Biosynthesis, Characterization, and Antibacterial Activity of Silver Nanoparticles Produced from Rice Straw Biomass

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Silver nanoparticles (AgNPs) were synthesized from $AgNO_3$ using rice straw biomass as the reducing agent at room temperature *via* light irradiation. Full wavelength scanning with UV/Vis spectrophotometer was used to study the effect of light intensity, reaction time, and concentrations of rice straw biomass and $AgNO_3$ during AgNPs synthesis. Surface plasmon resonance (SPR) showed that the peak wavelength of synthesized silver nanoparticles arose at 425 nm, the optimal light intensity observed was 60,000 lx, and the optimal reaction time was 140 min. The optimum concentrations of the rice straw biomass and $AgNO_3$ used were 4 mg/mL and 2 mM, respectively. The AgNPs were characterized by X-ray diffraction (XRD) analysis. The zeta potential of AgNPs reached -21.2 mV. In addition, the AgNPs synthesized by rice straw biomass revealed antimicrobial activity against *Escherichia coli*, *Pseudomonas aeruginosa*, *Bacillus subtilis,* and *Staphyloccus aureus*. The inhibition rate reached about 97.17 ± 2.01% when the concentration of AgNPs solution used was 8 μg/mL. In the detection of antimicrobial effect of AgNPs and antibiotics, the antibacterial activity was found to be superior to that of antibiotics alone.

Keywords: Biosynthesis; Silver nanoparticles; Rice straw biomass; Antibacterial activity; Antibiotics

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INTRODUCTION

It is well known that silver nanoparticles (AgNPs) are widely used in fields such as photonics, catalysis, bio-nanomaterials, and medicine due to its inherent morphology, composition, and crystal structure (Njagi *et al*. 2011; Lee *et al*. 2013; Gambino *et al*. 2015; Ghaedi *et al*. 2015). At present, the preparation methods used for AgNPs are mainly physical, chemical, and/or biological methods (Kumar and Yadav 2009). The nano-silver particles obtained by physical methods have wide distribution, high purity, and strong activity, but the process involves the use of high-pressure and temperature conditions. The energy consumption is large and the resulting particles have a tendency to agglomerate, so that the performance of AgNPs is reduced (Zhang *et al*. 2014). Although the chemical method can yield the AgNPs particles with a narrow particle size distribution range and which are difficult to agglomerate, the reaction process employs strong reducing agents such as hydrazine, dimethylformamide, sodium borohydride, and other toxic organic reagents, which pose a serious threat to the environment (Huang *et al*. 2011; Hebbalalu *et al*. 2013). In recent years, due to the increasing concern towards the subject of green chemistry, by virtue of it being simple, efficient, and pollution-free, the biological method with microorganisms and plants have attracted the interest of

researchers (Rani and Rajasekharreddy 2011). Plant systems have been used as a green route as a reliable method for the biosynthesis of nanoparticles owing to their environmentally friendly nature (Kumar and Yadav 2009). This route is simple, uses mild conditions, short synthesis time, and convenient access to materials; it is thus more suitable for the synthesis of AgNPs (Huang *et al*. 2011). It has been reported that the plant materials can be used to synthesize AgNPs, such as *Ficus carica*, *Coffea arabica*, *Rosmarinus officinlis*, *Lawsonia inermis*, and so on (Gupta *et al*. 2014; Boras *et al*. 2015; Ghaedi *et al*. 2015; Dhand *et al*. 2016).

As a large agricultural country, China is a major producer of rice, wheat, and maize. In 2010, China's rice production accounted for 26% of the world's total rice, while wheat and corn accounted for approximately 20% (FAO 2012). Thus, China can produce about 506 million tons (MT) of crop straw dry matter per year, through the analysis of straw residues (Jiang *et al*. 2012). These large amounts of straw can be regarded as waste because of their low utilization rate. At present, the main uses of crop straw residues are composting (Iranzo *et al*. 2004), clean energy (Banik and Nandi 2004), feed, and also as industrial raw materials for direct burning (McCarty *et al*. 2006) or covering the surface of paddy fields (Abdelhamid *et al.* 2004). Straw residues used as fertilizers can improve soil fertility by balancing the farmland ecosystem and increasing soil microbial colony structure (Ji *et al*. 2012). But the labor cost is high and the effect of fertilization results in a large amount of organic matter loss. Therefore, a key issue is to improve the utilization of crop straw resources. Rice is a staple food for half of the world population consumption (Arvanitoyannis and Tserkezou 2008), and the amount of straw produced is immeasurable. This means, not only to China but to the world agriculture, that solving the problem of rice straw resource utilization is a fundamental issue.

As far as crop yields are concerned, wheat, maize, and rice are the most productive crop stalks in the world. Recent research concerning corn straw has been more comprehensive, mainly concentrated on bio-energy. Moreover, much research in this area is focused on the assessment of the supply chain or the exclusion of economic benefits (Batidzirai *et al*. 2016).Our previous work has included a preliminary study on the effect of the synthesis of AgNPs using wheat straw biomass (Ma *et al*. 2016). It was found that the AgNPs particles could be efficiently synthesized within 90 min, and its antibacterial effect was noticeable. The object of this study was to utilize rice straw to synthesize silver nanoparticles. The biosynthesis conditions were investigated, and the main properties of AgNPs were analyzed. The antibacterial activities of synthetic AgNPs on common gram-positive and gram-negative bacteria were studied. Also the antibacterial activity of AgNPs on antibiotics was explored.

EXPERIMENTAL

Preparation of Rice Straw Biomass

Rice straw was obtained from the experimental foundation of Chengdu Institute of Biology, Chinese Academy of Science, Wenjiang, China. The biomass solution was prepared according to a previous method with slight modifications (Raju *et al*. 2012). The rice straw was first dried under sunlight for 5 days, and then fine powders were prepared with a pulverizer (XFB-200, Zhongxiang Pharmaceutical Machinery, Human, China) at 25,000 rpm for 15 min. Accurately 10 g of straw powder was added to 200 mL of distilled water and mixed evenly. The sample was then mixed by ultrasonication (KQ-

400KDB, Kunshan Ultrasound Instrument, Inc., Suzhou, China), followed by standing for 10 min at room temperature (25±1 °C). The supernatant solution was filtered through a filter paper. The filtrate was stored at 4 °C for further experiments and used within 1 week (Song and Kim 2009).

Synthesis of AgNPs

The rice straw biomass was mixed with $AgNO_3$ solution at various concentrations at room temperature. The effects of light intensity, reaction time, rice straw biomass concentration, and $AgNO_3$ concentration on the AgNPs synthesis were investigated. When the final concentration of the biomass in the reaction mixture was 4 mg/mL and that of $AgNO_3$ was 1 mM, the effects of various light intensities (30,000; 40,000; 50,000; 60,000; and 70,000 lx) were detected by Digital Lux Meter AR823 (Smart Seneor, HK, China). Control tests were conducted in the dark. Then, under the optimal illumination condition, the influence of various reaction time (40, 60, 80, 100, 120, and 140 min) on the reaction was investigated. Control tests had a reaction time of 0 min. The effect of different biomass concentrations (2, 4, 6, and 8 mg/mL) was also studied under an optimum light intensity and reaction time.

The control group did not contain the rice straw biomass. Finally, the effects of different concentrations of $AgNO_3$ (1, 2, 4, and 6 mM) on the yield of AgNPs were investigated under optimum light intensity, reaction time, and concentration of biomass. The control group was not added with $AgNO_3$ (Dhas *et al*. 2014). The AgNPs solution was prepared under the optimum conditions of 15,500 ×g centrifugation (YZB/GER 1841-2014, Thermo Fisher Scientific Technology, China), for 30 min, and the precipitated nanoparticles were collected. The AgNPs thus obtained were lyophilized at -80 °C for about 8 h (LGJ-18, Xiongdi Instrument, Ltd., Zhenzhou, China), and used for subsequent research.

UV-Vis Test

1 mL of the reaction solution was diluted 5 times with distilled water, and the UV-vis spectrum of the samples were scanned from 330 to 700 nm on a spectrophotometer (Analytikjena/Biometra ScanDrop 100, Analytik Jena AG Corp., Jena, Germany).

Zeta Potential and Average Particle Diameter Analysis

The samples were centrifuged at 15,500 ×g for 30 min, washed twice with distilled water, and then suspended in distilled water. The suspension was then subjected to ultrasonic treatment at room temperature for zeta potential measurement using a Malvern Zetas sizer Nano ZSP (Malvern Instruments Ltd. Malvern, UK) (Wei *et al*. 2012). The particle size distribution and average diameter of the dispersed particles was carried out by a particle size distribution system (Malvern Zetas sizer Nano ZSP, Malvern Instruments Ltd. Malvern, UK). Using a scattering angle of 173°, all samples were measured for 1 min at (25.0±0.1) °C.

X-Ray Diffraction Analysis

The AgNPs were lyophilized, and the resulting powders were characterized using a Rigaku Smart Lab X-ray diffractometer (Rigaku Corp., TKY, Japan) using CuK_α radiation (I = 30 mA, V = 40 KV, the scanning range of 35-80 °, λ = 1.54 Å).

Antibacterial Activity Test

Inhibitory effect and zone of inhibition

After weighing, the freeze-dried AgNPs were dispersed in distilled water to produce a stock solution with final concentration of 1024 μg/mL. The working suspensions were diluted in distilled water (Wei *et al.* 2012; Pereira *et al.* 2014). About 150 μL broth solutions (10^9 CFU/mL) of *Escherichia coli* (ATCC 25922), *Pseudomonas aeruginosa* (ATCC 27853), *Bacillus subtilis* (ATCC 6633), and *Staphylococcus aureus* (ATCC 29213) were inoculated in 5 mL of TSB (Tryptic Soy Broth) medium (024051, Huankai Microbe Science and technology Ltd., Guang Dong, China) containing various concentrations of AgNPs (2, 4, 6, 8, and 10 μg/mL) at 37 °C, 220 rpm. The OD_{600} was measured after 4 h of culture. The assays were performed in triplicate, and the inhibitory effect (%) was calculated using Eq 1 (Lee *et al*. 2013):

$$\text{Inhibitory effect (\%)} = (CFU_{control} - CFU_{treatment}) / CFU_{control} \times 100\ \% \quad (1)$$

The well-diffusion method was used to determine the antimicrobial activity in the solid LB (Lysogeny Broth) agar plate. Approximately 0.1 mL of test bacteria solutions from above cultures (10^9 CFU/mL) were inoculated into 100 mL of LB agar. Approximately 50 μL of AgNPs solution with concentrations of 90 or 120 μg/mL was added to each wells. The plate was incubated at 37 °C for 12 h and the size of the inhibition zone (n=3) was observed and recorded.

Determination of Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal concentration (MBC)

The MIC value was determined by standard microdilution broth susceptibility methods (CLSI 2012). The AgNPs (1024 μg/mL) were diluted to 2-256 times in 100 μL of pure TSB in a 96-well plate. We added 100 μL of strains suspension (10^8 CFU/mL) to each well, and the assembly was then incubated at 37 °C overnight. The positive control contained no AgNPs, and the negative control had no added bacteria. The MIC is defined as the lowest concentration of the antimicrobial agent that inhibits the visible growth of the microorganisms after overnight incubation (Panacek *et al.* 2006). MBC of AgNPs was tested according to Dhas *et al.* (2014) with slight modifications, where 100 μL of the aliquot was spread in TSA (Tryptic Soy Agar) medium (024051, Huankai Microbe Science and technology Ltd., Guang Dong, China). The TSA plate was observed after 24 h cultivation at 37 °C. The MBC was defined as the minimum concentration of the antimicrobial that completely kills all microorganisms and results in no viable microbial growth (Panacek *et al.* 2006; Ghaedi *et al.* 2015).

Combinational Antibacterial Analysis of AgNPs with Antibiotics

The well-diffusion method was used to determine the antimicrobial activity of AgNPs solution after mixing with a standard concentration of antibiotic solution (Ampicillin 10, Kanamycin 30, clindamycin 10, gentamicin 30, or vancomycin 30). The final concentration of nano-silver in the mixture was 120 μg/mL. The culture without the addition of AgNPs was used as the control sample. About 100 μL of *E. coli* (ATCC 25922), *P. aeruginosa* (ATCC 27853), *S. aureus* (ATCC 29213) (10^9 CFU/mL) were inoculated into 100 mL of LB agar. Approximately 100 μL of antibiotic/(antibiotic + AgNPs) solutions were added to the wells (Dhand *et al*. 2016). The plate was then

incubated at 37 °C for 12 h. The diameter (mm) of the inhibition zone was observed and recorded. The increase in the fold area (n=3) was calculated by Eq. 2 (Birla *et al*. 2009):

$$Increase\ in\ fold\ area = \frac{(Zone\ of\ inhibition\ of\ combination)^2 - (Zone\ of\ inhibition\ of\ antibiotics)^2}{(Zone\ of\ inhibition\ of\ antibiotics)^2} \quad (2)$$

RESULTS AND DISCUSSION

Optimization of Parameters

Effect of light intensity

When the biomass solution was added to the $AgNO_3$ solution under light conditions, a color change of the reaction mixture from colorless to yellow to orange was observed. No such changes were observed for samples incubated in the dark. After 90 min, the color of the biomass-treated samples continued to deepen and appeared reddish brown at the end. This observation was similar to the phenomenon of the synthesis of AgNPs using leaf extracts of *Rhizophora acipulata* (Dhas *et al*. 2014) and *Rosmarinus officinalis* (Ghaedi *et al*. 2015). The change in color was due to the surface plasmon resonance (SPR) effect of AgNPs, which is typical of noble metal nanoparticles (Zhang *et al*. 2014). All the reaction mixtures displayed a maximum absorption at 425 nm in its UV-Vis spectra, while the dark treatment group showed no characteristic peak (Fig. 1A). The motion of free electrons on the surface of AgNPs, which when equal to the frequency of the applied electromagnetic wave, results in enhanced plasma resonance effect (Mehmood *et al*. 2014). In addition, the UV spectrum appeared smooth without significant sharp peaks, indicating that the synthesized particles AgNPs were homogeneously dispersed without agglomeration (Fu *et al*. 2006). With the increase of light intensity, the height of the characteristic peak gradually increased, which further revealed that the light intensity is very important for the synthesis of AgNPs. However, higher light intensity had no noteworthy effect on the synthesis, indicating that the reaction has reached saturation point.

Reaction time

The effect of reaction time on the synthesis efficiency of AgNPs under 60,000 lx illumination is shown in Fig. 1B. After 40 min of reaction, the synthesis of AgNPs exceeded 50%, and the maximum reaction occurred at 140 min, which was much shorter than that of *Calotropis gigantea*, which took 3 h (Rajasekharreddy *et al*. 2010). The yield of AgNPs obtained in this study was greater than the previous results (Huang *et al*. 2011; Raut *et al*. 2014).

Biomass concentration

No color change was observed in the control tests without rice straw biomass when treated with 60,000 lx light intensity for 140 min, which indicated that the rice straw biomass was necessary for the synthesis of AgNPs (Fig. 1C). When the concentration of biomass was at 4 mg/mL, the yield of AgNPs produced was the highest, which was much lower than that of Sorghum biomass samples (Njagi *et al*. 2011). With the increasing concentration of the biomass, the peak position was almost constant, which

further indicated that the synthesized AgNPs particles had similar particle size at various concentration levels, which was consistent with the results of Wei *et al*. (2012).

Silver nitrate concentration

Under the light intensity of 60,000 lx, with a rice straw biomass concentration of 4 mg/mL and a reaction time 140 min, the change of silver nitrate concentrations indicated different effects on the synthesis and particle size of AgNPs. With the increasing concentration of silver nitrate solution from 1 mM to 6 mM, the characteristic peak height increased first and then decreased, and the maximum yield was observed at 2 mM, while the peak height shifted to long wavelength region (Fig. 1D). The position of characteristic absorption peak of nanoparticles is related to the size of the nanoparticles. The larger the particle size is, the larger the characteristic wavelength is, and a red shift occurs (Huang *et al*. 2009). When the concentration of silver nitrate increases, a secondary reaction occurs on the surface of silver nanoparticles, resulting in increased particle size of nanoparticles.

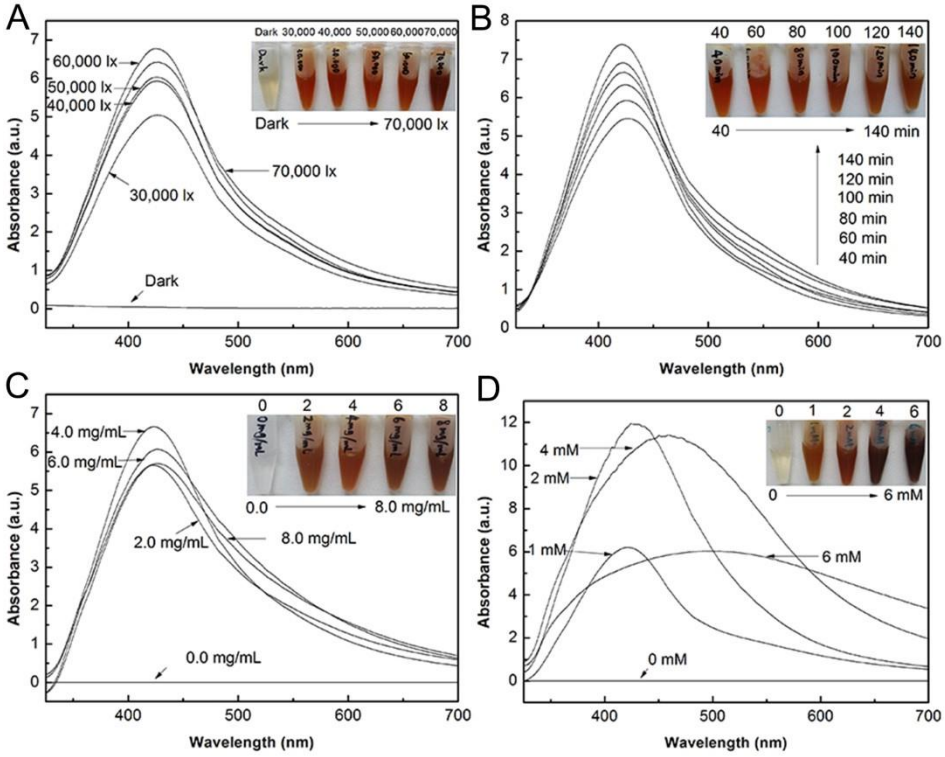

Fig. 1. Effects of reaction conditions on AgNPs synthesis with rice straw biomass: (A) light intensity; (B) reaction time; (C) biomass concentration; and (D) silver nitrate concentration

Characterization of AgNPs

Zeta potential and particle size distribution analysis

The zeta potential value is the total number of charges of the functional group with negative charge on the surface of AgNPs (Borase *et al*. 2013). The stability of the solution was characterized by zeta potential, the higher the absolute value was, and the electrostatic repulsion became stronger. The AgNPs in suspension became more dispersed and were difficult to agglomerate. This property is very important for the application and storage of AgNPs solution (Golińska *et al*. 2015). In the present study,

the zeta potential of AgNPs by rice straw biomass was -21.2 mV at pH 7.0 (Fig. 2A). The stability was found to be higher than using *Streptomyces albidoflavus* cell extract as the raw materials of AgNPs biosynthesis. The latter showed a zeta potential value of -8.5 mV, which was also less negative than that of the leaf of *Ficus carica* as raw material to synthesize AgNPs solution, where the zeta potential observed was -11.0 mV (Borase *et al*. 2015). Particle size analysis revealed that the average particle size of AgNPs was about 77.3 nm (Fig. 2B). Morones *et al.* (2005) demonstrated that the bonding strength between the AgNPs and the cell membrane depended on the surface will present a direct interaction; a higher percentage of surface will have a direct interaction with smaller AgNPs than larger ones. Ghorbani (2013) reported that the AgNPs synthesized by *Salmonella typhirium* showed an average diameter of 129.3 nm.

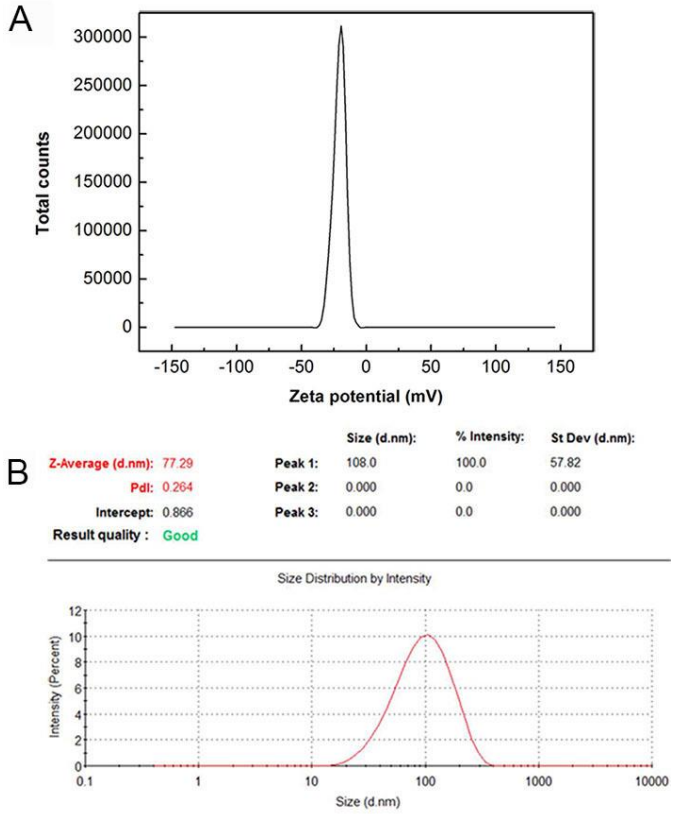

Fig. 2. Characterization of AgNPs by (A) zeta potential analysis and (B) size distribution, respectively.

X-ray diffraction (XRD) analysis

The AgNPs synthesized by the rice straw biomass were characterized by X-ray diffraction (XRD) analysis (Fig. 3). The XRD data were compared with the pure crystalline silver structure from the database of the Joint Committee on Powder Diffraction Standards (JCPDS, file nos. 04-0783) (ASTM04-0783 1999). Four intense diffraction peaks at 2θ values of 38.04°, 46.13°, 64.43°, and 76.61° were indexed to the 111, 200, 220, and 311 planes of silver reflections, respectively. The results demonstrated that the AgNPs synthesized from rice straw were composed of nanocrystals with the structure of an FCC (Patil *et al*. 2012) pattern. Other peaks shown in the diffraction spectrum might be the protein attached to the surface of the nano-silver particles or the biomass of the rice straw (Shankar *et al*. 2003).

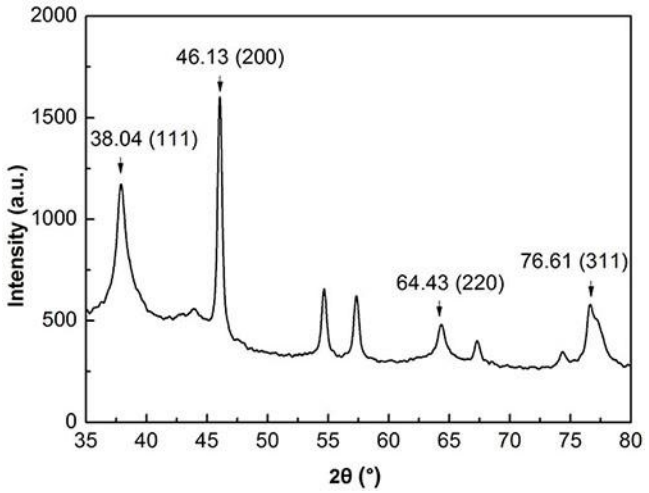

Fig. 3. Characterization of AgNPs by XRD. Four intense diffraction peaks at 2θ values of 38.04°, 46.13°, 64.43°, 76.61° corresponded to (111), (200), (220), and (311) planes of silver reflections, respectively.

Antimicrobial Performance of AgNPs

The antibacterial activity of AgNPs towards common gram-positive (*B. subtilis* and *S. aureus*) and gram-negative strains (*P. aeruginosa* and *E. coli*) (Fig. 4) were tested. The bacterial inhibition of AgNPs was found to be concentration-dependent (Fig. 4A), and the antibacterial activity increased with the increase of AgNPs concentration. When the concentration of AgNPs was 8 μg/mL, the inhibition rate of AgNPs to *P. aeruginosa* was about 97.17 ± 2.01%, which was higher than that of *S. aureus* (26.06 ± 0.20%), *E. coli* (45.64 ± 4.38%), and *B. subtilis* (80.10 ± 1.95%). This may be due to the increase of AgNPs concentration hindering *P. aeruginosa* biofilm formation, resulting in increased sensitivity of *P. aeruginosa* (Palanisamy *et al*. 2014). In contrast to the previous studies, Dhand *et al*. (2016) found that the synthesis of AgNPs from *Coffea arabica* seed extract was similar to that of *S. aureus* and *E. coli*, which was more sensitive to biosynthesis of AgNPs. In solid medium, Arun *et al*. (2015) and Wei *et al*. (2012) also reported that *B. subtilis* is more sensitive to AgNPs than *E. coli* (Fig. 4B). All these results implied that the AgNPs synthesized by rice straw biomass showed a substantial antimicrobial activity against gram positive and gram-negative bacteria.

The MIC and MBC of AgNPs against test microorganisms are presented in Table 1. These results demonstrate the AgNPs are effective in inhibiting gram-positive and gram-negative microorganism. Ghaedi *et al.* (2015) also reported lower MIC of AgNPs against *P. aeruginosa* (ATCC9027) (193.31 μg/mL) compared with *E. coli* (386.62 μg/mL) and *S. aureus* (773.24 μg/mL) and *B. subtilis* (1546.4 μg/mL) for AgNPs biosynthesized using *Rosmarinus officinalis* leaf extract as material.

Table 1. MIC and MBC of AgNPs

Strains	AgNPs	
	MIC (μg/mL)	MBC (μg/mL)
E. coli (ATCC25922)	16	32
P. aeruginosa (ATCC27853)	8	16
S. aureus (ATCC29213)	32	64
B. subtilis (ATCC6633)	16	32
MIC: minimum Inhibitory concentration; MBC: minimum bactericidal concentration; AgNPs: silver nanoparticles		

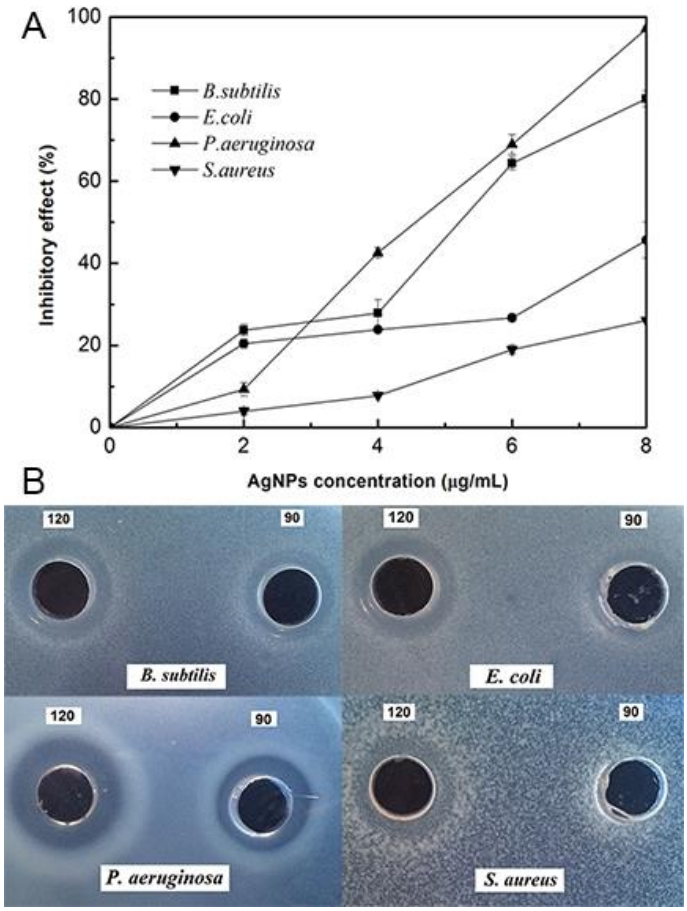

Fig. 4. Inhibitory effect of AgNPs on the growth of *B. subtilis*, *E. coli*, *P. aeruginosa*, and *S. aureus* in (A) TSB broth and (B) LB agar

Combinational Test of Antibiotics with AgNPs

The antibacterial effects of AgNPs mixed with Vancomycin, Kanamycin, Clindamycin, Gentamicin, and Ampicillin on the three pathogenic bacteria *E. coli*, *P. aeruginosa,* and *S. aureus* were found to be better than that of adding antibiotics alone. Bonde *et al.* (2012) came to a similar conclusion when AgNPs synthesized with *Murraya koenigii* were mixed with antibiotics, as well as previous report (Dar *et al*. 2012).

The AgNPs mixed with Kanamycin and Ampicillin showed strong antibacterial activity against gram-positive and gram-negative bacteria (Table 2), which was consistent with the results of Golińska *et al*. (2015). In particular, when AgNPs were mixed with Ampicillin, the antibacterial activity of *P. aeruginosa* was increased by adding antibiotics alone.

The calculated area multiplication factor was 0.82 (Fig. 5). The antibacterial effect of AgNPs mixed with Kanamycin on *S. aureus* was also considerably better than that of antibiotics added alone. The increased area multiplication factor in this case was 0.81, which was better than that of Birla *et al.* (2009), which was only 0.291. This is probably due to the use of rice straw biomass synthesis of AgNPs mixed with antibiotics to show a synergistic anti-bacterial effect (Borase *et al*. 2013).

Table 2. Antibacterial Activity of Antibiotics and Antibiotics Cooperate with AgNPs against *E. coli*, *P. aeruginosa*, and *S. aureus*

Antibiotics	*E. coli* (ATCC25922)			*P. aeruginosa* (ATCC27853)			*S. aureus* (ATCC29213)		
	A (mm)	A+AgNPs (mm)	Increase in fold area*	A (mm)	A+AgNPs (mm)	Increase in fold area*	A (mm)	A+AgNPs (mm)	Increase in fold area*
Vancomycin	16.08	16.88	0.10	15.36	16.36	0.13	15.46	16.98	0.21
Kanamycin	11.98	15.70	0.72	13.06	14.02	0.15	12.62	16.96	0.81
Clindamycin	21.98	23.00	0.09	22.02	22.64	0.06	22.40	24.10	0.16
Gentamicin	15.14	15.78	0.09	14.64	16.86	0.33	14.26	15.30	0.15
Ampicillin	13.10	15.80	0.45	13.38	18.04	0.82	13.04	16.48	0.60

Inhibition zones in diameter (mm) (n=3), and standard deviations were negligible. A: antibiotics; AgNPs: silver nanoparticles.
* The diameter of the well (8 mm) was used to calculate an increase in the fold area.

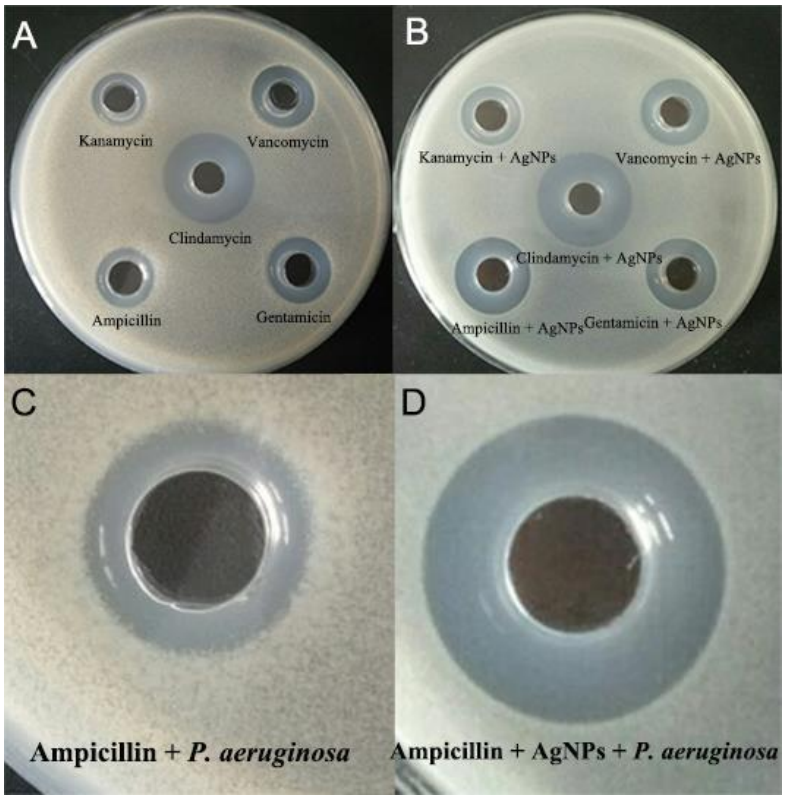

Fig. 5. (A) Zone of inhibition of *P. aeruginosa* with antibiotics; (B) AgNPs combined with antibiotics; (C) closer view of the zone of inhibition by ampicillin alone; and (D) AgNPs combined with antibiotics against *P. aeruginosa*

CONCLUSIONS

1. As a potential reducing agent and stabilizer, rice straw biomass can be used to synthesize silver nanoparticles efficiently under light conditions. The reaction conditions used were mild, the method utilizes green technology, and the synthesis efficiency was high.

2. When rice straw biomass was used as raw material to synthesize AgNPs, they promoted the diversification of the utilization and helped the recycling of straw resources.
3. XRD and zeta potential analysis showed that the synthesized AgNPs had tetrahedral centripetal crystal structure, which was consistent with the reported diffraction pattern of silver nanocrystals. The potential value of -21.2 mV, indicated that the AgNPs in solution had good stability, and suitable for long-term storage. The results of bacteriostatic experiments further confirmed the high efficiency and broad-spectrum antibacterial effect of AgNPs, especially for *P. aeruginosa*.
4. In the experiment of mixing with antibiotic solution, it was found that AgNPs can promote the inhibition of common antibiotics (*e.g.*, Ampicillin or Kanamycin) to various pathogens to varying degrees. The results showed that AgNPs may be used as adjuvant drug treatment by gram-positive bacteria or gram-negative bacteria that cause disease, revealing the potential application of AgNPs in biomedical and antibacterial materials.

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REFERENCES CITED

Abdelhamid, M. T., Horiuchi, T., and Oba, S. (2004). "Composting of rice straw with oilseed rape cake and poultry manure and its effects on faba bean (*Vicia faba* L.) growth and soil properties," *Bioresource Technology* 93(2), 183-189. DOI: 10.1016/j.biortech.2003.10.012

ASTM 04-0783 (1999). "Joint committee on powder diffraction standards powder diffraction files," ASTM International, West Conshohocken, PA.

Arun, G., Eyini, M., and Gunasekaran, P. (2015). "Green synthesis of silver nanoparticles using the mushroom fungus *Schizophyllum commune* and its biomedical applications," *Biotechnology and Bioprocess Engineering* 19(6), 1083-1090. DOI: 10.1007/s12257-014-0071-z

Arvanitoyannis, I. S., and Tserkezou, P. (2008). "Corn and rice waste: A comparative and critical presentation of methods and current and potential uses of treated waste," *Journal of Food Science and Technology* 43(6), 958-988. DOI: 10.1111/j.1365-2621.2007.01545.x

Banik, S., and Nandi, R. (2004). "Effect of supplementation of rice straw with biogas residual slurry manure on the yield, protein and mineral contents of oyster mushroom," *Industrial Crops and Products* 20(3), 311-319. DOI: 10.1016/j.indcrop.2003.11.003

Batidzirai, B., Valk, M., Wicke, B., Junginger, M., Daioglou, V., Euler, W., and Faaij, A. P. C. (2016). "Current and future technical, economic and environmental feasibility of maize and wheat residues supply for biomass energy application: Illustrated for

South Africa," *Biomass and Bioenergy* 92, 106-129. DOI: 10.1016/j.biombioe.2016.06.010

Birla, S. S., Tiwari, V. V., Gade, A. K., Ingle, A. P., Yadav, A. P., and Rai, M. K. (2009). "Fabrication of silver nanoparticles by *Phoma glomerata* and its combined effect against *Escherichia coli*, *Pseudomonas aeruginosa* and *Staphylococcus aureus*," *Letters in Applied Microbiology* 48(2), 173-179. DOI: 10.1111/j.1472-765X.2008.02510.x

Bonde, S. R., Rathod, D. P., Ingle, A. P., Ade, R. B., Gade, A. K., and Rai, M. K. (2012). "*Murraya koenigii*-mediated synthesis of silver nanoparticles and its activity against three human pathogenic bacteria," *Nanoscience Methods* 1(1), 25-36. DOI: 10.1080/17458080.2010.529172

Borase, H. P., Patil, C. D., Salunkhe, R. B., Suryawanshi, R. K., Salunke, B. K., and Patil, S. V. (2013). "Catalytic and synergistic antibacterial potential of green synthesized silver nanoparticles: Their ecotoxicological evaluation on *Poecillia reticulate*," *Biotechnology and Applied Biochemistry* 61(4), 385-394. DOI: 10.1002/bab.1189

Borase, H. P., Salunkhe, R. B., Patil, C. D., Suryawanshi, R. K., Salunke, B. K., Wagh, N. D., and Patil, S. V. (2015). "Innovative approach for urease inhibition by *Ficus carica* extract-fabricated silver nanoparticles: An in vitro study," *Biotechnology and Applied Biochemistry* 62(6), 780-784. DOI: 10.1002/bab.1341

CLSI (2012). "Methods for dilution antimicrobial susceptibility tests for bacteria that grow aerobically; approved standard, 9th ed.," *Clinical and Laboratory Standards Institute*, Wayne, PA, USA

Dar, M. A., Ingle, A., and Rai, M. (2012). "Enhanced antimicrobial activity of silver nanoparticles synthesized by *Cryphonectria* sp. evaluated singly and in combination with antibiotics," *Nanomedicine Nanotechnology Biology and Medicine* 9(1), 105-110. DOI: 10.1016/j.nano.2012.04.007

Dhand, V., Soumya, L., Bharadwaj, S., Chakra, S., Bhatt, D., and Sreedhar, B. (2016) "Green synthesis of silver nanoparticles using *Coffea arabica* seed extract and its antibacterial activity," *Materials Science and Engineering C* 58, 36-43. DOI: 10.1016/j.msec.2015.08.018

Dhas, S. P., John, S. P., Mukherjee, A., and Chandrasekaran, N. (2014). "Autocatalytic growth of biofunctionalized antibacterial silver nanoparticles," *Biotechnology and Applied Biochemistry* 61(3), 322-332. DOI: 10.1002/bab.1161

FAO (2012). "FAO Statistic Yearbook 2012: World food and agriculture," FAO Statistical Pocketbook World Food and Agriculture 79(7102), 572-574.

Fu, M., Li, Q., Sun, D., Lu, Y., He, N., Deng, X., Wang, H., and Huang, J. (2006). "Rapid preparation process of silver nanoparticles by bioreduction and their characterizations," *Chinese Journal of Chemical Engineering* 14(1), 114-117. DOI: 10.1016/S1004-9541(06)60046-3

Gambino, M., Marzano, V., Villa, F., Vitali, A., Vannini, C., Landini, P., and Cappitelli, F. (2015). "Effects of sublethal doses of silver nanoparticles on *Bacillus subtilis* planktonic and sessile cells," *Journal of Applied Microbiology* 118(5), 1103-1115. DOI: 10.1111/jam.12779

Ghaedi, M., Yousefinejad, M., Safarpoor, M., Khafri, H. Z., and Purkait, M. K. (2015). "*Rosmarinus officinalis* leaf extract mediated green synthesis of silver nanoparticles and investigation of its antimicrobial properties," *Journal of Industrial and Engineering Chemistry* 31, 167-172. DOI: 10.1016/j.jiec.2015.06.020

Ghorbani, H. R. (2013). "Biosynthesis of silver nanoparticles using *Salmonella typhirium*," *Journal of Nanostructure in Chemistry* 3(1), 1-4. DOI: 10.1186/2193-8865-3-29

Golińska, P., Wypij, M., Rathod, D., Tikar, S., Dahm, H., and Rai, M. (2015). "Synthesis of silver nanoparticles from two acidophilic strains of *Pilimelia columellifera* subsp. *pallida* and their antibacterial activities," *Journal of Basic Microbiology* 56(5), 1-6. DOI: 10.1002/jobm.201500516

Gupta, A., Bonde, S. R., Gaikwad, S., Ingle, A., Gade, A. K., and Rai, M. (2014). "*Lawsonia inermis*-mediated synthesis of silver nanoparticles: Activity against human pathogenic fungi and bacteria with special reference to formulation of an antimicrobial nanogel," *IET Nanobiotechnology* 8(3), 172-178. DOI: 10.1049/iet-nbt.2013.0015

Hebbalalu, D., Lalley, J., Nadagouda, M. N., and Varma, R. S. (2013). "Greener techniques for the synthesis of silver nanoparticles using plant extracts, enzymes, bacteria, biodegradable polymers, and microwaves," *ACS Sustainable Chemistry and Engineering* 1(7), 703-712. DOI: 10.1021/sc4000362

Huang, A., Xu, S., Wei, G., Ma, L., and Gao, C. (2009). "A trilayer dermal equivalent containing silver nanoparticles with enhanced antibacterial property," *Chinese Journal of Polymer Science* 27(6), 865-871. DOI: 10.1142/S025676790900459X

Huang, J., Zhan, G., Zheng, B., Sun, D., Lu, F., Lin, Y., Chen, H., Zheng, Z., Zheng, Y., and Li, Q. (2011). "Biogenic silver nanoparticles by *Cacumen Platycladi* extract: Synthesis, formation mechanism, and antibacterial activity," *Industrial and Engineering Chemistry Research* 50(15), 9095-9106. DOI: 10.1021/ie200858y

Iranzo, M., Cañizares, J. V., Rocaperez, L., Sainzpardo, I., Mormeneo, S., and Boluda, R. (2004). "Characteristics of rice straw and sewage sludge as composting materials in Valencia (Spain)," *Bioresource Technology* 95(1), 107-112. DOI: 10.1016/j.biortech.2004.01.013

Ji, X., Wu, J., Peng, H., Shi, L., Zhang, Z., Liu, Z., Tian, F., Huo, L., and Zhu, J. (2012). "The effect of rice straw incorporation into paddy soil on carbon sequestration and emissions in the double cropping rice system," *Journal of the Science of Food and Agriculture* 92(5), 1038-1045. DOI: 10.1002/jsfa.5550

Jiang, D., Zhuang, D., Fu, J., Huang, Y., and Wen, K. (2012). "Bioenergy potential from crop residues in China: Availability and distribution," *Renewable and Sustainable Energy Reviews* 16(3), 1377-1382. DOI: 10.1016/j.rser.2011.12.012

Kumar, V., and Yadav, S. K. (2009). "Plant-mediated synthesis of silver and gold nanoparticles and their applications," *Journal of Chemical Technology and Biotechnology* 84(2), 151-157. DOI: 10.1002/jctb.2023

Lee, H.-J., Song, J. Y., and Kim, B. S. (2013). "Biological synthesis of copper nanoparticles using *Magnolia kobus* leaf extract and their antibacterial activity," *Journal of Chemical Technology and Biotechnology* 88(11), 1971-1977. DOI: 10.1002/jctb.4052

Ma, Q., Li, J., Xu, L., Sun, Q., Yong, B., and Wang, Y. (2016). "Biosynthesis of silver nanoparticles using wheat straw biomass under light radiation and their antibacterial activity," *BioResources* 11(4), 10190-10200. DOI: 10.15376/biores.11.4.10190-10200

McCarty, J. L., Justice, C. O., and Korontzi, S. (2006). "Agricultural burning in the Southeastern United States detected by MODIS," *Remote Sensing of Environment* 108(2), 151-162. DOI: 10.1016/j.rse.2006.03.020

Mehmood, A., Murtaza, G., Bhatti, T. M., and Kausar, R. (2014). "Enviro-friendly synthesis of silver nanoparticles using *Berberis lycium* leaf extract and their antibacterial efficacy," *Acta Metallurgica Sinica* 27(1), 75-80. DOI: 10.1007/s40195-014-0025-7

Morones, J. R., Elechiguerra, J. L., Camacho, A., Holt, K., Kouri, J. B., Ramírez, J. T., and Yacaman, M. J. (2005). "The bactericidal effect of silver nanoparticles," *Nanotechnology* 16(10), 2346–2353. DOI: 10.1088/0957-4484/16/10/059

Njagi, E. C., Huang. H., Stafford, L., Genuino, H., Galindo, H. M., Collins, J. B., Hoag, G. E., and Suib, S. L. (2011). "Biosynthesis of iron and silver nanoparticles at room temperature using aqueous sorghum bran extracts," *Langmuir* 27, 264-271. DOI: 10.1021/la103190n

Palanisamy, N. K., Ferina, N., Amirulhusni, A. N., Mohd-Zain, Z., Hussaini, J., and Ping, L. J. (2014). "Antibiofilm properties of chemically synthesized silver nanoparticles found against *Pseudomonas aeruginosa*," *Nanobiotechnology* 12(1), 1-7. DOI: 10.1186/1477-3155-12-2

Panacek, A., Kvitek, L., Prucek, R., Kolar, M., Vecerova, R., Pizúrova, N., Sharma, V. K., Nevecna, T., and Zboril, R. (2006). "Silver colloid nanoparticles: Synthesis, characterization, and their antibacterial activity," *Journal of Physical Chemistry B* 110(33), 16248. DOI: 10.1021/jp063826h

Patil, R. S., Kokate, M. R., and Kolekar, S. S. (2012). "Bioinspired synthesis of highly stabilized silver nanoparticles using *Ocimum tenuiflorum* leaf extract and their antibacterial activity," *Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy* 91(91), 234-238. DOI: 10.1016/j.saa.2012.02.009

Pereia, L., Dias, N., Carvalho, J., Fernandes, S., Santos, C., and Lima, N. (2014). "Synthesis, characterization and antifungal activity of chemically and fungal-produced silver nanoparticles against *Trichophyton rubrum*," *Journal of Applied Microbiology* 117(6), 1601-1613. DOI: 10.1111/jam.12652

Rajasekharreddy, P., Rani, P. U., and Sreedhar, B. (2010). "Qualitative assessment of silver and gold nanoparticle synthesis in various plants: A photobiological approach," *Journal of Nanoparticle Research* 12(5), 1711-1721. DOI: 10.1007/s11051-010-9894-5

Raju, D., Mehta, U. J., and Ahmad, A. (2012). "Phytosynthesis of intracellular and extracellular gold nanoparticles by living peanut plant (*Arachis hypogaea* L.)," *Biotechnology and Applied Biochemistry* 59(6), 471-478. DOI: 10.1002/bab.1049

Rani, P. U., and Rajasekharreddy, P. (2011). "Green synthesis of silver-protein (core-shell) nanoparticles using *Piper betle* L. leaf extract and its ecotoxicological studies on *Daphnia magna*," *Colloids and Surfaces A: Physicochemical and Engineering Aspects* 389(1), 188-194. DOI: 10.1016/j.colsurfa.2011.08.028

Raut, R. W., Mendhulkar, V. D., and Kashid, S. B. (2014). "Photosensitized synthesis of silver nanoparticles using *Withania somnifera* leaf powder and silver nitrate," *Journal of Photochemistry and Photobiology B, Biology* 132C, 45-55. DOI: 10.1016/j.jphotobiol.2014.02.001

Shankar, S. S., Ahmad, A., and Sastry, M. (2003). "Geranium leaf assisted biosynthesis of silver nanoparticles," *Biotechnology Progress* 19(6), 1627-1631. DOI: 10.1021/bp034070w

Song, J. Y., and Kim, B. S. (2009). "Rapid biological synthesis of silver nanoparticles using plant leaf extracts," *Bioprocess and Biosystems Engineering* 32(1), 79-84. DOI: 10.1007/s00449-008-0224-6

Wei, X., Luo, M., Li, W., Yang, L., Liang, X., Xu, L., Kong, P., and Liu, H. (2012). "Synthesis of silver nanoparticles by solar irradiation of cell-free *Bacillus amyloliquefaciens* extracts and $AgNO_3$," *Bioresource Technology* 103(1), 273-278. DOI: 10.1016/j.biortech.2011.09.118

Zhang, M., Zhang, K., De, G. B., Verstraete, W., and Field, R. (2014). "The antibacterial and anti-biofouling performance of biogenic silver nanoparticles by *Lactobacillus fermentum*," *Biofouling* 30(3), 347-357. DOI: 10.1080/08927014.2013.873419

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