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Biotech SI

<https://doi.org/10.17113/ftb.63.02.25.8925>

original scientific paper

Agro-Waste Driven Green Synthesis and Characterization of Silver Nanoparticles Using *Oryza sativa* and Spent *Coffea robusta*

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Received: 29 October 2024

Accepted: 30 April 2025



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SUMMARY

Research background. Agricultural waste was utilised to synthesise silver nanoparticles (AgNPs) via green synthesis, a sustainable alternative to traditional synthesis techniques that use hazardous chemicals and extensive processing. AgNPs were produced from spent coffee grounds, *Coffea robusta*, and rice husks, *Oryza sativa*, both prevalent agricultural wastes rich in bioactive substances including proteins, flavonoids, and phenolic acids, which act as natural reducing agents.

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Experimental approach. The formation and stability of AgNPs were confirmed using various methods. UV-Vis spectroscopy displayed surface plasmon resonance (SPR) peaks at 450 nm, signifying AgNP formation, while FTIR identified functional groups responsible for the bio-reduction and stabilisation of nanoparticles. XRD confirmed the crystalline, face-centred cubic structure. Zeta potential analysis showed stable dispersion, and particle size analysis highlighted consistent sizing. The antibacterial activity of AgNPs was assessed by testing their effectiveness against both gram-positive and gram-negative bacteria.

Results and conclusions. Synthesis of AgNPs from spent coffee grounds and rice husks rich in biomolecules served as effective reducing and stabilizing agents. FTIR analysis identified functional groups involved in nanoparticle reduction and stabilization, while XRD verified their face-centred cubic (FCC) crystalline structure. Zeta potential measurements demonstrated stable dispersions, with particle sizes of approximately 187 nm spent coffee ground-AgNPs and 198 nm for rice husk-AgNPs. The synthesized AgNPs also exhibited strong antibacterial activity against both gram-positive and gram-negative bacteria.

Novelty and scientific contribution. The green synthesis approach for AgNPs utilising agriculture waste such as spent coffee grounds and rice husks as natural reducing and stabilizing agents. This study highlights the innovative use of biomolecule-rich materials, producing stable AgNPs with strong antibacterial properties, and establishes a sustainable foundation for advancing nanotechnology applications.

Keywords: agro-waste; green synthesis; *Oryza sativa*; spent *Coffea robusta*; silver nanoparticles

INTRODUCTION

Nanotechnology has emerged as a transformative field dominated by silver nanoparticles (AgNPs) due to their exceptional properties and diverse applications (1). Conventional AgNP synthesis often relies on hazardous chemicals and complex processes which pose significant environmental concerns (2). Consequently, green synthesis has gained attention as a sustainable alternative that utilises environmentally benign materials as reducing agents (3). Agricultural waste rich in biomolecules with reducing capabilities offers a promising platform for AgNP production (4) adhering to the principles of sustainable development and circular economy (5). Agricultural waste rich in proteins, phenolics, and flavonoids acts as bio-reductant agents in the biological synthesis of AgNPs (6) and this approach aids waste management by transforming waste into valuable products. It supports resource conservation by utilising waste materials efficiently, thereby addressing the

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environmental challenges associated with traditional nanoparticle production methods (7). Although various bio-based approaches have been investigated, the potential of rice husks and spent coffee grounds as precursors for AgNP synthesis remains relatively understudied.

Coffee is a popular non-alcoholic beverage obtained from the seeds of the *Coffea* plant which are then roasted (8). Coffee consumption is increasing globally (9) and the coffee industry generates significant amounts of waste, including spent coffee grounds (SCG). SCG are a rich source of biologically active compounds, such as flavonoids, phenolic compounds, and phytonutrients but can also contribute to environmental pollution due to the high levels of tannins and caffeine (10). The polyphenolic compounds in SCG, including chlorogenic acids and melanoidins, interact with metal salt solutions to form metal atoms through a reduction mechanism (11). Additionally, coffee beans contain two main alkaloids, caffeine and trigonelline, and some other compounds like adenine, xanthine, hypoxanthine, and guanine which can facilitate the formation of nanostructures through oxidative coupling (12).

Rice husks (*Oryza sativa*) are a byproduct of the rice milling industry often discarded as agricultural waste (13). Despite its abundance and renewability, rice husk presents a significant opportunity for sustainable nanoparticle synthesis (14). It is a rich source of biologically active compounds, such as phenolic acids and flavonoids, which can facilitate the reduction of metal ions to produce AgNPs (15,16). The AgNPs synthesised from rice husk are safe for humans and the environment and have been shown to have higher reducing power and ROS scavenging capability making them potential antioxidant compounds (17). The use of SCGs and rice husks as reducing agents provides a cost-effective, sustainable, and eco-friendly approach to AgNP production. This method contributes to the sustainable utilisation of agro-waste and a circular bioeconomy, thereby helping to mitigate the environmental impact associated with traditional nanoparticle synthesis.

The main focus of this study is to develop a sustainable and environmentally friendly method in synthesizing silver nanoparticles (AgNPs) using agricultural waste materials. By utilizing rice husks and coffee grounds as reducing agents, we aimed to synthesize nanoparticles and subsequently characterize their properties. This characterization process is essential for evaluating the effectiveness and quality of the produced nanoparticles. Additionally, we understand the potential benefits of AgNPs in various fields, including antimicrobial, catalytic, and biomedical applications. Furthermore, this study paves the way to the advancement to the advancement of green nanotechnology by demonstrating the feasibility and sustainability of using agricultural wastes for nanoparticle synthesis.

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MATERIAL AND METHODS

Preparation of SCG

Spent coffee ground (SCG) extract was prepared using SCG from *Coffea robusta* beans obtained from Ziq Bakery and Cake, Gong Badak, Terengganu, Malaysia. Initially, the SCG was rinsed with deionized water and filtered to remove excess impurities. The cleaned SCG was then dried in a universal oven (UN55, Memmert, Schwabach, Germany) at 60 °C overnight until complete desiccation, then blended to obtain a fine dry powder using a blender (MX-GM1011, Panasonic, Shah Alam, Malaysia). A total of 15 g of powdered SCG was mixed with 150 mL deionized water and 30 mL absolute ethanol (8.33 % *m/V*) in a 5:1 ratio for the extraction of bioactive compounds. This mixture was stirred and heated at 80 °C for 35 min in a water bath (WNB 7, Memmert, Schwabach, Germany). After the mixture was cooled down, the mixture was centrifuged at 2500 rpm for 15 min using a high-speed floor-top refrigerated centrifuge (CR22N, Hitachi, Hitachinaka, Japan) and then filtered to separate solid particles from the liquid phase and freeze-dried to preserve the material for further use.

Preparation of rice husk (Oryza sativa) extract

Rice husk was sourced from MAAS Agros Technology in Kajang, Selangor. A mass of 500 g sample of rice husk was rinsed with deionized water. The rinsed rice husk was dried overnight in a universal oven at 60 °C. The dried rice husk was then powdered using a blender into a fine powder.

For the extraction process, 300 g of rice husk powder was combined with 1000 mL of deionized water (3:10 *m/V* ratio) and heated at 60 °C for 30 min in a water bath. Following heating, the mixture was centrifuged at 2500 rpm for 15 min, which was followed by filtration, and then it was collected and freeze-dried using a freeze dryer (Genesis 35EL Pilot Freeze Dryer, SP Scientific VirTis, Gardiner, New York, USA) for further use.

Synthesis of silver nanoparticles from SCG extract

The SCG extract was added to the 10 mM silver nitrate (AgNO_3) (Bendosen Laboratory Chemicals, Bendosen, Norway) solution in a 1:1 ratio. This mixture was homogeneous and stored in a dry, dark place for 5 h (Fig. S1). The colour of the solution changes. The colour change is indicative of the formation of nanoparticles. These changes are observed and recorded hourly from 0 to 5 h.

Synthesis of silver nanoparticles from rice husk (O. sativa) extract

Silver nitrate solution of 10 mM was prepared from solid AgNO_3 . The freeze-dried rice husk extract was mixed with the AgNO_3 solutions in a 1:4 (*v/v*) ratio (Fig. S2). The reaction mixtures were

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then heated at 75 °C for 15 min in a water bath and subsequently stored in the dark for 96 h to prevent photodegradation. The colour change of the solution is indicative of nanoparticle formation. The colour changes were observed and recorded hourly.

UV-Vis spectroscopy

The absorbance of the solution containing nanoparticles was determined by using a UV-Vis Spectrophotometer apparatus (Shimadzu UV-1800 240 V, Kyoto, Japan) over a wavelength of 350 to 650 nm (18). The reduction of green synthesised silver ions was recorded using the UV-Vis spectrum of the reaction silver nanoparticles mixture.

Particle size analysis

The size of AgNPs synthesised was analysed using a zeta sizer. The hydrodynamic size (Z average), polydispersity index (PDI) and Zeta potential of AgNPs was measured by dynamic light scattering (DLS) (Litesizer 500, Anton Paar, Graz, Austria) (18).

Fourier transform infrared spectroscopy (FTIR) analysis

The chemical functional groups of synthesised AgNPs were analysed using the FTIR (IR Traces-100, Shimadzu, Kyoto, Japan) with in the spectral range of 4000–400 cm⁻¹ (18).

X-ray diffraction (XRD) analysis

The XRD (SmartLab X-ray Diffractometer, Rigaku, Tokyo, Japan) profile of biosynthesised AgNPs was conducted using freeze-dried AgNPs at 20 °C -80 °C, 40 kV and 30 Ma (18).

Antimicrobial assay

The agar disc diffusion method was used to evaluate the antimicrobial activity of rice husk and spent coffee ground silver nanoparticles synthesized AgNPs exhibited strong antibacterial properties against Gram-positive bacteria (*Staphylococcus aureus* and *Bacillus subtilis*) and Gram-negative bacteria (*Pseudomonas aeruginosa* and *Escherichia coli*) (Fig. S3). These bacteria were cultured and streaked on the nutrient agar. Using a densitometer, the streaked bacteria were used to prepare the suspension of 0.5 McFarland standard (equivalent to 1.5·10⁸ CFU/mL). The prepared bacterial inoculum was uniformly swabbed on the Mueller-Hinton agar (MHA) plate. Then, rice husk and spent coffee ground extract-AgNPs were added to the sterile plain disc and placed onto an MHA plate using concentrations of 400, 200, and 100 mg/mL for both samples. The zone of inhibition was measured

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using a vernier calliper after incubating for 24 h at 37 °C. The gentamicin disc was the positive control, and distilled water was the negative control, both used to assess the effectiveness of the antimicrobial assay.

Statistical analysis

All the data are presented as mean±standard deviation (S.D.). ANOVA and Tukey's HSD test was used for the statistical analysis using the IBM SPSS Statistics version 29.0 software, setting the significance at a p-value of less than 0.05 (19,20).

RESULTS AND DISCUSSION

Biosynthesis and spectroscopic analysis of AgNPs derived from agricultural waste

The synthesis of silver nanoparticles (AgNPs) was successfully achieved through a green approach utilizing extracts from *Oryza sativa* (rice husk) and spent coffee grounds as reducing agents. This environmentally friendly method involved the reduction of Ag⁺ ions to AgNPs, visually indicated by a distinct change of colour from light yellow to brownish black (21). The colour intensity of *Oryza sativa* extract in 10 mM AgNO₃ solution increased from 0 to 96 h (Fig. S4a) of storage, meanwhile for spent coffee ground in 10mM AgNO₃ from 0 to 5 h (Fig. S4b). The mechanism, attributed to the excitation of electrons via surface plasmon resonance (SPR), confirmed the formation of AgNPs (22). Phenolic compounds from the plant extracts contributed a vital role in this bio-reduction process, acting as both reducing and stabilizing agents for the nanoparticles (23). Optimization of parameters such as metal ion concentration, temperature, and reaction time was essential for achieving optimal AgNP formation (24). The interaction of silver ions with phenolic acids from rice husk and coffee grounds was confirmed by the change of colour from light yellow to brownish black, indicating increased stable AgNP formation. Similar to Vasylijev, the phenolic compounds in the black currant pomace extract contained phenolic compounds which may have contributed for the improved stability of AgNPs with this extract (25). The extracts were freeze-dried before being combined with AgNO₃ for synthesis. Freeze-drying removes 98 % of water from the samples, allowing long-term storage while conserving the bioactive phenolic acids responsible for AgNP formation (26).

The UV-Vis spectra were utilized to determine the structure of the AgNPs by analyzing their free surface electron plasmon oscillations. The SPR peak, indicating AgNP formation, typically appears in the visible range (350-650 nm), influenced by particle size, shape, and environment (27). For rice husk-synthesized AgNPs, as indicated in Fig. 1a, a weak or nearly absent SPR peak at 450 nm was observed after 96 h, suggesting AgNPs formation. The

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phenolic acid components peaks in the spectra obtained for AgNPs synthesized were observed at approximately 430 nm, as stated by Lieu (15).

The concentration of the AgNPs is directly proportional to the intensity of the absorption peak. A higher concentration of nanoparticles or larger nanoparticles can result in a more intense absorption peak (28). The increasing absorbance over time suggested the growth of larger particles, indicating a bathochromic shift as the SPR peak shifted toward longer wavelengths (29). In SCG-mediated AgNP synthesis, a rapid colour change from light to dark brown within 30 min confirmed the formation of AgNPs, with a peak at 450 nm intensifying over time, reaching maximum absorbance after 4 h (Fig. 1b). Similarly silver nanoparticles from Spent coffee ground shows peak at 430 nm SPR band (30).

The UV-Vis analysis confirmed AgNP synthesis from both rice husk and SCG extracts, with a consistent SPR peak around 450 nm. These results align with previous studies, indicating SPR bands in similar ranges (30). Spherical metal nanoparticles (NPs) can only produce a single SPR band, while anisotropic particles may generate two or more SPR bands depending on their shape (18). In the current study, a single SPR peak was observed which proposed that synthesized silver nanoparticles were spherical in shape. This study highlights the feasibility of using sustainable, cost-effective, and environmentally friendly methods for AgNP synthesis from agricultural wastes, with potential applications across various fields.

Functional group analysis of AgNP by FTIR

FTIR analysis was performed to identify the functional groups and biomolecules involved in the bio-reduction of AgNPs derived from rice husk and SCG. This technique provides critical insights into the surface chemical composition and reactive sites of the AgNPs which are essential for understanding their surface reactivity (31). For the rice husk-derived AgNPs, the FTIR spectrum, as depicted in Fig. 2a, showed several significant absorption peaks. The significant peak at 3332.99 cm^{-1} (O-H stretching), indicates the presence of carboxylic acids (32). The O-H stretching is probably originating from the phenolic O-H groups in the lignin structure of rice husk (33,34). Additional prominent peaks were observed at 2931.80 cm^{-1} corresponding to CH_2 stretching, 2360.87 cm^{-1} (alkynes or ammonium), 1635.64 cm^{-1} (C-N and C-C stretching, associated with proteins or amides), 1319.31 cm^{-1} (N=O stretching, nitro compounds), 1081.41 cm^{-1} (C-N stretching, amines), and 648.08 cm^{-1} (C-Cl stretching, alkyl groups) (18,35). A broad band at 3286 cm^{-1} and a peak at 2924 cm^{-1} shown in Fig. 2b, in the FTIR spectrum of SCG-AgNPs indicates distinct features (36). The peak at 3286 cm^{-1} corresponds to N-H and O-H stretching vibrations and the peak at 2924 cm^{-1} corresponds to asymmetric stretching of the C-H bond in methyl groups (37). The broad band at 3286 cm^{-1}

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suggested the presence of -OH groups in alcohols and phenolic compounds, which function as capping and stabilizing agents. For the rice husk-derived AgNPs, the polyphenols and polysaccharides act as capping and stabilising agents (38) with the peaks at 1357 cm^{-1} and 1273 cm^{-1} associated with the bending vibrations of C-H bonds in methyl and methylene groups which also influence the size, shape, and stability of the AgNPs. The absorption peak at 663.51 cm^{-1} indicated C-Cl stretching vibrations, reflecting the presence of halogen compounds that act as stabilising agents to prevent nanoparticle agglomeration (39). Various functional groups were present in rice husk and SCG extracts as confirmed by FTIR analysis, thus this agricultural waste is a potential resource for the green synthesis of silver nanoparticles, an approach that contributes to the advancement of sustainable and eco-friendly nanotechnology.

X-ray diffraction spectroscopy (XRD) analysis of silver nanoparticles synthesized from rice husk and spent coffee grounds

The molecular and crystalline structures of AgNPs were analyzed through X-ray diffraction spectroscopy (XRD), which provides critical insights into the crystalline structure and phase identification of materials. XRD analysis of the molecular and crystalline AgNP structures provided valuable information about the physicochemical properties and degree of crystallinity of the synthesised AgNPs (40). The XRD pattern of the rice husk-derived AgNPs (Fig. 3a) exhibited distinct peaks at 38° , 43° , 64° , and 77° , corresponding to the (111), (200), (220), and (311) planes, respectively, confirming the presence of a face-centred cubic (FCC) structure (41), that is, the synthesised nanoparticles were crystalline. This was in line with previous studies where similar XRD patterns were reported for AgNPs synthesised using apricot and black currant pomace extracts, as well as rice husks subjected to acid-alkali pretreatment (15,24). In addition, the XRD analysis of SCG-derived AgNPs (Fig. 3b) revealed prominent peaks across the 2θ range from 20° to 80° , with distinct peaks at 27.24° , 38.01° (111), 41.11° (200), 64.3° (220), and 77.3° (311) corresponding to planes of Bragg's reflection, confirming a face-centred cubic crystalline structure (39). Notably, the peak at 38.01° indicates Ag (0) with an FCC structure, corroborating the effective reduction of Ag^+ ions mediated by both SCG and rice husk (41). Overall, these findings demonstrate the successful formation of crystalline silver nanoparticles using agricultural waste, highlighting their potential for sustainable and eco-friendly applications in nanotechnology (18).

Particle size and zeta potential

The particle analysis of AgNPs produced from rice husk and SCG extracts revealed that the AgNPs synthesised from rice husk had a larger average size of $(198.59 \pm 3.65)\text{ nm}$ (Fig. 4a), attributed

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to the complex chemical composition of the rice husk which influences nanoparticle formation. Furthermore, these particles demonstrated a zeta potential of (-21.2 ± 2.0) mV and a PDI of 26.9 % (Fig. 4b). SCG-derived AgNPs had an average size of 187 nm (Fig. 4c), typical of biologically synthesised nanoparticles. However, their stability was moderate, as indicated by a zeta potential of -11.8 mV and a PDI of 22.1 % (Fig. 4d), which suggests inadequate electrostatic repulsion, therefore the potential for particle aggregation. The multiple peaks in the particle diameter distribution of rice husk indicate polydispersity and the particle's negative charge tends to enhance stability by inhibiting aggregation (42). The negatively charged surface of the nanoparticles contributes to the anionic capping agents, including polyphenols and flavonoids from black currant and apricot pomace extracts that are coordinated to the surface of the silver nanoparticles (24). Optimising the synthesis conditions is critical to achieving AgNPs with desirable size and stability for specific applications. Previous studies have demonstrated that both the concentration of silver nitrate (AgNO_3) and the pH of the reaction medium significantly influence the characteristics of the resulting nanoparticles. Elevated pH accelerates the reduction of Ag^+ ions resulting in smaller AgNPs due to accelerated crystallisation. Additionally, higher pH can mitigate nanoparticle aggregation by fully charging the particle surface, thereby enhancing electrostatic repulsion. Conversely, increasing AgNO_3 concentration can also lead to the formation of smaller nanoparticles (42).

Antimicrobial analysis of AgNPs

Organic nanoparticles provide thermal stability, tensile strength, prolonged shelf life, and antibacterial properties (43). AgNPs exhibit antibacterial activity against both gram-negative and gram-positive bacteria including multidrug-resistant strains (40). Correspondingly, the present study showed that AgNPs from rice husk and SCG exhibit similar antibacterial activity against both gram-positive and gram-negative bacteria (Table 1 and Table 2). Rice husk silver nanoparticles have antimicrobial activity against bacteria at 400 mg/mL concentration. The maximum zone of growth inhibition against *Staphylococcus aureus* and *Bacillus subtilis* are 10 mm and 12 mm in size, respectively, as shown in Table 1. The inhibition zone against *Pseudomonas aeruginosa* and *Escherichia coli* is 11 mm for both bacteria at 400 mg/mL. Furthermore, spent coffee ground AgNPs (Table 2) have antimicrobial activity against bacteria maximum at a 400 mg/mL concentration. The maximum growth inhibition zone against *S. aureus* and *B. subtilis* is 9 and 12 mm in size, respectively. Inhibition zones against *P. aeruginosa* and *E. coli* are 9 and 11 mm at 400 mg/mL.

The antimicrobial efficacy of AgNPs is markedly influenced by their physicochemical properties, including shape, size, concentration, and colloidal state (44). These properties enable AgNPs to interact with or penetrate cell walls and membranes, thereby exerting their antimicrobial

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effects (40). A recent study discovered that rice husk inhibited the growth of *S. aureus*, *Escherichia coli*, and *Salmonella enterica* demonstrating its potential as an antimicrobial agent (45). Moreover, rice husk extract was effective against clinical strains of *S. aureus* isolated from skin wound infections (17,46). It has also been reported that SCG samples were more effective against gram-positive than gram-negative bacteria (47). Both rice husk and SCG-derived silver nanoparticles synthesised exhibited potent antimicrobial properties, demonstrating potential for biomedical applications (48,49).

CONCLUSIONS

In conclusion, this study successfully demonstrated a green synthesis method that produces silver nanoparticles (AgNPs) from agricultural waste, particularly spent coffee ground (SCG) and rice husk. The findings demonstrated that both rice husk and SCG extracts act as reducing and stabilizing agents by utilizing their abundant biomolecule content, which includes flavonoids, phenolics, and other organic components. These bioactive compounds emphasized the multiple uses of agricultural waste in synthesis of AgNP by enhancing in the stability of nanoparticles and reducing silver ions. In both rice husk and SCG-mediated syntheses, AgNP production was confirmed by the biosynthesis process's significant colour change and surface plasmon resonance (SPR) peaks around 450 nm. The potential of these waste materials as bio-reductants was further demonstrated by FTIR analysis, which pinpointed the precise functional groups involved in the reduction and stabilization processes. The produced AgNPs' face-centered cubic (FCC) crystalline structure has been demonstrated by X-ray diffraction (XRD) investigations, which is consistent with known properties of silver nanoparticles. Zeta potentials indicated stable nanoparticle dispersions, and particle analysis showed diameters of about 187 nm for SCG-AgNPs and 198 nm for AgNPs produced from rice husks. By recycling agricultural waste, this study demonstrates the viability of employing rice husks and SCG as affordable, sustainable precursors for AgNP synthesis, which is consistent with the ideas of the circular economy. The produced AgNPs showed encouraging antibacterial qualities, suggesting potential applications in the environmental and biomedical fields. By encouraging the production of eco-friendly, resource-efficient nanoparticles, this endeavour advances green nanotechnology.

FUNDING

Authors would like to thank the Research Intensified Grant Scheme (RIGS) by Universiti Malaysia Terengganu [UMT/RIGS/2023/55438] for the financial support in the completion of this work.

CONFLICT OF INTEREST

The authors declare that they have no known financial or personal conflicts of interest that could have affected the work presented in this paper.

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SUPPLEMENTARY MATERIAL

All supplementary material are available at www.ftbcom.hr.

AUTHORS' CONTRIBUTION

Nithiskanna and Nurul Afifah contributed to the investigation, the writing of the original draft, and participated in reviewing and editing the manuscript. Wan Nor Dalila Wan Fauzi and Hawa Dalily Mohd Jefri were involved in the writing of the original draft, as well as reviewing and editing. Jasvini Bala Murally and Liam Jing Zhi contributed to the investigation and the writing of the original draft. Seeram Ramakrishna and Amirul Al-Ashraf Abdullah were responsible for the conceptualization, visualization, and supervision of the study. Sevakumaran Vigneswari contributed to the conceptualization, visualization, project administration, and funding acquisition, as well as reviewing and editing the manuscript.

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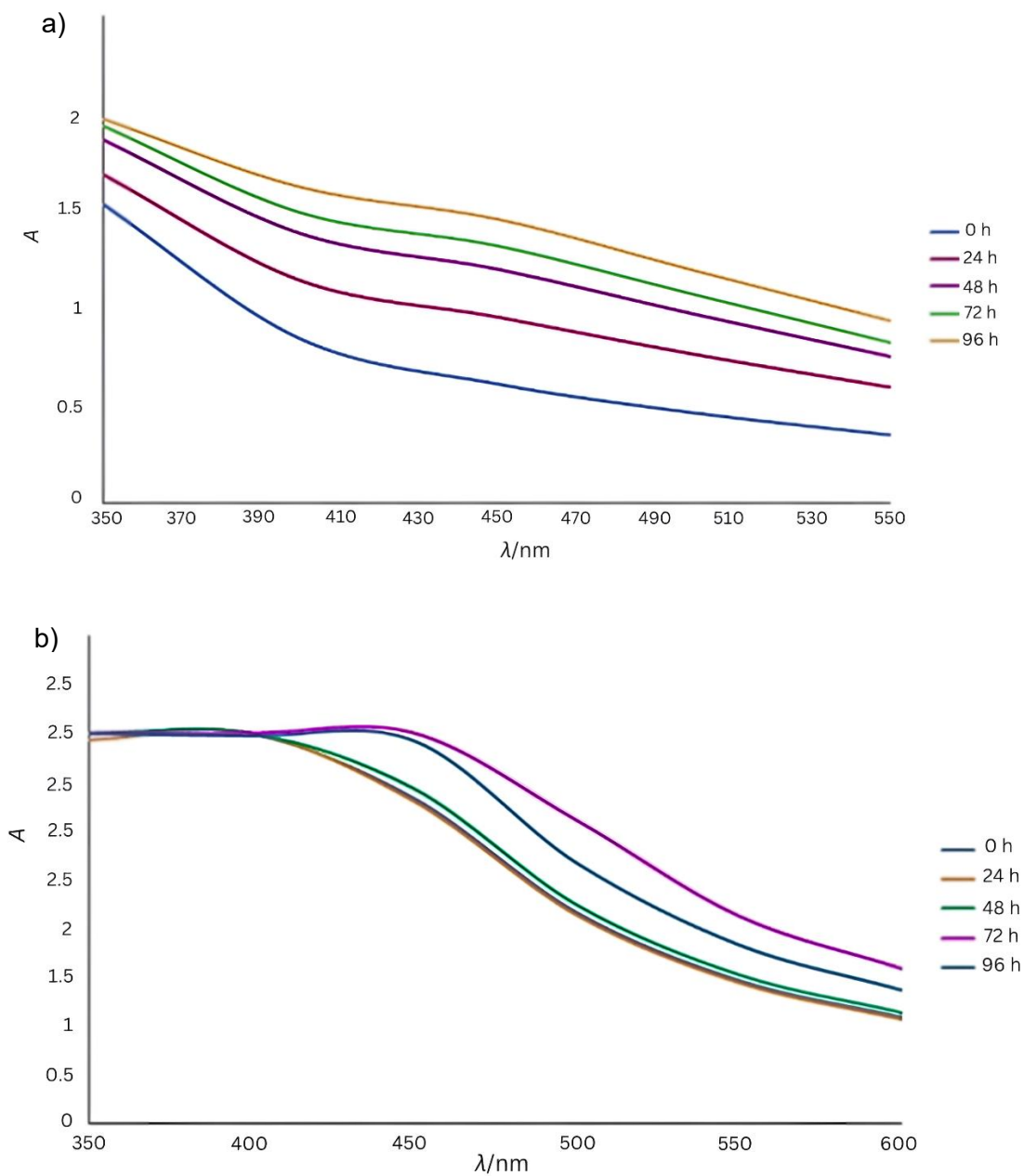


Fig. 1. Absorbance peak of UV-Vis spectra of: a) rice husk-AgNPs and b) spent coffee ground-AgNPs (SCG)

a)

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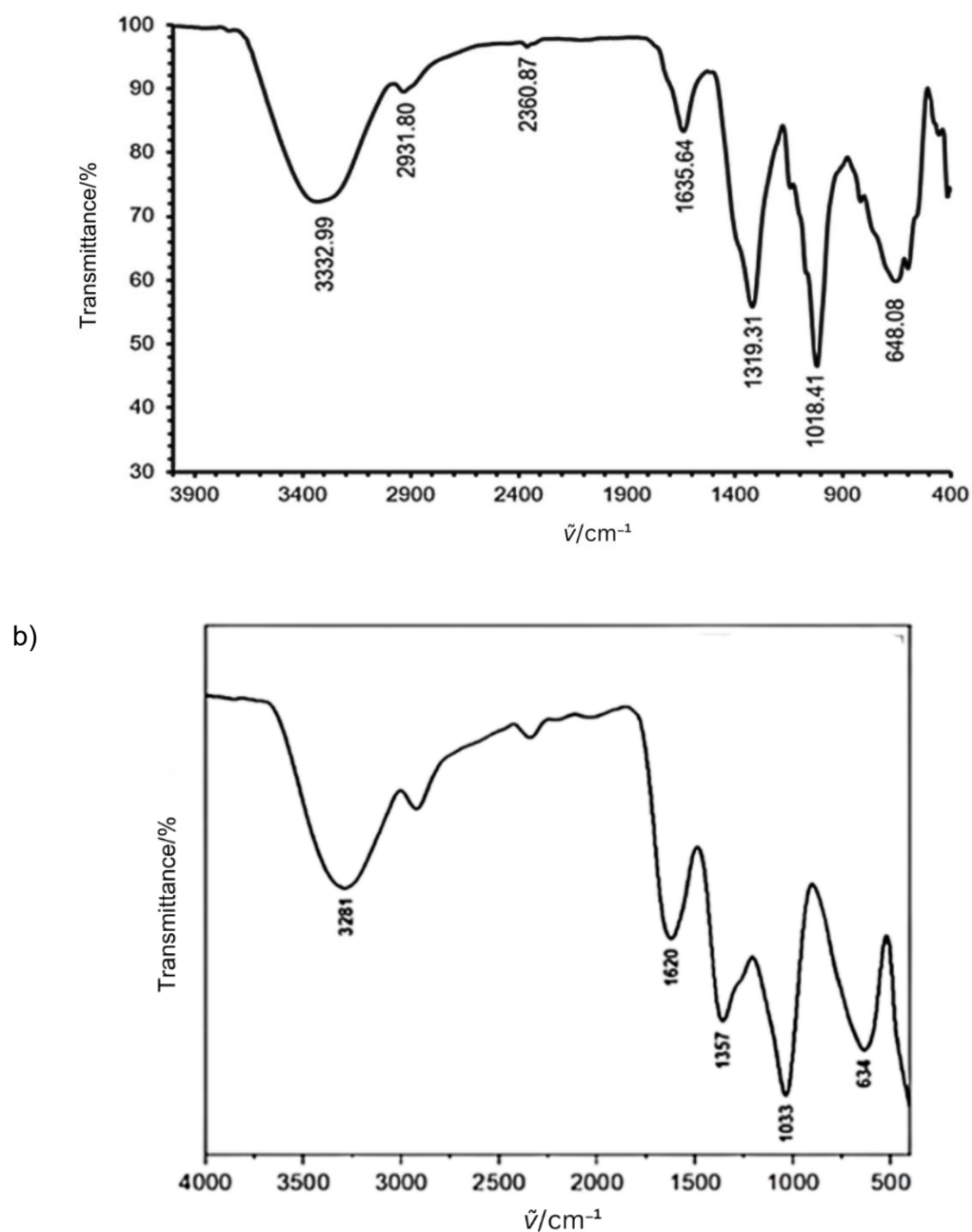


Fig. 2. FTIR spectra of AgNPs from: a) rice husk AgNP and b) spent coffee ground

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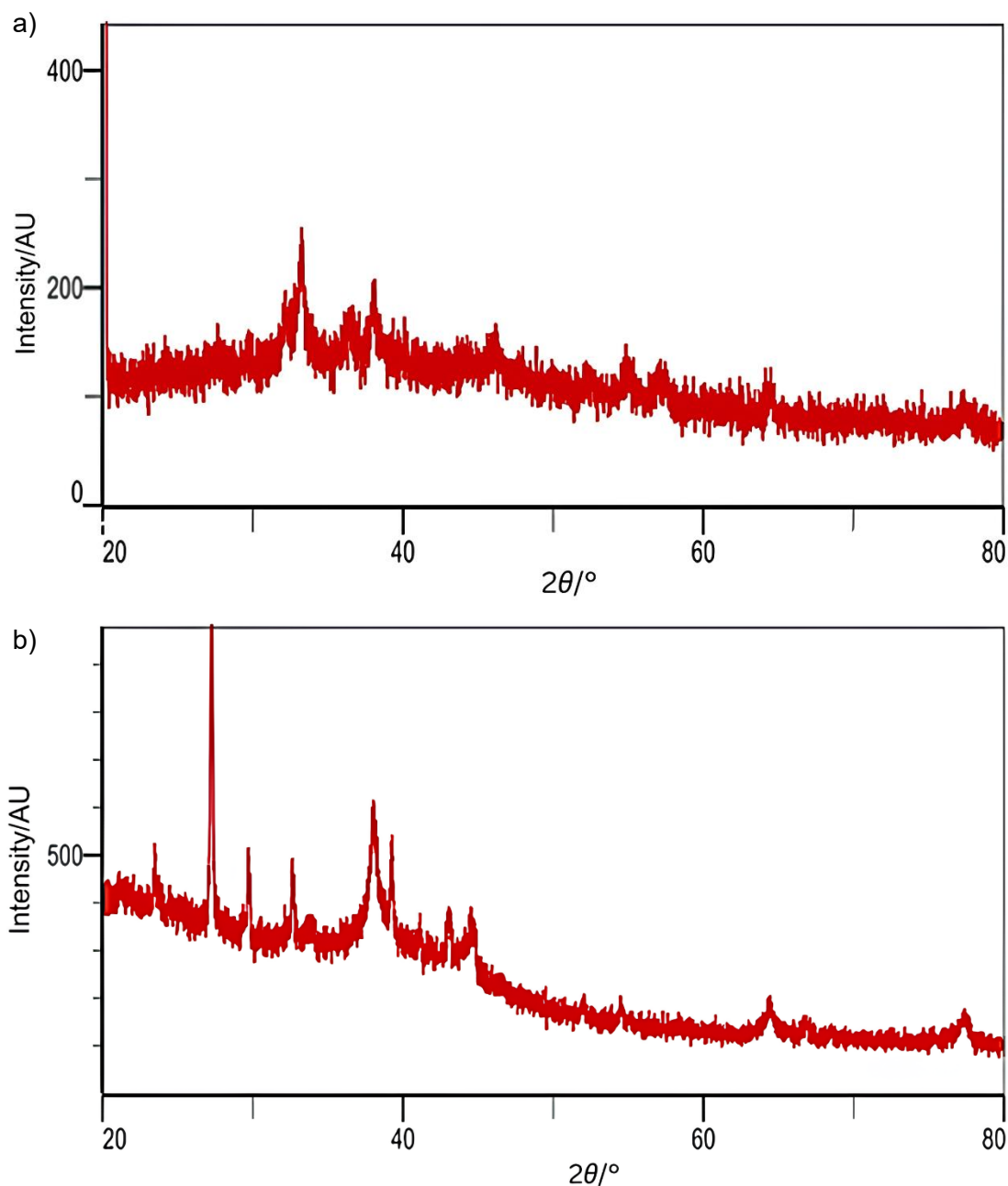
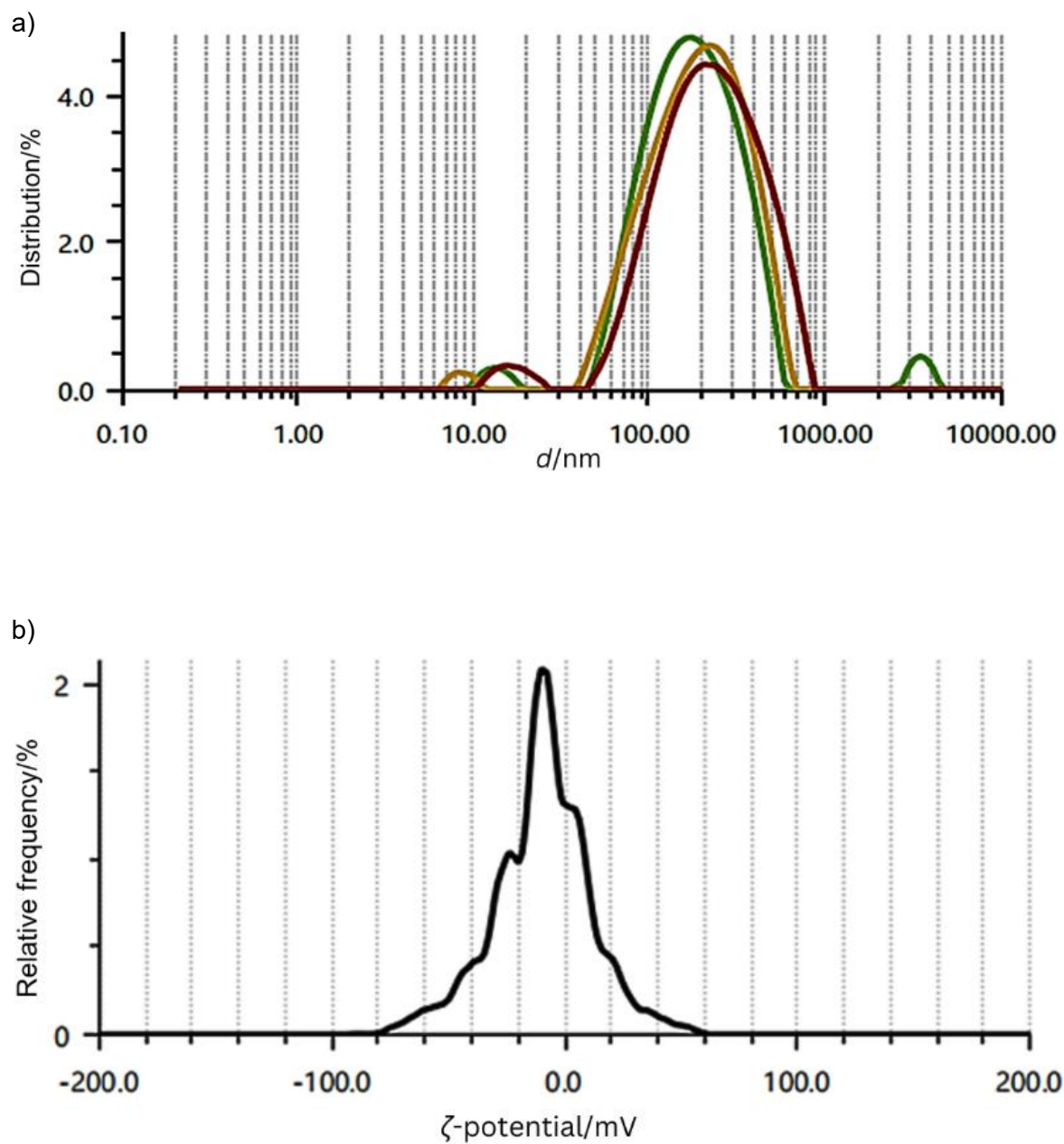


Fig. 3. XRD analysis of AgNPS from: a) rice husk *Oryza sativa* and b) spent coffee ground

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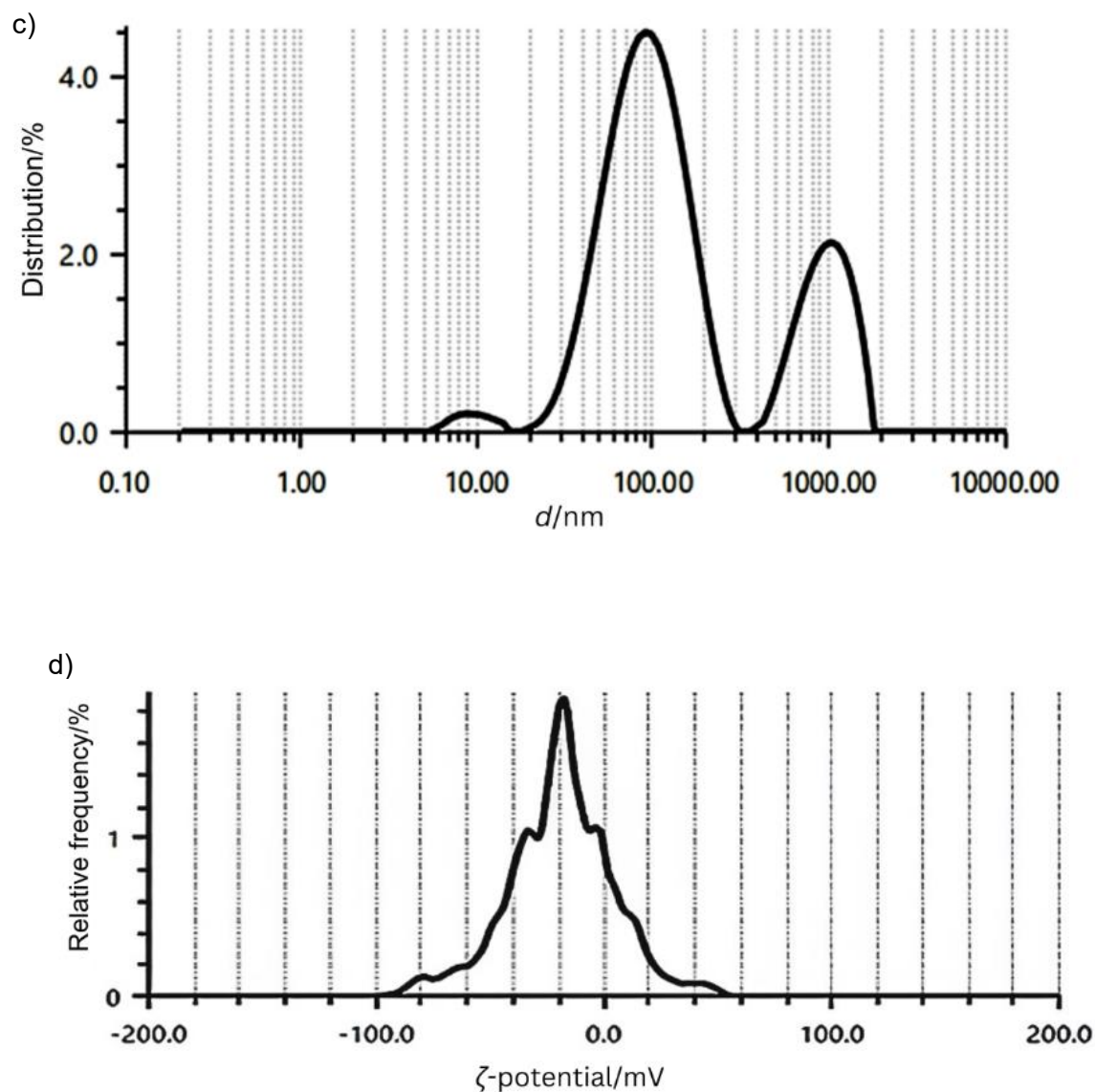


Fig. 4. Particle size and zeta potential of AgNPs from: a and b) rice husk and c and d) spent coffee ground

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Table 1. Antibacterial activity of rice husks silver nanoparticles exhibited against both Gram-positive and Gram-negative bacteria

Organism	Gentamicin (mm)	Rice husk AgNP (mm)			Rice husk extract (γ =10 mg/mL) (mm)	Silver nitrate (AgNO ₃) (γ =1.7 mg/mL) (mm)	Control (mm)
		Concentration (mg/mL)					
		400	200	100			
<i>Pseudomonas aeruginosa</i>	(21±0.7)	(10±0.7) ^a	(9±0.7) ^{ab}	(8±0.7) ^b	0	(7±0)	0
<i>Escherichia coli</i>	(20±0)	(11±0) ^a	(9±0) ^{ab}	(8±0) ^b	0	(7±0)	0
<i>Staphylococcus aureus</i>	(21±0)	(9±0.7) ^a	(8±0.7) ^{ab}	(7±0.7) ^b	0	(7±0)	0
<i>Bacillus subtilis</i>	(22±0)	(10±0.7) ^a	(9±0.7) ^{ab}	(9±0.7) ^b	0	(7±0.7)	0

*Data are presented as mean value±S.D. (N=3). Tukey's HSD test indicates that different letters (^{a,b}) denote significant differences within the concentration at the p<0.05 level

Table 2. Antibacterial activity of spent coffee ground silver nanoparticles exhibited against both Gram-positive and Gram-negative bacteria

Organism	Gentamicin (mm)	Spent coffee ground AgNP (mm)			Spent coffee ground extract (γ=10 mg/mL) (mm)	Silver nitrate (AgNO ₃) (γ=1.7 mg/mL) (mm)	Control (mm)
		Concentration (mg/mL)					
		400	200	100			
<i>Pseudomonas aeruginosa</i>	(21±0.7)	(9±0) ^a	(8±0) ^b	(7±0) ^c	0	(7±0)	0
<i>Escherichia coli</i>	(20±0)	(10±0.7) ^a	(8±0.7) ^b	(7±0.7) ^c	0	(7±0)	0
<i>Staphylococcus aureus</i>	(22±0)	(9± 0.7) ^a	(8±0) ^b	(7± 0) ^c	0	(7±0)	0
<i>Bacillus subtilis</i>	(22±0)	(9±0.7) ^a	(8±0.7) ^b	(7± 0.7) ^c	0	(7±0.7)	0

*Data are presented as mean value±S.D. (N=3). Tukey's HSD test indicates that different letters (^{a,b}) denote significant differences within the concentration at the p<0.05 level

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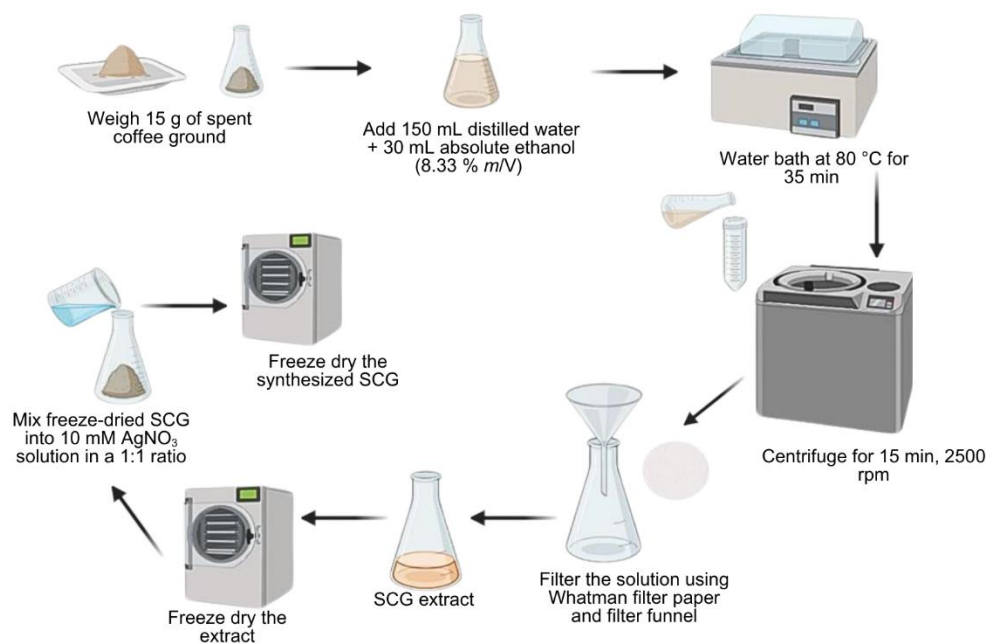


Fig. S1. Schematic illustration of the SCG AgNPs (created using BioRender.com)

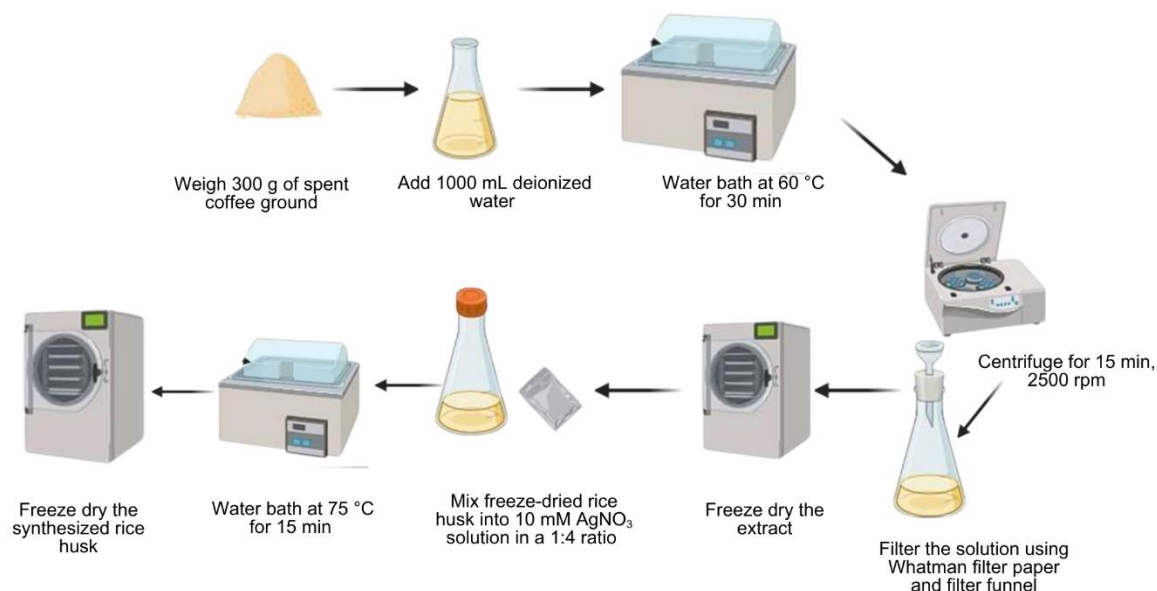


Fig. S2. Schematic illustration of the rice husk AgNPs (created using BioRender.com)

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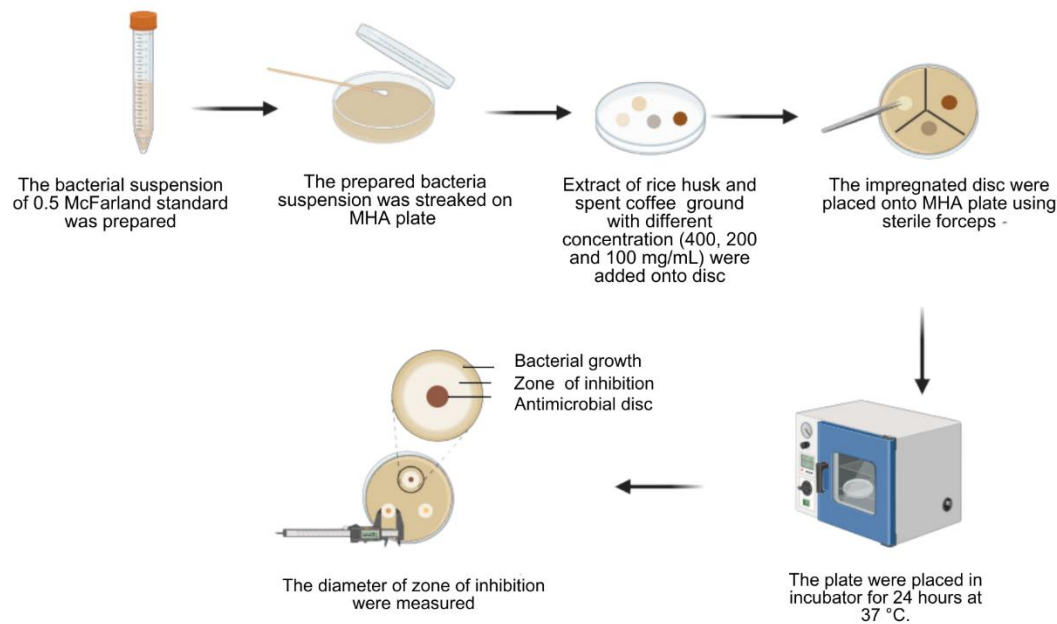


Fig. S3. Schematic illustrating the for AgNP antimicrobial test (created using BioRender.com)

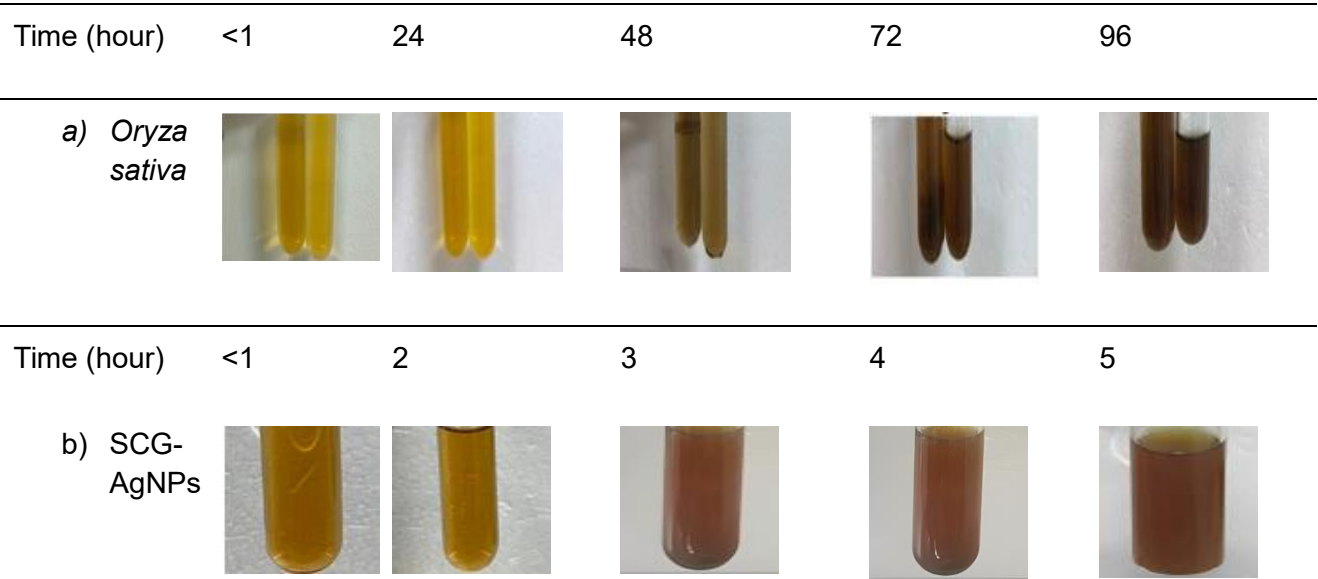


Fig. S4. The synthesis of AgNP based on colour changes at different time interval for a) rice husk and b) spent coffee ground