Zinc, magnesium, and calcium are particularly interesting nanoparticle-forming elements because of their broad applications and biological significance (Umudi et al., 2021). Zinc-based nanoparticles have attracted considerable attention because of their antimicrobial, catalytic, optical, and agricultural applications. Magnesium-based nanoparticles and magnesium-containing nanomaterials are also being investigated for biomedical, catalytic, and environmental applications, while calcium-based nanoparticles have relevance in areas such as biomedicine, agriculture, catalysis, and environmental technologies (Etus and Erienu, 2026). Despite these potential applications, nanoparticle synthesis must be carefully controlled because particle properties are influenced by precursor concentration, pH, temperature, reaction time, extract concentration, and, importantly, the phytochemical constituents of the biological reducing system (Edo et al., 2025). Although numerous studies have investigated plant-mediated synthesis of nanoparticles, an important research gap remains concerning the systematic relationship between the phytochemical composition of a single plant extract and its suitability for synthesizing different nanoparticles. Many studies report the successful synthesis of nanoparticles using plant extracts but provide limited information on the specific phytochemicals present in the extracts or their possible roles in nanoparticle formation. In addition, considerable attention has been given to individual nanoparticle types, particularly silver and gold nanoparticles, while comparatively fewer studies have systematically examined the potential of *C. papaya* leaf aqueous extract for the green synthesis of zinc, magnesium, and calcium nanoparticles within a related experimental framework. This creates a need to establish whether the phytochemical constituents of *C. papaya* leaves provide an appropriate biochemical basis for the formation and stabilization of these different nanoparticles (Joshi et al.,

2023; Jacintha et al., 2025). Therefore, this study focuses on the phytochemical profiling of aqueous *C. papaya* leaf extract as a basis for its application in the green synthesis of zinc, magnesium, and calcium nanoparticles. The study will identify and characterize the major classes of phytochemicals present in the aqueous extract and relate their presence to the potential reducing, chelating, and stabilizing activities required during nanoparticle formation. By linking phytochemical composition with green nanoparticle synthesis, the research seeks to address the existing knowledge gap concerning the biochemical basis of *C. papaya*-mediated synthesis of multiple metal nanoparticles. The findings are expected to contribute to the development of safer, simpler, cost-effective, and environmentally sustainable approaches to nanoparticle production and provide useful information for future investigations into plant-based nanotechnology.

MATERIALS AND METHODS

The reagents and materials required for this study include; Mayer's reagent, tollen's reagent, sulphuric acid (H_2SO_4), chloroform, HNO_3 , 10% ferric chloride solution, lead acetate solution. Materials are; 2 test tube racks, 2 plastic bowls, one (20 liters) plastic containers, digital camera, and funnels. All solutions were prepared using distilled water.

Preparation of Sampling Container

To obtain accurate results, proper sampling procedures were adopted as prescribed by Muthukumar et al., (2024) to eliminate potential contamination from the samples containers. Sample containers were washed with nitric acid, 0.1M HNO_3 , overnight, rinsed with distilled water, and air-dried. Sample containers were clearly and properly labeled to enhance record keeping. The control samples were labeled.

Collection, Preparation and Extraction of Pawpaw (*Carica papaya*) Plant Leaves

In order to carry out proper and effective collection, preparation and extraction, the extraction procedures and protocols outlined in the Standard Methods for extraction of plants by Ogwuche and Ibe, (2019) and Elemike et al., (2020) was thoroughly followed. Pawpaw (*Carica papaya*) plants leaves extract were collected from Agbarho town, in Ughelli North Local Government Area. It was authenticated by botanists in the Department of Botany, Delta State University, Abraka, Delta state. The plants leaves were washed with deionized water to remove dirty and impurities, air-dried at laboratory room temperature ($\sim 27^\circ\text{C}$) and in the absence of sunlight for 14 days as authorized by Elemike et al. (2021). Dried Pawpaw (*Carica papaya*) plant leaves extract was weighed, pulverized to smaller sizes using a domestic mortar to reduce the surface area, and further grinded to powder form using industrial blender. Concentration of the Dried Pawpaw (*Carica papaya*) plant leaves extract were made by weighing 16 g of the dried Pawpaw (*Carica papaya*) plant leaves and dunked inside 1000 mL of distilled water and heat for 1hr at 85°C , to obtain bioactive compounds concentrations. The homogenized solution was allowed to cool down naturally; the aqueous solution was gently screened through a Whatman filter paper (diam. 45 mm). The pure liquid (supernatant) solution was stored for further purposes as shown in figure 1.

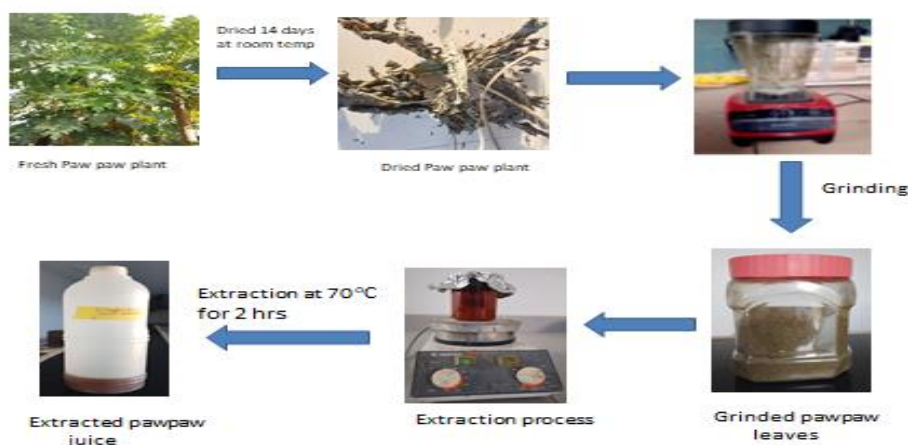


Figure 1: Flow diagram for the extraction of pawpaw plant leaves

Phytochemical Screening of the Plant Extract

The phytochemical screening processes were similar to work of Ogwuche and Ibe, (2019); with a few minor modifications. The aqueous extract of the pawpaw leaves was subjected to phytochemicals screening as shown in figure 2. This process would allow to know if the aqueous extracts contains the required phytochemical to synthesize the nanoparticles.

Test for Alkaloids

To about 0.5 g of the aqueous extract pawpaw, two drops of Mayer's reagent were added along the sides of test tube. Appearance of white creamy precipitate was observed which indicated the presence of alkaloids.

Test for Reducing Sugar

To about 0.5 g of the aqueous extract in the test tube, tollen's reagent was added and heated moderately. The presence of silver mirror on the walls of the test tube indicates the presence of reducing sugars.

Test for Steroids

0.5 g of the aqueous extracts was taken in a test tube and dissolved with chloroform (10 mL), and then three drops of concentrated sulphuric acid was added by the side of the test tube. Result showed negative to steroids. Positive result should give two layers. The upper layer should give red and sulphuric acid layer should give yellow with green fluorescence.

Test for Terpenoids

0.5 g of extract was mixed in 2 mL of chloroform and 3 mL of concentrated H_2SO_4 was carefully added to form a layer. A reddish brown colouration of the interface indicate the

presence of terpenoids. Meanwhile, in this work, it was found negative.

Test for Saponin

To about 0.5 g of the aqueous extract in the test tube was added 5 mL of distilled water. It was shaken vigorously. Persistent foam for about 2 minutes confirmed the presence of saponin.

Test for Anthraquinones

0.5 g of the aqueous extract was boiled with 1 mL concentrated HNO_3 and shaken well. A dark orange colour indicates the presence of anthraquinones on addition of the concentrated acid. Meanwhile, in this work, result obtained showed negative.

Test for Phenolic Compounds

To 0.5 g of the aqueous extract, few drops of 10% ferric chloride solution were added. Green blue colour appears which indicated the presence of phenolic compounds.

Test for Tannins

About 0.5 g of the aqueous extract, 5% ferric chloride solution was added, a dark green precipitate was observed which showed the presence of tannins in the plant.

Test for Flavonoids

To 0.5 g of aqueous extract was added little quantity of water. A few drop of lead acetate solution was added and a light yellow precipitate was observed which indicated the presence of flavonoid.

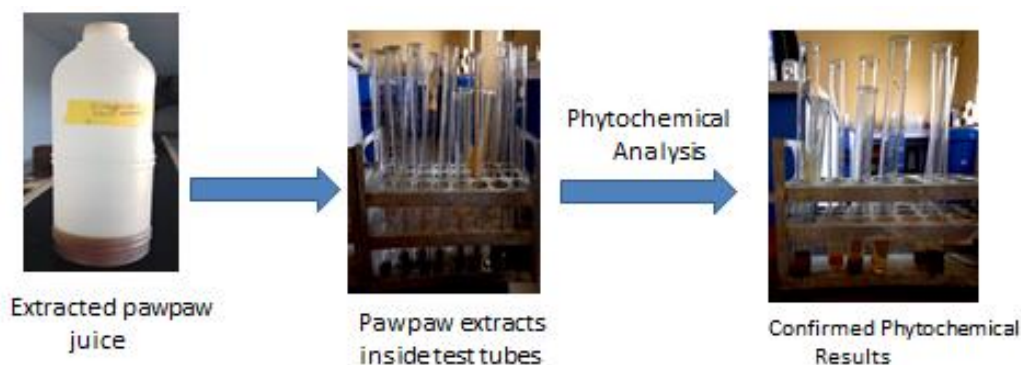


Figure 2: Flow diagram for the Phytochemical screening of pawpaw plant leaves

RESULTS AND DISCUSSION

This section presents the analyzed and characterized results of the phytochemical screening as presented according to the specific objective sited in this study.

Phytochemical Characterization of Aqueous Plant Extract of Pawpaw Leaf

The result of the phytochemical experiment on the aqueous extract of pawpaw leaf extract reveals the presence of some phytochemicals is presented in Table 1. The results of the

phytochemical analysis on pawpaw leaf extract. Result showed that the presence of saponins, alkaloids, tannis, phenolic compounds, flavonoids and reducing sugar which may be responsible for the reduction of metallic acetate precursors. It is believed that the plant extract has information to give in the bio-reduction of the metal precursor in the synthesis of nano-metal particles (Hadjilambrinos et al., 2024). Similar result was obtained in the findings of Kristanti et al. (2021) on the phytochemical characterization of pawpaw leaf extract.

Table 1: Phytochemical Analysis of Aqueous Extract Pawpaw Leaves

Tests Performed	Obtained Results
State	Liquid
Colour	Reddish brown
Saponin	+
Alkaloids	+
Steroids	+
Tannins	+
Terpenoids	+
Flavonoids	+
Reducing Sugar	+
Phenolic compounds	+
Anthraquinones	-

Keys; (+) detected, (-) Not detected

Discussion of Phytochemical Characterization of Aqueous *Carica papaya* Leaf Extract

The phytochemical analysis of the aqueous extract of *Carica papaya* (pawpaw) leaves revealed the presence of several important secondary metabolites, as presented in Table 1. The extract was reddish-brown and remained in liquid form after extraction. The detected phytochemicals included saponins, alkaloids, steroids, tannins, terpenoids, flavonoids, reducing sugars, and phenolic compounds, while anthraquinones were not detected. The broad range of phytochemicals identified indicates that *C. papaya* leaves contain diverse bioactive constituents that may contribute significantly to their biological and nanoparticle-synthesis potentials.

Saponins were detected in the extract. Saponins are amphiphilic compounds containing hydrophilic and hydrophobic portions and are known for their surface-active properties. Their presence may be important during green nanoparticle synthesis because they can interact with metal ions and nanoparticle surfaces, thereby contributing to stabilization and reducing particle aggregation. Similarly, the detection of alkaloids indicates the presence of nitrogen-containing secondary metabolites with considerable biological activity. Alkaloids possess functional groups capable of interacting with metal ions and may participate indirectly in the reduction and stabilization processes involved in nanoparticle formation. Similar result was obtained in the findings of Kristanti et al. (2021) on the phytochemical characterization of pawpaw leaf extract.

Tannins and phenolic compounds were also detected. These compounds are particularly significant because they contain hydroxyl and other functional groups capable of donating electrons. Such electron-donating properties can facilitate the conversion of metal ions from their precursor salts into nanoscale materials. Phenolic compounds are also recognized for their antioxidant activity, which makes them important candidates for the reduction of metallic precursors. Their adsorption onto newly formed nanoparticle surfaces may further provide a protective coating that enhances colloidal stability. Thus, the presence of tannins and phenolics supports

the potential suitability of the aqueous *C. papaya* leaf extract as a green reducing and capping system. Flavonoids were detected in the extract and may also contribute substantially to nanoparticle synthesis. Flavonoids contain several hydroxyl groups and conjugated structures that can participate in electron transfer and metal chelation. Their presence may therefore facilitate nucleation and growth of nanoparticles while simultaneously helping to stabilize the resulting particles. The detection of reducing sugars is similarly important because reducing sugars can donate electrons to metal ions, potentially contributing to their reduction during the synthesis process. The extract also tested positive for steroids and terpenoids. Although their direct reducing capacity may differ from that of phenolics and flavonoids, these compounds possess functional groups capable of interacting with metal ions and nanoparticle surfaces. They may therefore contribute to surface stabilization and modification of the synthesized nanoparticles. In contrast, anthraquinones were not detected, indicating that these compounds are unlikely to play a significant role in the proposed bio-reduction process.

Overall, the presence of multiple phytochemical classes demonstrates that aqueous *C. papaya* leaf extract contains a complex mixture of potential reducing, chelating, and stabilizing agents. This supports its application in the green synthesis of zinc, magnesium, and calcium nanoparticles. The findings are consistent with the observations reported by Kristanti et al. (2021), who identified several phytochemical constituents in *C. papaya* leaf extracts. The result also agrees with the general observation that plant-derived phytochemicals can facilitate nanoparticle formation through their reducing and stabilizing activities (Hadjilambrinos et al., 2024). Importantly, the present findings provide a biochemical basis for further investigation of the extract in the synthesis and characterization of the targeted nanoparticles.

CONCLUSION

The phytochemical characterization of the aqueous extract of *Carica papaya* leaves demonstrated the presence of a broad range of bioactive constituents, including saponins, alkaloids, steroids, tannins, terpenoids, flavonoids, reducing sugars, and phenolic compounds, while anthraquinones were not detected. The reddish-brown colour and liquid nature of the extract further indicate the successful extraction of soluble plant constituents. The diversity of phytochemicals identified suggests that *C. papaya* leaves possess considerable biochemical potential for green nanoparticle synthesis. The detected phenolic compounds, flavonoids, tannins, and reducing sugars are particularly important because their functional groups can participate in electron donation, metal-ion reduction, and chelation. These properties may facilitate the conversion of metallic acetate precursors into nanoparticles. In addition, saponins, alkaloids, steroids, and terpenoids may contribute to the stabilization and capping of newly formed nanoparticles, thereby helping to reduce particle aggregation and improve their stability. The absence of anthraquinones does not substantially diminish the potential of the extract because several other phytochemical classes capable of supporting nanoparticle formation were detected. Overall, the findings establish that aqueous *C. papaya* leaf extract contains a combination of phytochemicals capable of acting as natural reducing, chelating, and stabilizing agents. The result therefore provides a strong biochemical basis for employing the extract in the environmentally friendly synthesis of zinc, magnesium, and calcium nanoparticles. The study further demonstrates the potential of *C. papaya* leaves as an inexpensive and sustainable biological resource for nanoparticle production, while subsequent characterization of the synthesized nanoparticles is necessary to confirm their size, morphology, composition, crystallinity, and stability.

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