

Phyto Constituents of *Ananas comosus* Leaf Extract Enhancing the Copper Nanoparticles Synthesis

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ABSTRACT

Ananas comosus leaf is one of the agricultural wastes that has resulted in environmental pollution. Recently, this waste has been explored for its potential in synthesising environmentally friendly metallic nanoparticles. *Ananas comosus* leaf extract has been proposed as a viable and economical option for reducing, capping, and stabilising agents in synthesising copper nanoparticles. This study used *Ananas comosus* leaf extract and copper sulphate as precursor materials to produce copper nanoparticles in the green synthesis process. The results show that the absorption spectra in the UV-visible range exhibit a peak absorption value at a wavelength of 238 nm. Fourier Transform Infrared analysis revealed the presence of bioactive compounds in the *Ananas comosus* leaf extract,

which are responsible for reducing Cu^{2+} ions to Cu^0 nanoparticles. The Field Emission Scanning Electron Microscope analysis indicated that the copper nanoparticles have a face-centred cubic crystal structure, which is significant for innovative nanoparticle applications. The average particle size was determined to be 56.71 nm. X-ray diffraction analysis revealed peaks at 43.46° , 50.57° , and 74.23° , corresponding to the (111), (200), and (220) lattice planes, respectively, confirming the structural properties of the synthesised nanoparticles. Energy Dispersive X-ray examination found that the

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synthesised copper nanoparticles are primarily composed of copper, accounting for 95.24% of the total composition. The phytochemicals in the extract have been effectively utilised in the green synthesis of copper nanoparticles, demonstrating potential applications in medicine and agriculture.

Keywords: *Ananas comosus*, copper nanoparticles, Energy Dispersive X-ray, Field Emission Scanning Electron Microscope, Fourier Transform Infrared, green synthesis, UV-visible, X-ray diffraction

INTRODUCTION

Ananas comosus is a botanical species characterised by its tropical origins and fruit, suitable for human consumption (Ali et al., 2020). The MD2 variety of *Ananas comosus* is widely cultivated in Malaysia due to its significant popularity in global markets (Ramli, 2020). The annual global harvest of *Ananas comosus* fruit for consumption and use in the food and beverage sectors amounts to around 30 million tonnes, which also generates a significant quantity of *Ananas comosus* waste (van Tran et al., 2023). The primary waste components of *Ananas comosus* consist of the crown, peel, stem, and core (Zulpahmi et al., 2023). Agricultural industries worldwide generate substantial quantities of biomass residues, posing environmental challenges. These waste materials frequently end up in landfills, where a significant portion is converted into greenhouse gases and methane (van Tran et al., 2023).

Therefore, biotechnological and adaptable methodologies can ensure the sustainable use of by-products derived from *Ananas comosus* (Fouda-Mbanga & Tywabi-Ngeva, 2022). This is primarily due to the abundant presence of phytochemicals, antioxidants, biofuels, and phenolic compounds (Sarangi et al., 2022). According to Vijayaram et al. (2023), green synthesis tools are considered more appropriate for developing nanoparticles within the 1 to 100 nm size range. Green synthesis methods are increasingly replacing physical and chemical processes as they utilise natural and eco-friendly substances (Ying et al., 2022). Particle size affects various features and functions (Buniyamin et al., 2022). Metal nanoparticles can be synthesised in the presence of green materials and under appropriate conditions (Vijayaram et al., 2023). In previous studies, several metallic nanoparticles have been synthesised using plant extracts.

For instance, zinc oxide nanoparticles (ZnO NPs) have been synthesised using the unripe fruit extract of *Aegle marmelos* (Am-ZnO NPs), the leaf extract of *Lawsonia inermis*, whereas iron oxide nanoparticles have been produced with the assistance of *Tamarindus indica* fruit extract (Senthamarai & Malaikozhundan, 2022; Vinothini et al., 2023; Malaikozhundan et al., 2024). The current status of bio-assisted synthesis primarily encompasses various interconnected factors, including the composition and concentration of the biological reducing agent, the starting concentration of precursor salts, the stirring speed, the reaction duration, the pH level, temperature, and exposure to light. These factors

influence the properties of the synthesised nanoparticles. Therefore, optimising a biological methodology is a comprehensive undertaking that requires a substantial investment of time and resources (Crisan et al., 2022). Among various metal nanoparticles, copper stands out from others, such as gold, silver, and zinc, due to its larger surface area, cost-effective production, and notable antibacterial and antioxidant properties (Malaikozhundan, Krishnamoorthi et al., 2022). Copper nanoparticles have received significant attention from the public (Crisan et al., 2022). They include distinctive characteristics that render them highly relevant in many domains, including their utilisation as antioxidants, anticancer agents, antibacterial agents, nanosensors, nanocatalysts, and materials with exceptional strength (Alahdal et al., 2023). These nanoparticles' production has been achieved by reducing aqueous copper ions using various plant extracts (Hano & Abbasi, 2022). The wide range of accessible oxidation states of copper (Cu^0 , Cu^+ , Cu^{2+} , and Cu^{3+}) allows for diverse reactions and promotes reactivity in copper-based materials.

This reactivity can occur through both one- and two-electron pathways (Alahdal et al., 2023). Several protective compounds have been recommended to mitigate the process of oxidation (Alahdal et al., 2023). Capping agents play a crucial and diverse role in the synthesis of nanoparticles. Plant extracts contain a wide range of compounds, including amines, amides, alkaloids, flavonoids, phenols, terpenoids, proteins, and pigments (Thatyana et al., 2023). Previous studies have also shown the presence of various phytochemical compounds in different parts of *Ananas comosus*, as portrayed in Table 1. The bioactive compounds used as reducing and capping agents during nanomaterial synthesis were identified to assess the rate of the mechanism, surface morphology, and size of the nanoparticle, along with its stability (Buniyamin, Halim et al., 2023). Therefore, using plant extracts presents a viable approach for efficiently forming metallic nanoparticles through environmentally friendly methodologies (Alahdal et al., 2023).

The use of plants as reducing agents in the synthesis of nanoparticles effectively eliminates harmful toxic substances (Malaikozhundan, Lakshmi et al., 2022). The secondary metabolites derived from plants contribute to the stability of nanoparticles, prevent agglomeration, and facilitate control over their shape and size (Sidhu et al., 2022). Additionally, biomolecules present during the synthesis step can prevent particle agglomeration, resulting in dispersed nanoparticles and a high surface area-to-volume ratio (Buniyamin, Mahmood et al., 2023).

To the best of our knowledge, previous research has concentrated on copper nanoparticles derived from fresh aqueous extracts of *Ananas comosus* fruit and peel (Ranjitham et al., 2015; Mitra et al., 2024), neglecting other parts of *Ananas comosus*. Therefore, this study aims to focus on the novelty of the *Ananas comosus* leaf by utilising it for the synthesis of copper nanoparticles that enhance environmental sustainability and renewability due to its abundance compared to the fruit, which is predominantly utilised in

the food and beverage industry. This effort facilitates the exploration of new opportunities for significant impact, applications, or future additional research. These findings help Malaysia enhance its scientific research in nanotechnology while addressing the issue of excessive waste generated from the *Ananas comosus*.

Table 1
Phytochemical compound of Ananas comosus

| Plant Part | Chemical Compounds | References |
|------------|---|--|
| Fruits | Alkaloids: Phenolamine, piperidine, plumerane, pyridine, pyrrole, quinoline Flavonoids: Flavonols, flavonones, flavones, flavonoid carbonoside, flavanols Lipids: Free fatty acids, glycerol ester, lysophosphatidylcholine, sphingolipids Amino acids and derivatives, proteins Lignans and coumarins Nucleotides and derivatives Organic Acids: Acetic acid, citric acid, oxalic acid, malic acid Phenolic Acids: Chlorogenic acid, gallic acid, gentisic acid, syringic acid, vanillin, ferulic acid, sinapic acid, isoferulic acid, o-coumaric acid polyphenols: gallic acid, catechin, epicatechin, ferulic acid saponins, tannins, steroids, terpenoids, naphthoquinone, inulin, phenols Bromelain, other compounds: 1-hexanol, nonanal | Chen et al., 2023; Rahman et al., 2020; Hikal et al., 2021; Spanier et al., 1998; Gao et al., 2022; Salome et al., 2011 |
| Stem | Flavonoids, alkaloids, saponins, tannins, phytosterol, carbohydrates, bromelain, proteins | Febrianti et al., 2016; Rahman et al., 2020 |
| Peel | Protein, alkaloids, tannins, flavonoids, steroids, saponins, terpenoids, inulin, glycosides, phenolic compounds, polyphenols, anthraquinones, coumarins, triterpenes, sterols, oxalate, phytate, quinine, carbohydrate, bromelain | Rahman et al., 2020 |
| Crown | Bromelain, protein: Ribulose biphosphate carboxylate, peroxidase, acid, alkaline phosphatase, nuclease, α -amylase Polyphenols: Ferulic acid, syringic acid | Rahman et al., 2020 Hikal et al., 2021 |
| Pulp | Esters: Methyl hexanoate, Ethyl hexanoate, Methyl 3-(methylthio) propanoate, Methyl octanoate, Ethyl deanoate Aromatic compounds: A-terpineol, nonanal, decanal | Teai et al., 2001 |
| Shell | Phenolic compounds: Myricetin, salicylic acid, tannic acid, trans-cinnamic acid, p-coumaric acid | Hikal et al., 2021 |
| Core | Ascorbic acid, phenolic acids: Ferulic acid, syringic acid | Hikal et al., 2021 |
| Leaves | Flavonoids, terpenoids, phytosterol, amino acids, phenols, tannins, carbohydrates, glycosides, proteins, alkaloids, saponins, triterpenoids, steroids, citric acids, bromelain Phenolic compounds: Caffeic acid, chlorogenic acid, p-coumaric acid | Rahman et al., 2020; Hikal et al., 2021 Ma et al., 2007 |

MATERIALS AND METHOD

Oven (Brand: Memmert/UF260), soil crusher with sieve (Brand: Retsch) to grind the dried chopped leaf, copper (II) sulphate (Brand: Sigma Germany) as the precursor salt, Whatman No. 1 filter paper (Brand: Cytiva) to filter the extract, glass Petri dish to dry the mixture of copper nanoparticle solution, universal bottle to store the dried copper nanoparticles, high-speed refrigerated centrifuge (Brand: Hitachi) and centrifuge tubes to centrifuge the *Ananas comosus* leaf extract and colloidal solution of copper nanoparticles, UV-Visible Diffuse Reflectance Spectroscopy (DRS) to measure the optical properties of copper nanoparticles (Model: Cary 5000 Varian, using a PbSmart detector and controlled by Cary WinUV software), X-ray diffraction (XRD) operated using the PANalytical X'pert PRO XRD model with CuK α radiation ($\lambda=1.17545 \text{ \AA}$) at ca. $2\theta = 5^\circ\text{-}90^\circ$ with 45 kV and a scan speed of $0.417782^\circ/\text{sec}$ at 40 mA, with a 2 degree/min setting, Fourier Transform Infrared (FTIR) Spectrophotometer measured using the Perkin Elmer Spectrum 400 within the region 400 to 4000 cm^{-1} by the Attenuated Total Reflectance (ATR) technique, Field Emission Scanning Electron Microscope (FESEM) (Brand: Thermo Scientific Apreo 2) and Energy Dispersive X-ray (EDX) microanalysis (Brand: Thermo Scientific Apreo 2) to analyse the structure, shape, and characteristics of synthesised copper nanoparticles, and OriginPro 2024 (Graphing and Analysis) software for plotting graphs of results obtained.

Preparation of Leaf Extract

The preparation of *Ananas comosus* leaf extracts was conducted with minor modifications and the inclusion of an additional method. The *Ananas comosus* leaf, obtained from a plantation owned by the Malaysian Pineapple Industrial Board (MPIB) located in Chin Chin, Melaka, Malaysia, was subjected to a washing process, followed by chopping and subsequent drying at a temperature of 60°C (Hamdiani & Shih, 2021). The dried leaf was ground into a fine powder. The leaf extract was prepared by combining 20 g of powdered leaf with 200 mL of distilled water in a 250 mL conical flask. The mixture was then boiled and stirred constantly using a magnetic stirrer on a hot plate set at 80°C for 30 minutes (Olajire & Mohammed, 2019). After boiling, the mixture was cooled and subsequently centrifuged at 15,000 revolutions per minute (rpm) to separate and eliminate sediment. The filtrate was then subjected to three rounds of filtration using Whatman No. 1 filter paper to enhance clarity (Hassanein et al., 2018). The sample was stored at 4°C in a Schott bottle for subsequent use in synthesising copper nanoparticles (Hamdiani & Shih, 2021).

Phytochemical Screening of *Ananas comosus* Leaf Extract

The extract was used for phytochemical analysis following established frameworks, with minor modifications to the quantities of chemicals and extract used. The qualitative tests for secondary metabolites were conducted as follows in Table 2.

Table 2

Chemical tests for various compounds in Ananas comosus leaf extract

| Test | Procedure | Indication of Presence | References |
|---------------|---|---|------------------------|
| Flavonoid | Add leaf extract to 1.5 mL of diluted NaOH. | Yellow precipitate formation | Hassan et al., 2020 |
| Phenols | Mix 2 mL of leaf extract with a small amount of 10% aqueous ferric chloride. | Blue or green colour | Rajkumar et al., 2022 |
| Tannins | Add ferric chloride to 2 mL of leaf extract. | Black or brownish-blue colour | Rajkumar et al., 2022 |
| Saponins | Mix 1 mL of leaf extract with 5 mL of water and shake rapidly. | Formation of significant lather | Hassan et al., 2020 |
| Steroids | Add 1 mL of leaf extract to 1 mL of glacial acetic acid, 1 mL of acetic anhydride, and 2 drops of concentrated H ₂ SO ₄ . | Colour change from red to bluish-green | Godlewska et al., 2022 |
| Alkaloids | Mix 1 mL of leaf extract with Dragendorff's reagent. | Orange colour | Godlewska et al., 2022 |
| Quinones | Mix 1 mL of leaf extract with 1 mL of concentrated H ₂ SO ₄ . | Red colour | María et al., 2018 |
| Terpenoids | Mix 5 mL of leaf extract with 2 mL of chloroform, then add 3 mL of concentrated H ₂ SO ₄ . | The reddish-brown colour at the interface | Dubale et al., 2023 |
| Carbohydrates | Mix 5 mL of Benedict reagent with leaf extract, boil for 2 minutes, then cool. | Red precipitate | Pooja et al., 2022 |

Synthesis of Copper Nanoparticles Using *Ananas comosus* Leaf Extract

The method used was based on recent research by Ranjitham et al. (2015) with minor modifications. Copper nanoparticles were synthesised by dissolving 1.6 g of copper (II) sulphate in 500 mL of distilled water to generate a stock solution (0.02 g/mL) with a blue colouration. Subsequently, 200 mL of prepared *Ananas comosus* leaf extract was introduced into the aqueous solution of copper sulphate. The reaction mixture was subjected to vigorous agitation using a water bath shaker, and the colour changes of the colloidal solution were used as evidence of the presence of synthesised copper nanoparticles. The mixture then underwent a separation process using centrifugation, followed by two rounds of rinsing with distilled water until the filtrate achieved purity. The copper nanoparticles were subsequently dried at a temperature of 60°C for 24 hours, resulting in powdered copper nanoparticles. The specimen was kept for subsequent characterisation analysis using UV-visible diffuse Reflectance Spectroscopy (DRS), X-ray Diffraction (XRD), Energy Dispersive X-ray Spectroscopy (EDX), Fourier Transform Infrared Spectroscopy (FTIR), and Field Emission Scanning Electron Microscopy (FESEM). The parameters for

this research were fixed as outlined below. At the end of the investigation, all results with graphs and peaks were plotted using OriginPro 2024 (Graphing and Analysis) software. The parameters listed in Table 3 were based on the optimal results obtained in the prior study by Ranjitham et al. (2015).

Table 3

Experimental parameters for the synthesis of copper nanoparticles using Ananas comosus leaf extract

| Parameter | Details |
|---------------|--|
| Composition | 1:4 volume ratio of copper sulphate aqueous solution to <i>Ananas comosus</i> leaf extract used for synthesising copper nanoparticles. |
| Temperature | Synthesis was conducted at 80°C in a water bath. |
| Time Interval | The formation of a colloidal solution of synthesised copper nanoparticles was recorded at 60 minutes. |

RESULTS AND DISCUSSION

Phytochemical Screening of *Ananas comosus* Leaf Extract

The findings of the phytochemical analysis of the aqueous extract derived from *Ananas comosus* are presented in Table 4. The results demonstrate the presence of secondary metabolites, including phenol, tannin, saponin, steroid, alkaloid, quinone, anthraquinone, terpenoid, carbohydrates, coumarin, and flavanone. Positive (+) and negative (-) marks are used to denote the presence and absence of specific phytochemical elements, respectively. Phenolic compounds are significant as phytoconstituents due to their inherent capacity to act as reducing agents in redox reactions (Amini & Akbari, 2019). They can be categorised into distinct groups, including coumarins and quinones (Gan et al., 2019).

Secondary metabolites derived from plants, such as steroids, saponins, tannins, terpenoids, polyols, alkaloids, flavonoids, and phenolics, exhibit robust detoxifying, reducing, and stabilising properties (Sidhu et al., 2022). Previous studies have also demonstrated that carbohydrates in plant extracts stabilise and reduce agents during nanoparticle formation (Rani et al., 2023). This assertion is supported by prior scholarly

Table 4

Phytochemical screening of Ananas comosus leaf extract

| Phytochemicals | Phenol | Tannin | Saponin | Steroid | Alkaloid | Quinone | Terpenoid | Carbohydrates | Coumarin | Flavanone |
|----------------|---------------|--------|---------|---------|----------|---------|-----------|---------------|----------|-----------|
| | Results (+/-) | + | + | + | - | + | - | + | + | + |
| | - | - | - | + | - | + | - | - | - | - |

investigations, which have shown the use of phenolic acids derived from plants in the green synthesis of nanoparticles. Nanoparticle synthesis involves the use of reducing agents and stabilisers, which play dual roles in the process (Amini & Akbari, 2019). Therefore, this qualitative screening aids in identifying the phytochemical compounds present in the extract of *Ananas comosus* leaf.

Visual Observation of Synthesised Copper Nanoparticles Using *Ananas comosus* Leaf Extract

The formation of copper nanoparticles was detected through a discernible alteration in the colour of the reaction mixture. Initially, the copper sulphate solution exhibited a pale blue colour. When the leaf extract of *A. paniculata* was added, the copper sulphate solution underwent a colour transformation from light blue to green, eventually turning a dark greenish-brown colour due to the synthesis of copper nanoparticles (Rajeshkumar et al., 2021). This study demonstrated that the blue aqueous copper sulphate solution changed to dark greenish-brown and eventually dark brown upon the introduction of *Ananas comosus* leaf extract into the aqueous solution of copper sulphate, given the heat and time required for synthesising copper nanoparticles, a pink precipitate formed at the base of the conical flask by the end of the synthesis process. The initial observation in this study was the presence of a pink precipitate consisting of colloidal copper nanoparticles. Previous investigations have shown that colloidal copper solutions' pink to violet colour is directly associated with the observed copper surface plasmon band (Bárta et al., 2010). Subsequently, this precipitate was dried in an oven, forming black powdered copper nanoparticles. The powdered nanoparticles were then subjected to characterisation analysis using spectroscopy, microscopy, and diffraction techniques.

UV-visible Spectrophotometry

A diffuse reflectance analysis was conducted to obtain the spectral data of reflection performance. As a result, the energy band gaps of the nanoparticles could be detected (Buniyamin et al., 2022). The investigation focused on copper nanoparticles synthesised using *Ananas comosus* leaf extract, with the analysis performed using UV-visible diffuse Reflectance Spectroscopy (DRS). The black powdered copper nanoparticles were evaluated based on their reflectance activity, as measured by UV-visible diffuse reflectance (UV-visible DRS). The synthesised copper nanoparticles exhibited surface plasmon resonance (SPR) spectra with absorption peaks ranging from 200 to 800 nm. The presence of copper nanoparticles was verified by observing a distinct peak at 238 nm using a UV-visible diffuse reflectance (UV-visible DRS) spectrophotometer. The data were plotted using OriginPro 2024 (Graphing and Analysis) software, as depicted in Figure 1. A prominent peak in absorbance was detected, attributed to the ionisation of phenolic groups found

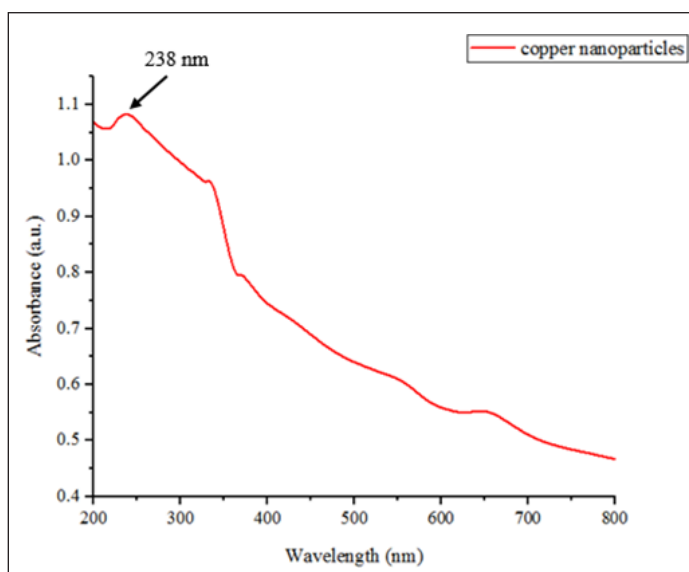


Figure 1. The UV-visible diffuse Reflectance Spectroscopy (DRS) of synthesised copper nanoparticles using *Ananas comosus* leaf extract

within the plant extract. Copper nanoparticles typically demonstrate a distinct surface plasmon resonance peak within the wavelength range of 560–570 nm (Mohamed, 2020). The observed variations in reduction, capping, and stabilising processes can be attributed to the diverse properties of the plant extracts utilised. It may also be caused by using different types of UV-visible spectrophotometers. UV-visible spectroscopy is commonly associated with absorption spectroscopy, measured through transmission. On the other hand, DRS specifically refers to diffuse reflection spectroscopy. Typically, the former is employed for solutions or thin films, whereas DRS is increasingly utilised. Spectroscopy is a commonly employed, fundamental method of spectrophotometry used to analyse powders and surfaces and requires minimal sample preparation (Morozzi et al., 2021).

X-ray Diffraction Analysis (XRD)

X-ray diffraction (XRD) is a non-destructive technique used to analyse the chemical composition and crystallographic structure of dried synthesised copper nanoparticles, regardless of whether they exhibit an amorphous or crystalline nature. Figure 2 depicts the analysis conducted on the synthesised copper nanoparticles. The pattern shows peaks at 2θ values of 43.46° , 50.57° , and 74.23° , which correspond to the lattice planes (111), (200), and (220), respectively. All observed peaks correspond to the face-centred cubic (FCC) crystal structure and show satisfactory agreement with the standard Joint Committee on Powder Diffraction Standards (JCPDS) No. 04-0836 reference. The observed peak positions align with documented findings related to metallic copper, and no additional peaks associated

with impurities were observed in the sample according to the JCPDS (Tsilo et al., 2023). Therefore, the synthesised copper nanoparticles exhibit a high level of purity.

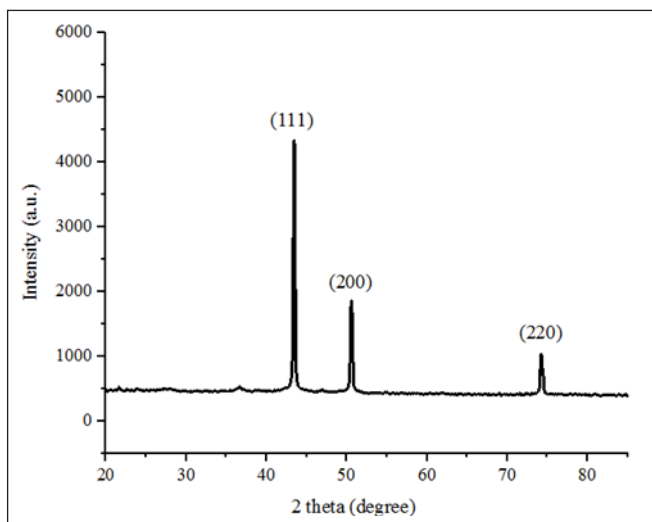


Figure 2. The X-ray diffraction analysis (XRD) of synthesised copper nanoparticles using *Ananas comosus* leaf extract

Energy Dispersive X-ray (EDX)

The Energy Dispersive X-ray (EDX) technique can perform qualitative and quantitative investigations of the elemental composition that could potentially contribute to the production of nanoparticles. Based on Table 5, using energy-dispersive X-ray analysis facilitated the identification of the composition of synthesised copper nanoparticles, which consisted of 95.24% pure copper, 4.08% carbon, and 0.68% oxygen, as determined by weight percentage. The study further revealed that the synthesised copper nanoparticles consisted of 79.68% pure copper, 18.07% carbon, and 2.25% oxygen, as determined by atomic percentage. Copper served as a prominent constituent element within the copper nanoparticles, demonstrating a correlation between the copper content and the overall percentage of copper present in these nanoparticles. The Energy Dispersive X-ray analysis depicted in Figure 3(a) shows the EDX pattern obtained from copper nanoparticles synthesised using *Ananas comosus* leaf extract, revealing a significant concentration of copper. This research suggests that the copper particles obtained exhibit high purity. The utilisation of Energy Dispersive X-ray microanalysis facilitates the acquisition of morphological data pertaining to the synthesised copper

Table 5
The weight and atomic percentage of element presence in the synthesised copper nanoparticles

| Result | Copper | Carbon | Oxygen |
|----------------|--------|--------|--------|
| Mass Norm. (%) | 95.24 | 4.08 | 0.68 |
| Atom (%) | 79.68 | 18.07 | 2.25 |

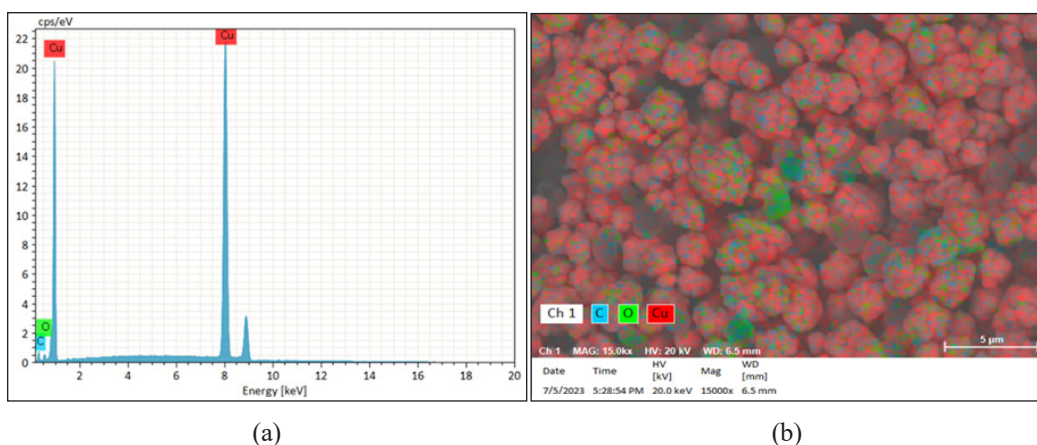


Figure 3. (a) The Energy Dispersive X-ray (EDX) of synthesised copper nanoparticles using *Ananas comosus* leaf extract (b) The portrayed composition of elements present in the synthesised copper nanoparticles using *Ananas comosus* leaf extract

nanoparticles. Based on the data presented in Figure 3(b), it can be observed that the *Ananas comosus* leaf extract, containing copper (Cu), carbon (C), and oxygen (O) elements, plays a significant role in the formation of face-centred cubic copper nanoparticles. Face-centred structures are known to have a higher degree of space efficiency. The observed red colouration of the copper nanoparticles can be attributed to the densely packed arrangement of copper atoms, which contributes to their enhanced stability. The blue colour refers to the carbon elements, while the green indicates the presence of oxygen elements. The colours were used to differentiate the presence of elements in the synthesised copper nanoparticles. Prior studies have indicated that the face-centred cubic structure is a crucial factor in the exploration of innovative nanoscale structures (Cheedarala & Song, 2020). It has been shown that the structure of copper nanoparticles was capped and stabilised by the carbon and oxygen present in minute quantities, which originate from the extract of *Ananas comosus* leaf. The resulting strong structure, as depicted in Figure 3(b), can be attributed to the capping and stabilising mechanism. Therefore, this study has demonstrated the efficacy of phytochemical constituents derived from *Ananas comosus* leaf extract in synthesising copper nanoparticles. X-ray diffraction (XRD) analysis validated the successful formation of a face-centred cubic (FCC) crystalline structure. The diffraction patterns revealed distinct peaks that correspond to the FCC lattice planes, confirming the presence of this structural configuration.

Fourier Transform Infrared Spectroscopy (FTIR)

Fourier Transform Infrared (FTIR) analysis was conducted to investigate the impact of capping or reducing agents and to demonstrate the characteristics of different functional

groups within the plant extract responsible for the synthesis of copper nanoparticles through the reduction of copper. The FTIR study covered the spectral region of 400–4000 cm^{-1} , as depicted in Figures 4(a) and 4(b). Figure 4(a) displays the FTIR spectra of *Ananas comosus* leaf extract, revealing a prominent peak at 3347 cm^{-1} . This spectral feature is attributed to the O-H stretching motion of an alcohol molecule. The observed peaks at 2998, 1378, and 1325 cm^{-1} correspond to the absorption of C-O single bonds. The presence of absorption bands in 1723 and 1649 cm^{-1} suggests C=O carbonyl and C=C stretching vibrations, respectively, likely due to the cyclic aromatic ring. The carboxylic acid functional group shows characteristics of both alcohols and ketones, encompassing the O-H bond characteristic of alcohols and the C=O carbonyl bond typical of ketones.

Hence, carboxylic acids exhibit a pronounced spectral band from 2800 cm^{-1} to 3500 cm^{-1} corresponding to the O-H bond stretching in alcohols. Secondary amines display a singular N-H bond, observed at 3343 cm^{-1} , resulting in a solitary peak. The amide functional group includes both the N-H amine bond and the C=O carbonyl bond, presenting a prominent and moderately wide absorption band in the 3100 cm^{-1} to 3500 cm^{-1} range, corresponding to the N-H amine bond stretching. The observed peaks indicate the presence of functional groups commonly found in the principal chemicals of the *Ananas comosus* leaf extract. The synthesised copper nanoparticles displayed several distinctive peaks in the *Ananas comosus* leaf extract, as shown in Figure 4(b), with modest variations in peak positions and intensities.

FTIR analysis can also be used quantitatively to determine the concentration of certain functional groups, provided that the underlying chemistry is understood and standard reference materials are available. Figure 4(b) illustrates the correlation between the transmittance of the synthesised copper nanoparticles and the absorption of infrared light. When a sample absorbs a significant portion of incident light, transmission through the sample is nearly non-existent. The synthesised copper nanoparticles exhibit a high level of absorbance and a correspondingly low level of transmission. The sample concentration may also influence the intensity of absorption bands in an IR spectrum. According to the Beer-Lambert equation, absorbance is directly proportional to both the concentration of the absorbing substance in the solution and the length of the light path (Bhanvase & Barai, 2021).

The results showed that the composition of the synthesised copper nanoparticles retained the organic constituents derived from the *Ananas comosus* leaf extract. The observed peaks in the nanoparticles suggest functional groups from organic compounds such as flavonoids, alkaloids, and terpenoids encapsulated them. Past research has shown that most flavonoids possess a C6-C3-C6 structure with two benzene rings (A and B) linked by a heterocyclic pyrone ring (C) containing oxygen (Dias et al., 2021). Flavonoid classification is based on specific chemical groups. For instance, flavanols are characterised by a hydroxyl group at

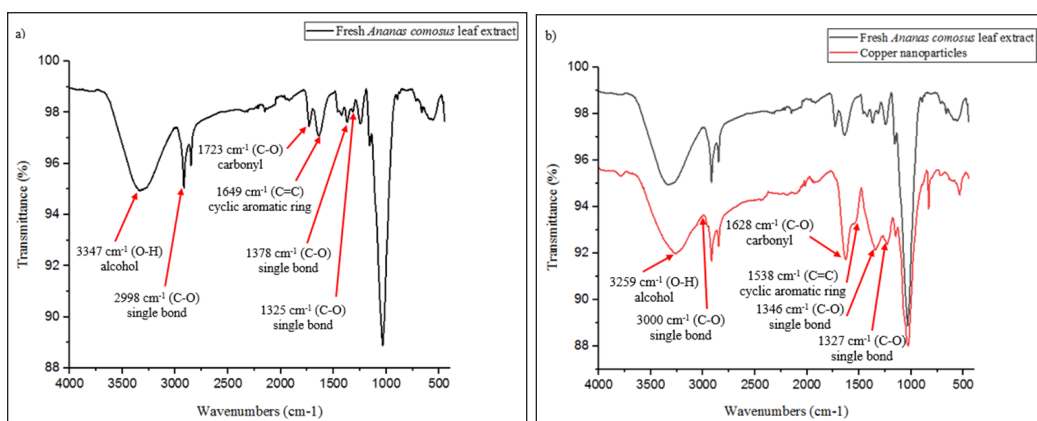


Figure 4. (a) The Fourier Transform Infrared Spectroscopy (FTIR) analysis of fresh *Ananas comosus* leaf extract (b) The Fourier Transform Infrared Spectroscopy (FTIR) analysis of fresh *Ananas comosus* leaf extract and synthesised copper nanoparticles

the 3-position, while flavanones have a C2–C3 double bond, confirming the presence of flavonoid compounds in the synthesised copper nanoparticles (Kumar & Pandey, 2013). Additionally, terpenoids, which are derived from terpenes and include oxygen atoms, likely derive their oxygen from the carbonyl group of the fresh *Ananas comosus* leaf extract, connected to the cyclic aromatic ring of the terpenoid (Adefegha et al., 2022). Alkaloids, chemicals typically containing one or more nitrogen atoms in a heterocyclic ring structure (amine functional group), are also present (Lamponi, 2021). This research confirmed that amines in the fresh *Ananas comosus* leaf extract exhibit a singular N-H bond at 3343 cm^{-1} . Thus, the study indicates that the surface of the copper nanoparticles was effectively covered and stabilised by these organic molecules during synthesis, suggesting the nanoparticles' non-oxidative and highly pure nature.

Field Emission Scanning Electron Microscopy (FESEM)

Figure 5 depicts the synthesised copper nanoparticles' morphological characteristics and structural configuration. The images were obtained using Field Emission Scanning Electron Microscopy (FESEM) with magnifications ranging from the smallest to the largest. The synthesised copper nanoparticles exhibited a tendency to form a disordered agglomeration. The dispersion of the copper nanoparticles is characterised by a loose structure, which leads to agglomeration due to adhesion. Only a limited number of agglomerations occurred, resulting in non-uniform copper nanoparticles. The copper nanoparticles synthesised in this study exhibited strong aggregation due to the dense packing of pure copper atoms. Figure 5 demonstrates that the shape of copper nanoparticles is irregular, while Figure 6 shows that the predominant diameters of the synthesised copper nanoparticles are 51.91

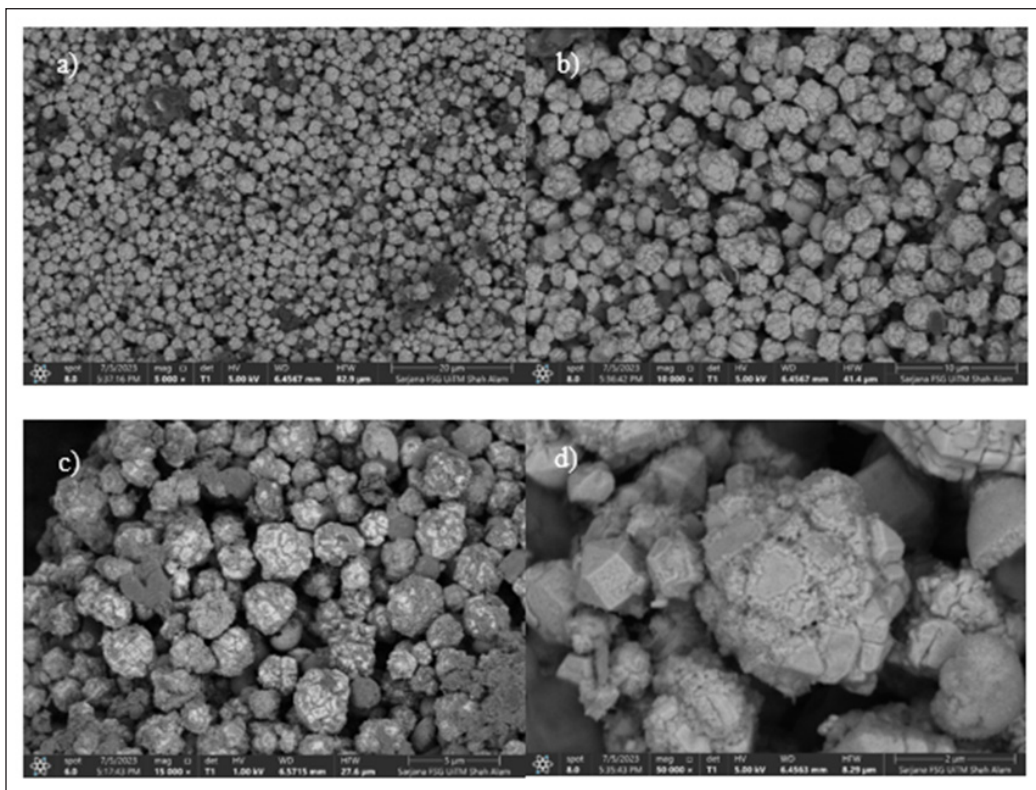


Figure 5. The morphological surface structure of synthesised copper nanoparticles using *Ananas comosus* leaf extract using (a) 1000 \times , (b) 10000 \times , (c) 15000 \times , and (d) 50000 \times magnification of Field Emission Scanning Electron Microscopy (FESEM)

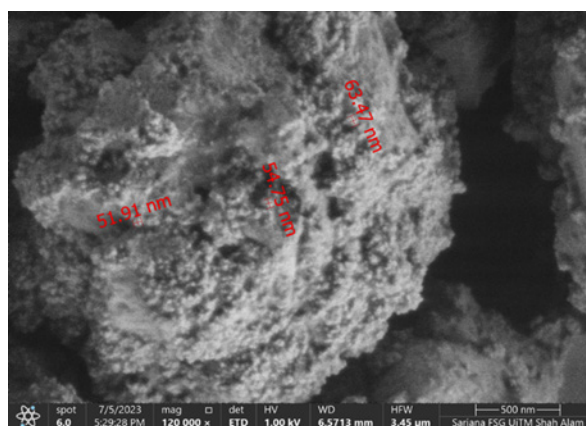


Figure 6. The average size of synthesised copper nanoparticles using *Ananas comosus* leaf extract under 120000 \times magnification of Field Emission Scanning Electron Microscopy (FESEM)

nm, 54.75 nm, and 63.47 nm. The average size of the synthesised copper nanoparticles was 56.71 nm. Determining the shape and average size of synthesised nanoparticles allows for the assessment of their specifications and effectiveness in various applications, such as medicinal, antibacterial, antifungal, magnetic, and electrical effects. The findings of this experiment indicate that the structure of the copper nanoparticles remains stable and consistent with the specified size of the nanoparticles.

CONCLUSION

In the current study, *Ananas comosus* leaf extract was successfully used to synthesise copper nanoparticles through the processes of reduction, capping, and stabilisation by the secondary metabolites present in the extract. The existence of functional groups from secondary metabolites such as flavonoids, alkaloids, and terpenoids derived from the extract facilitated the synthesis process. The synthesised copper nanoparticles succeeded in terms of their crystallographic, optical, and surface morphological properties. Since the synthesis of copper nanoparticles only involves raw materials from *Ananas comosus* leaf and a small amount of copper sulfate, the production cost is very affordable, especially for smallholders. In the future, implementing technological improvements aimed at enhancing the safety of nanoparticles created through green synthesis could potentially boost the development of agricultural economics, particularly in applications such as plant pathogen treatments and microbiology.

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