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Silver nanoparticle biosynthesis by using phenolic acids in rice husk extract as reducing agents and dispersants

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ABSTRACT

Rice husk extract, obtained using acid and alkali pretreatment extraction (AAPE), contains bioactive compounds and exhibits reducing abilities. Phenolic composition in rice husk extract was analyzed and the mechanism of silver nanoparticle (AgNP) biosynthesis by using AAPE rice husk extract was investigated in this study. Stable and spherically shaped AgNPs with a size of <15 nm were prepared under the following conditions: 0.001 M AgNO₃, AAPE rice husk extract diluted 10 times, pH 10, and reacted at 25 °C for 60 min. Synergistic effects among phenolic acids contributed to the formation of AgNPs, with the acids acting as excellent reducing agents (owing to their abundant hydroxyl groups) and excellent dispersants (owing to their derived C=O groups), which enhanced the NPs' stability. Caffeic acid (CA) was demonstrated to synthesize AgNPs independently and is suggested to be the most crucial compound for reducing Ag⁺ during the biosynthesis with rice husk extract. A possible mechanism and reaction process for the formation of AgNPs synthesized using CA in rice husk extracts is proposed.

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1. Introduction

Silver nanoparticles (AgNPs), between 1 and 100 nm in size, have gained interest because of their distinctive physico-chemical and optical properties and are employed in fields such as biological detection [1], conduction [2], catalysis [3], antimicrobials [4,5], and wound healing [6]. AgNPs can be prepared through various chemical and physical methods, such as through chemical reduction with sodium

borohydrate as the reducing agent and by using capping agents to stabilize the solution and prevent flocculation. The disadvantages of these methods are typically that they require highly toxic chemicals or complex steps to achieve purification [7,8]. Biosynthesis of nanostructure materials is not only a favorable method of fabricating AgNPs but also an ecofriendly approach because it minimizes the use of substances that are hazardous to human health and the environment [9,10].

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In the search for a clean and nontoxic AgNP synthesis method, biosynthesis is a desirable candidate because it is analogous to the preparation of nanocrystals through chemical reduction. The raw materials used are biocompatible and reducible bioresources. Several studies have reported using natural material sources such as phenolics in plants, enzymes in microorganisms, and honey for synthesizing AgNPs [11–13]. Bioreduction of Ag^+ to yield metal NPs has been performed using various cereals, such as the bran of husk extract from *Oryza sativa* [13]; *Sorghum bicolor* [14]; and the seeds of *Macrotyloma uniflorum* [15].

The reduction reaction in AgNP biosynthesis is performed under mild temperature and pressure conditions, the metal precursor is used at a low concentration, and the nanometal product is stable [16]. Rice husk contains bioactive compounds, such as phenolic acids, which promote the antioxidant activity of rice in an antioxidant defense system [17,18]. Approximately 20% of paddy weight is the husk, which is discarded during the rice husking process and forms a major proportion of agricultural waste in Taiwan. Accordingly, the present study investigated the biosynthesis of AgNPs using rice husk extract as a reduction agent and stabilizer.

Phytochemicals are regarded as the major ingredients involved in the biosynthesis of AgNPs [7,15], and phenolic acids are the major phytochemicals in rice husk [19]. Phenolic acids have been reported to possess hydroxyl and carbonyl groups, which can bind to metals. Phenolic compounds may inactivate ions through chelation, and their chelating ability is likely related to the high nucleophilic characteristic of their aromatic rings [15]. Therefore, phenolic acids should have high antioxidant activity.

For most applications, the properties of metal NPs are determined by their size, shape, composition, and structure [20]. The preparation of high-quality AgNPs with controllable physicochemical properties is of great importance. Therefore, the effects of experimental parameters such as pH, incubation temperature, extract concentration, and AgNO_3 concentration on the formation of AgNPs were studied to identify desirable biosynthesis conditions when acid and alkali pretreatment extraction (AAPE) rice husk extract is used to obtain well-dispersed and stable AgNPs. The Ag^+ reduction process corresponded to a change in the structure of the phenolic acids, and the stabilization mechanism of AgNPs by phenolic acids was deduced in this study. The phenolic acids that contributed primarily to the reduction reaction during the biosynthesis were also analyzed.

2. Materials and methods

2.1. Materials and chemicals

Fresh rice (*O. sativa japonica*, Kaohsiung 145) husk was collected from the area surrounding Yilan County, Taiwan. The rice husk was hot-air-dried until its water content was less than 5% at 60 °C. The dried rice husk was dry-milled, and powder smaller than 60 mesh was collected as rice husk powder (RHP).

Sodium hydroxide, hydrochloric acid, methanol, and ether were purchased from Merck (Darmstadt, Germany). Ethyl

acetate was purchased from Macron Chemicals (Center Valley, PA, USA). Folin–Ciocalteu reagent, Na_2CO_3 , 4-hydroxybenzoic acid (4-hyd BA), 3-hydroxybenzoic acid (3-hyd BA), caffeic acid (CA), syringic acid (SA), gallic acid (GA), vanillic acid (VA), ferulic acid (FA), *p*-coumaric acid (*p*-Cou), AgNO_3 , and reagents for preparation of buffer solution were purchased from Sigma–Aldrich Chemical Co. (St. Louis, MS, USA). Protocatechuic acid (proto CA) and Ag powder (0.6–2 μm , 99.9%) were purchased from Alfa Aesar Co. (Ward Hill, MA, USA).

2.2. Extraction of phenolics from rice husk

For AAPE, phenolic acids were extracted using the method reported by Tang [21] with minor modifications. RHP was hydrolyzed with concentrated sulfuric acid under a 1:8 (w/v) ratio, and this mixture was heated for 40 min at 115 °C in a high-pressure reactor (Model 4522M, Parr Instrument Company, Moline, IL, USA). The cooled reaction product was filtered and the retentate was washed with deionized water to neutralize its pH, after which it was dried for 6 h at 50 °C to become acid-hydrolyzed rice RHP (AHRHP). The AHRHP was reacted with 8.7% sodium hydroxide under a 1:30 (w/v) ratio, and the mixture was heated for 118 min at 132 °C in a high-pressure reactor. The alkali-treated AHRHP solution was adjusted with concentrated hydrochloric acid to a pH of approximately 2.0–3.0 to avoid the $\text{pK}_{\text{a}1}$ of the phenolic acids (approximately 4–5). The solution was extracted with 4 °C ether:ethyl acetate (1:1, v/v) and was shocked ultrasonically (LEO-150, Leo Ultrasonic Co., Ltd, New Taipei City, Taiwan) for 30 min. The mixture was then centrifuged (Model 7780, Kubota Corporation, Japan) at 2500 $\times g$ for 15 min at 4 °C, after which the supernatant was collected. Sediment was extracted three times following the outlined procedure. These extracts were concentrated and blow-dried with nitrogen. The dried extracts were dissolved in methanol and used as AAPE solution.

For hot water extraction (HWE), RHP and deionized water (1:1, w/v) were heated for 30 min at 90 °C. The cooled crude extract was centrifuged at 8000 $\times g$ for 30 min. The filtered supernatant was used as the HWE solution.

2.3. Determination of phenolics in rice husk

The total phenolic content was determined using the Folin–Ciocalteu method [22]. A total of 0.1 mL of the extract was placed in a test tube and then 0.50 mL of Folin–Ciocalteu reagent and 6.00 mL of deionized water were added. After incubation for 2 min, 2 mL of 15% Na_2CO_3 was added, after which the solution was left for 0.5 min; water was then added to bring the volume to 10.0 mL. Absorbance was measured through UV–vis spectrophotometry (U2001 UV–vis Spectrophotometer, Hitachi, Japan) at 755 nm after incubation for 2 h. The content of the phenolic compounds was analyzed according to the method of Tang [21] by using high-performance liquid chromatography.

2.4. AgNP biosynthesis conditions

The AgNPs were prepared with a 1:3 volume ratio of diluted AAPE rice husk extract to AgNO_3 , subjected to 5 min of ultrasonic vibration, and then reacted in a water bath. Biosynthesis

was performed under the following reaction conditions: AAPE rice husk extracts were diluted 2–50 times with methanol; the AgNO_3 concentrations used were 0.00001–1 M; and the reactions proceeded under pH 2–12 conditions for between 10 min and 5 months at 25, 50, or 80 °C. The bio-reduced aqueous components were used to obtain the UV–vis spectrum of the solution. Particle suspensions were diluted 10 times with distilled water to avoid errors due to the high optical density of the solution; measurements were performed at wavelengths of 300–700 nm. Appropriate AgNP biosynthesis conditions were preliminarily evaluated according to the sharpest peak at lower wavelengths in the UV–vis spectrum of the bio-reduced aqueous solution. The sharpest peak represents the highest yield and most even dispersion, and peaks at lower wavelengths indicate smaller AgNP sizes [8,15]. The biosynthesis of AgNPs was also performed using the HWE rice husk extract.

2.5. Analysis of the morphology of AgNPs

Three microliters of AgNP solution was dripped onto a formvar-carbon-coated 200-mesh copper grid (Ted Pella Inc., Redding, CA, USA), and the solvent was evaporated for 15 min. The sample was imaged through transmission electron microscopy (TEM; JEM-1400, JEOL, Peabody, MA, USA) at 80 kV to determine the size distribution and agglomeration of the particles.

2.6. Identification of phenolic acids contributing to AgNP biosynthesis

Because the nine main phenolic acids contained in rice husk are GA, proto CA, 4-hyd BA, 3-hyd BA, VA, CA, SA, *p*-Cou, and FA [23,24], phenolic acid mixtures consisting of eight standard phenolic acids were employed in the lack-one test. “Lack-one test” indicates that one of the phenolic acids was missing in each mixture and the concentration was adjusted to equal to that of rice husk extract, and the reaction environment used in the appropriate conditions for the biosynthesis of AgNPs prepared from AAPE (Table 1). The biosynthesis ability of various phenolic acids was evaluated by using the UV–vis spectra to identify the dominant phenolic acids contributing to the AgNP biosynthesis.

3. Results and discussion

3.1. Appropriate AgNP biosynthesis conditions

A transparent solution was observed when AAPE rice husk extract reacted with AgNO_3 at pH < 6, whereas a yellow–brown product was observed when the reaction environment was adjusted to pH > 6. The highest peak in the UV–vis spectra was obtained when the reaction environment was adjusted to pH 10, and no obvious peak was noted after the reaction at pH 4 (Fig. 1a).

The UV–vis spectra were used to determine the structure of the AgNPs based on their free surface electron plasmon oscillations. The peaks in the spectra reflect the size and shape of the AgNPs because they are surface plasmon

resonance (SPR) peaks [15]. The peak width and height of these SPR peaks indicate the dispersity and yield of AgNPs, respectively, and the peak shifts to a shorter wavelength as the AgNP size decreases [25]. In this study, no clear peaks were observed at pH 4 (Fig. 1a), which demonstrated that an acidic environment is detrimental to the synthesis of AgNPs using rice husk extract. By contrast, a high AgNP yield was obtained in an alkaline environment. These phenomena can be attributed to the dissociation of phenolic hydroxyl groups ($\text{p}K_{\text{a}2} > 8$) and an increase in the absolute value of the zeta potential stabilizing AgNP dispersion at a high pH [26], because phenolic acids are considered the major bioactive components in the biosynthesis of AgNPs using rice husk extract [23]. However, an extremely alkaline environment is not conducive to the formation of AgNPs.

All of the peaks were located within 445–450 nm, indicating that there was no substantial difference in the size of AgNPs formed in differently alkaline environments. The reactions at pH 9 and 10 resulted in higher and narrower peaks, which indicated that the AgNP yield was larger and that the particle size distribution was more even than for other pH levels. Therefore, a pH of 10 was identified as optimal and was used in the synthesis of AgNPs using AAPE rice husk extract in the follow-up experiments.

After 1-h synthesis reaction at various temperatures, peaks in the AgNPs UV–vis spectra were observed at 445 (25 °C), 450 (50 °C), and 455 nm (80 °C) (Fig. 1b). The peak became higher and broader as the reaction temperature increased, indicated that the yield increased and the particle size distribution became more uneven. Precipitate was actually observed when the reaction occurred at 50 and 80 °C. The peak corresponding to the AgNPs synthesized at 25 °C was the sharpest and was observed at the shortest wavelength, demonstrating that these AgNPs were smaller and more even. Khan et al. [7] reported that AgNP synthesis mediated using *Pulicaria glutinosa* extract was completed after 80 min and 20 h of reaction at 90 °C and ambient temperature, respectively. At high temperatures, there was insufficient time for the phenolic acids in the rice husk abstract to capture the rapidly synthesized AgNPs, which caused the NPs' flocculation. Therefore, 25 °C was concluded to be a desirable reaction temperature for AgNP synthesis.

The AgNP yield decreased when the AAPE rice husk extract was diluted. Almost no peak was observed when the synthesis was performed with the rice husk extract diluted 50 times, which was regarded as the limiting concentration for AgNP synthesis (Fig. 2a). A zigzag curve was observed in the spectrum when the stock solution of rice husk extract was used and precipitate was produced. The position of the peak was at a longer wavelength when a higher concentration of extract was used, as indicated by Fig. 2b, which shows the full-wavelength scanning spectra of the reaction products (reacted with 2–50 times extract dilutions) adjusted to the same peak height. This may have been because the strong reducing power of high-concentration rice husk extract rapidly formed a certain amount of AgNPs and there was insufficient time for the phenolic acids to prevent AgNPs aggregation, thus resulting in larger particles. Additionally, Ostwald ripening may have occurred during the 60-min reaction, with just-formed small AgNPs tending to achieve a thermodynamically stable state [7].

Table 1 – Phenolic acid mixtures in lack-one tests.

Phenolic acid	Concentrate in sample (ppm)									
	A ₀ ^a	Lack of GA	Lack of proto CA	Lack of 4-hyd BA	Lack of 3-hyd BA	Lack of VA	Lack of CA	Lack of SA	Lack of <i>p</i> -Cou	Lack of FA
Gallic acid (GA)	0.5	–	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5
Protocatechuic acid (proto CA)	0.9	0.9	–	0.9	0.9	0.9	0.9	0.9	0.9	0.9
4-Hydroxybenzoic acid (4-hyd BA)	5.4	5.4	5.4	–	5.4	5.4	5.4	5.4	5.4	5.4
3-Hydroxybenzoic acid (3-hyd BA)	170	170	170	170	–	170	170	170	170	170
Vanillic acid (VA)	22	22	22	22	22	–	22	22	22	22
Caffeic acid (CA)	22	22	22	22	22	22	–	22	22	22
Syringic acid (SA)	32	32	32	32	32	32	32	–	32	32
<i>p</i> -Coumaric acid (<i>p</i> -Cou)	433	433	433	433	433	433	433	433	–	433
Ferulic acid (FA)	98	98	98	98	98	98	98	98	98	–

^a A₀ represents the solution with a similar phenolic acid composition to the AAPE extract.

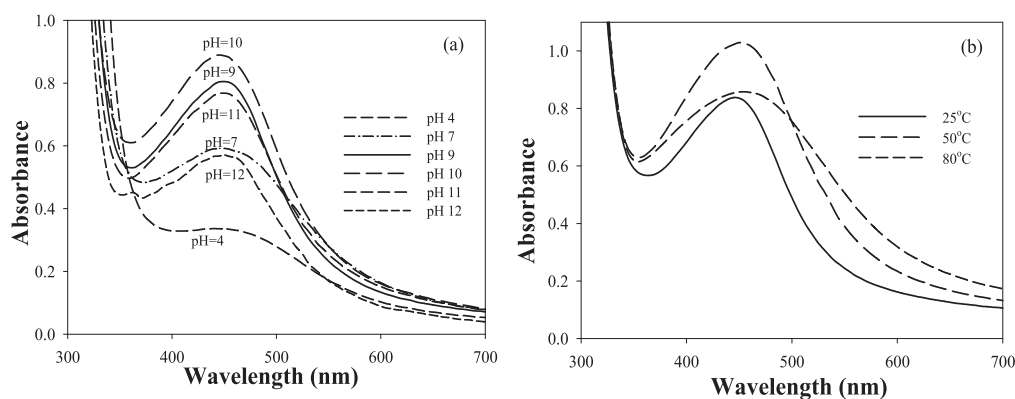


Fig. 1 – UV–vis spectra of AgNP biosynthesized using 1 mL of AAPE that had been diluted 10 times and reacted with 2 mL of 0.001 M AgNO₃ for 60 min under various pH (a) and temperature (b) conditions.

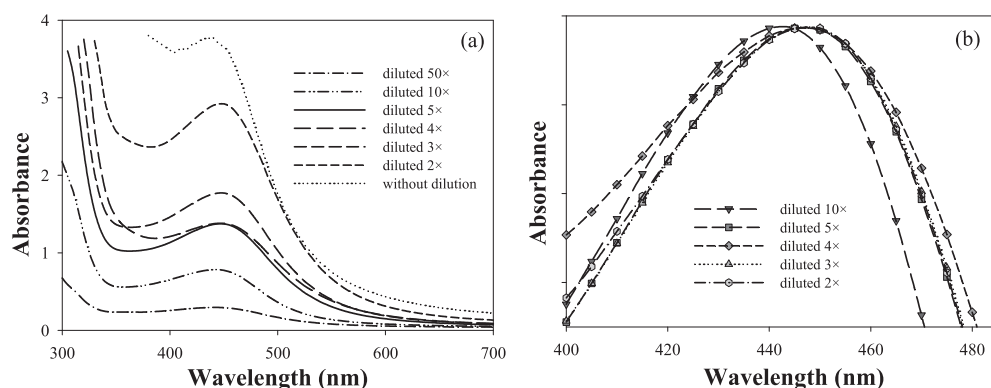


Fig. 2 – UV–vis spectra (a) and adjustment of peaks to equal peak height (b) of AgNPs biosynthesized using 1 mL of AAPE rice husk extract diluted to different concentrations, adjusted to pH 10, and reacted with 3 mL of 0.001 M AgNO₃ for 60 min at 25 °C.

A 10-times extract dilution resulted in the sharpest peak and the peak at the lowest wavelength; thus, this is suggested to be a desirable rice husk extract concentration for the production of a small and even AgNP distribution.

No peak was observed in the UV–vis spectra at 440–460 nm when the AgNO₃ concentration was lower than 0.0001 M (Fig. 3a), indicated that AgNPs could not be synthesized at such low concentrations. The peaks became broader and

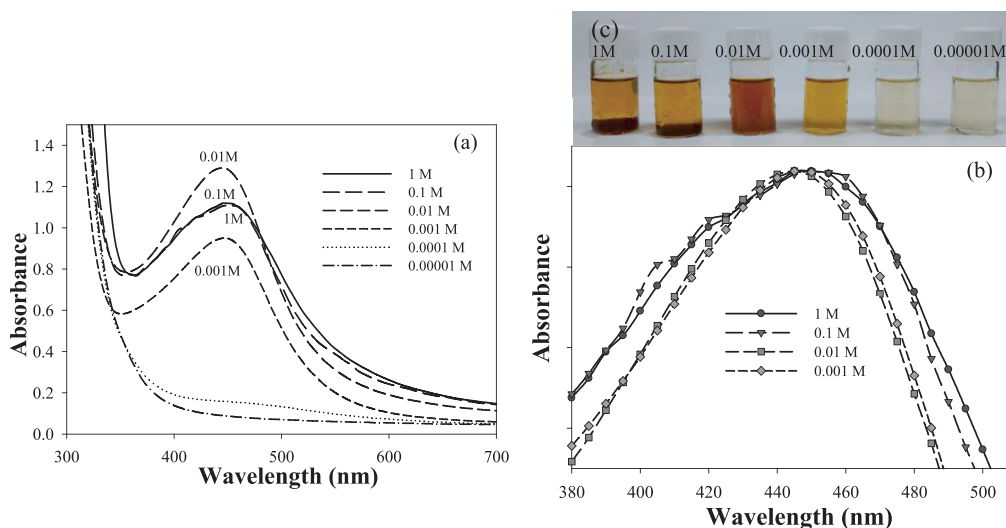


Fig. 3 – UV–vis spectra (a), adjustment of peaks to equal height (b), and visual observation (c) of AgNP biosynthesized using 1 mL of AAPE rice husk extract diluted 10 times, adjusted to pH 10, and reacted with 3 mL of AgNO_3 of different concentrations for 60 min at 25 °C.

clearly shifted to higher wavelengths when high concentrations were used, as indicated by a plot of the curves adjusted to the same peak height (Fig. 3b); the AgNPs thus became larger and more uneven. A zigzag curve was observed in the spectrum (Fig. 3b) and precipitates were observed (Fig. 3c) when the AgNO_3 concentration was increased to 0.1 M or 1 M. Similar results were obtained for AgNP biosynthesis using tansy fruit as the reducing agent [27]. Suspended particles were observed during the initial stage of synthesis when the 0.01 M AgNO_3 was used as the reactant; however, precipitates formed after setting for a few hours. An AgNO_3 concentration of 0.001 M is suggested as desirable as the reactant in AgNP biosynthesis, which enables the phenolic acids to fully perform the function of the dispersants to prevent precipitation.

The peaks at 445 nm became higher with longer synthesis times (Fig. 4), indicated that the AgNP yield increased with the

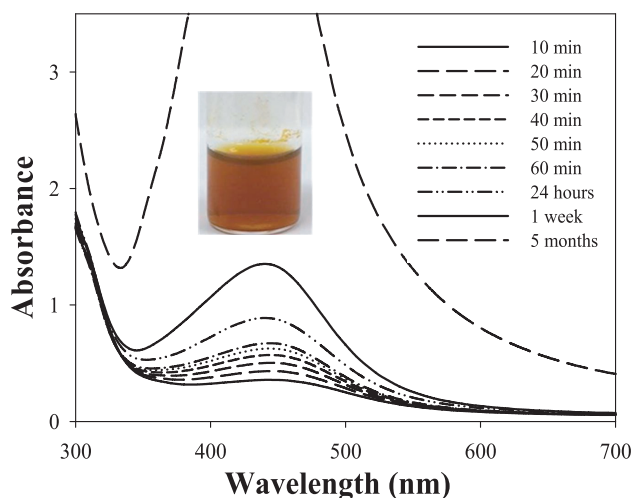


Fig. 4 – UV–vis spectra under appropriate conditions after different reaction times.

reaction or setting time. In a previous study on the synthesis of AgNPs from the aqueous extract of *O. sanctum* leaves, the plasmon peak and its position as a function of the reaction time did not change for 3–4 min, but the intensity increased because of nucleation. As the reduction proceeded, the increase in intensity was accompanied by a shift in the position of the peak toward higher wavelengths, indicating an increase in the size of the Ag particles because of diffusion growth, aggregation, or a combination of both [15].

The synthesis reaction proceeded quickly under the following conditions: 0.001 M AgNO_3 , AAPE rice husk extract diluted 10 times, pH 10, and reacted at 25 °C. Under these conditions, AgNPs could be detected through UV–vis spectrophotometry within 10 min reaction. Moreover, a darker AgNP solution without precipitates was obtained for the synthesis sample that was left for 5 months (inset of Fig. 4). This indicated that the reduction of Ag^+ and formation of AgNPs continued to occur when rice husk extract was used during the synthesis and setting, and that the phenolic acids, acting as dispersants, successfully captured the AgNPs and stabilized their structure [26,28] to prevent AgNPs aggregation and maintain their dispersion. No sedimentation for the AgNPs in aqueous solution during their setting for 5 months, these AgNPs can be inferred to be surrounded by phenolic acids that formed negatively charged layers and presented a space hindrance to prevent the aggregation of AgNPs by electrostatic repulsion and keep them dispersed evenly and stably.

3.2. Morphology of biosynthesized AgNPs

To further confirm the morphology of the synthesized AgNPs, TEM analysis of the AgNPs biosynthesized using AAPE and HWE rice husk extracts was performed; the results are presented in Fig. 5. The TEM images reveal that the AgNPs were spherical. The largest particles had diameters of 15.45 and 47.90 nm for AgNPs biosynthesized using AAPE and HWE rice husk extracts, respectively, and the particle size distribution

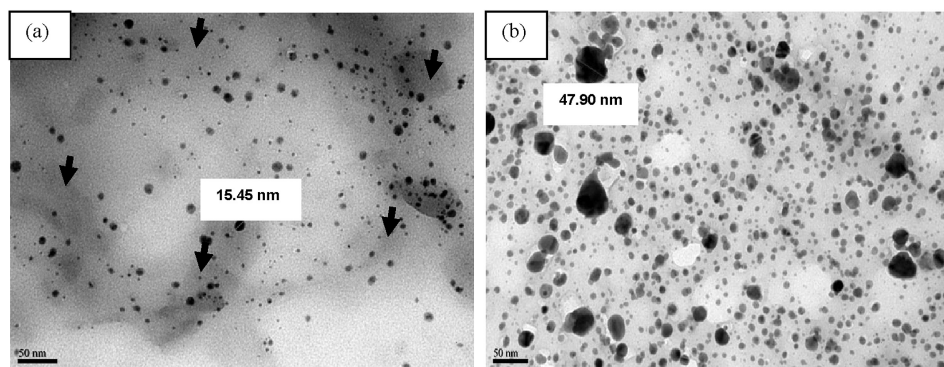


Fig. 5 – TEM images (300,000 \times) of biosynthesized AgNPs by using AAPE conditions (a) and HWE (b) rice husk extract.

when the AAPE was used was the more even. This may have been because more phenolic acids in the extract had a superior dispersion effect, which prevented AgNP aggregation.

When biosynthesized using a plant extract, AgNPs are captured and covered by phenolic compounds [15]. Baishya et al. [11] also discovered that AgNPs were capped with phenolic compounds of *Bryophyllum pinnatum* leaf extracts, and TEM images of the product exhibited a gray outer ring of AgNPs. In the present study, black AgNPs were dispersed in gray cloud-like regions which were darker in Fig. 5a than in Fig. 5b, indicated that the abundant phenolic compounds captured the AgNPs and prevented contact between them and their possible agglomeration. The growth in particle size due to the NP surface area effect was thus inhibited.

3.3. Dominant compounds in the reduction of Ag^+ during biosynthesis

Only CA changed the transparent Ag^+ solution to yellow–brown, as determined by visual observation, during AgNP synthesis using individual phenolic acids (inset of Fig. 6a). A UV–vis spectrum peak representing the characteristic absorption of AgNPs was observed at approximately 425 nm (Fig. 6a). These results revealed that only CA could independently reduce Ag^+ to form AgNPs, even though the *p*-Cou

content in rice husk extract is the largest [23], because CA has one more $-\text{OH}$ on benzene than *p*-Cou. This structure strengthens the resonance effect of benzene and C_3 , and the reduction reaction occurred more easily in an alkaline environment, facilitating the formation of AgNPs.

To further confirm that CA was the dominant compound reducing Ag^+ during AgNP biosynthesis with rice husk extract, lack-one tests were performed to analyze the effect of the absence of one phenolic acid in the mixture during the formation of AgNPs. The results demonstrated that AgNPs could be synthesized regardless of which phenolic acid was absent from the mixture, although their yellow–brown color was lighter than when the mixture containing all nine phenolic acids was used. A synergistic effect is suggested to occur when all the phenolic acids are included. Most of the peaks in the spectra obtained for AgNPs synthesized using the phenolic acid mixture were observed at approximately 430 nm, except that blue-shift and red-shift of the peaks occurred when the phenolic acid mixture was lacking *p*-Cou (425 nm) and CA (445 nm) in the lack-one tests, respectively (Fig. 6b). The red-shift corresponded to the formation of larger AgNPs and revealed that CA facilitated the synthesis of small AgNPs. By contrast, the blue-shift revealed that the synergistic effect among *p*-Cou and the other phenolic acids promoted faster synthesis and resulted in larger AgNPs.

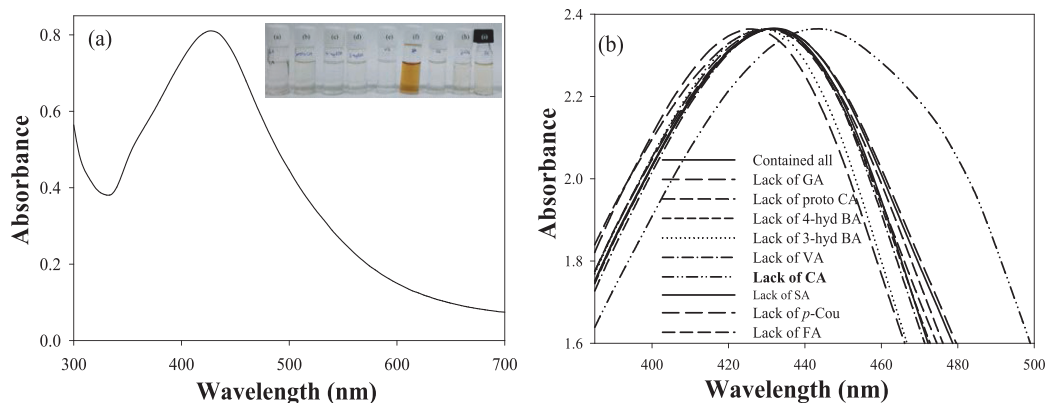


Fig. 6 – UV–vis spectrum of AgNP synthesized using 20 ppm CA under appropriate conditions (a) and using lack-one phenolic acid mixtures, adjusting to equal peak height (b). Inset of Fig. 6a: visual observation of AgNPs synthesized using 1 mL of GA (a), proto CA (b), 4-hyd BA (c), 3-hyd BA (d), VA (e), CA (f), SA (g), *p*-Cou (h), and FA (i) at pH 10 and with 3 mL of 0.001 M AgNO_3 , reacted for 60 min at 25 °C.

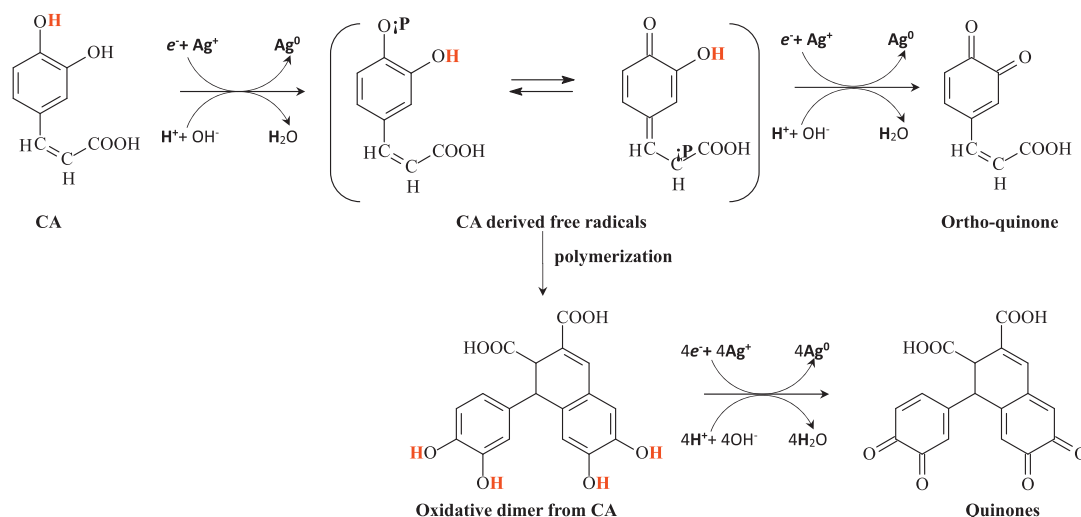


Fig. 7 – Possible mechanism of the formation of AgNP synthesized using CA in rice husk extract.

Most related studies in the literature have concluded that phenolic acids are the major bioactive components in the biosynthesis of AgNPs [7,15,29]. Phenolic acids have been reported to possess hydroxyl and carbonyl groups that can inactivate ions through chelation. This chelating ability of phenolic compounds is likely related to the high nucleophilicity of their aromatic rings [15]. Therefore, phenolic acids should have high antioxidant activity. According to the Fourier transform infrared spectroscopy results of Khan et al. [7], AgNPs were bound to oxygen derived from the hydroxyl groups in *P. glutinosa* compounds based on the formation of a new C=O group that was an aldehyde, ketone, or carboxylic acid. This finding suggested that Ag^+ was reduced by some hydroxyl groups and bound with C=O groups. A similar result was reported by Bankar et al. [30] in their study of AgNP synthesis using banana peel extract.

Tazaki et al. [31] reported that the hydroxyl group on CA releases one hydrogen ion and one electron to form a CA-derived free radical during oxidation. The CA-derived free radical is unstable and causes another hydroxyl group to release a hydrogen ion and electron. This oxidation forms a stable *o*-quinone compound. Another possible mechanism of oxidative dimerization of CA-derived free radicals occurs through a coupling reaction of semiquinone radicals as an intermediate of one-electron oxidation. Four hydroxyl groups on oxidative CA dimers may also be oxidized to release four hydrogen ions and four electrons to form a quinone with four ketones. Arakawa et al. [32] demonstrated that CA is more easily oxidized at a higher pH because two protons are released for the two-electron oxidation of CAF through the semiquinone.

Integrating the hypotheses of Arakawa et al. [32], Khan et al. [7], and Tazaki et al. [31] with the results of the present study, a possible mechanism is proposed for the formation of AgNPs synthesized using CA in rice husk extract, the reaction process of which is presented in Fig. 7. In an alkaline environment, CA releases electrons to reduce Ag^+ to become Ag and form CA-derived free radical. The CA-derived free radical reduces another Ag^+ and is itself oxidized to form *o*-quinone.

In addition, a CA dimer is formed by the coupling of two CA-derived free radicals. The CA dimer offers four electrons for the reduction of Ag^+ to Ag and is oxidized itself to form quinones. The *o*-quinone and other quinone compounds can combine AgNPs to form a steric hindrance around the AgNPs, which prevents aggregation and stabilizes the dispersion of AgNPs.

4. Conclusions

AAPE rice husk extract contains sufficient phenolic acids to biosynthesize AgNPs. Small spherical AgNPs with high crystallinity, even particle size distribution, and storage stability can be obtained under appropriate biosynthesis conditions. The synergistic effect of phenolic acids in rice husk extract was demonstrated to synthesize and stabilize AgNPs. An alkaline environment benefits the electron transfer of phenolic acids to reduce Ag^+ to form AgNPs, and the C=O on the formed quinone compounds bind and disperse AgNPs. CA is the dominant compound in rice husk extract contributing to the reduction of Ag^+ and can independently biosynthesize AgNPs. In addition to identifying a possible mechanism describing the formation of AgNPs using CA and other phenolic acids, this study manipulated the yield and characteristics of the obtained AgNPs through adjusting the conditions of the biosynthesis by using AAPE rice husk extract, demonstrating how the biosynthesis could apply to the requirements of distinct industries. These results proved the capability of biomaterials and revealed a potential application of rice husk as a crucial source of reducing agents and dispersants for the biosynthesis of AgNPs.

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