Research Article

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Characterizations and analysis of the antioxidant, antimicrobial, and dye reduction ability of green synthesized silver nanoparticles

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Abstract: The current study was conducted to assess the potential of ginger rhizome extract (*Zingiber officinale*) for the synthesis of silver nanoparticles (AgNPs) through the green method and its mitigating activity against pathogenic bacterial strains. AgNPs were synthesized

through a simple one-step approach and characterized by UV-Visible (UV-Vis) spectroscopy, powder X-ray diffraction (PXRD), transmission electronic microscopy (TEM), and energy dispersive X-rays spectroscopy (EDS). PXRD and TEM results of AgNPs showed the face central cubic structures and predominantly spherical structures with a size of 6.5 nm. EDS analysis confirms the elemental silver in nanoparticles. Moreover, the impact of the pH, as well as temperature, during the synthesis of AgNPs has also been investigated. At 25°C and pH 5, there was no significant peak for AgNPs in the absorption spectra. However, with an increase in temperature from 25°C to 85°C and pH 5 to pH 11, particles started attaining the spherical shape of different sizes due to an increase in the reduction rate. The AgNPs displayed effective results against selected pathogenic strains, *Pseudomonas aeruginosa* (MTCC 424), Methicillin-resistant *Staphylococcus aureus* (ATCC 43300), and fungus *Candida albicans* (KACC 30003). The prepared AgNPs exhibited excellent antioxidant activity and catalytic reduction of methyl orange with the pseudo-first-order rate constant of 3.9×10^{-3} .

Keywords: green synthesis, silver nanoparticles, plant extract, antibacterial activity, agar well diffusion

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1 Introduction

Silver NPs have extraordinary optical, electrical, and thermal properties and therefore are utilized in products ranging from cell to an organism. Other applications including photonic devices and molecular diagnostics take benefit of the exclusive optical characteristics of these nanomaterials [1]. The use of AgNPs is increasing in the mutual application of antimicrobial coatings. Therefore, the number of metallic nanoparticles' application increases for biomedical research, as therapeutic potential [2], contrast agents [3], organic synthesis [4,5], and theranostic agents for bio-imaging [6]. Different kinds of nanomaterials

developed such as copper [7], titanium, nickel [8], palladium [9], gold [10], and silver [11]. Among all, silver nanoparticles are the most efficient because they have a strong antimicrobial effect on fungus [12], bacteria [13], and other eukaryotic microorganisms, as theranostic [6] and antitumor agents [14]. Owing to exceptional characteristics such as excellent catalytic properties, conductivity, antibacterial activity, and chemical stability, colloidal silver is of specific interest [15]. Different procedures used for manufacturing nanoparticles frequently involve toxic, reducing agents and chemical solvents/surfactants [16–18], which typically produce a bulk amount of lethal wastes [19]. Hereafter, greener processes eliminate the use of dangerous reagents [20] and are of low cost, more economical, and alternative to the conventional approaches.

Many bacterial and fungal species were also explored for the synthesis of AgNPs [21,22], but most of them generate intracellular NPs. This synthesis always requires longer response time, so further extraction and retrieval steps are also required. On the contrary to microbial NPs synthesis, the plant-based green synthesis makes nanoparticles more biocompatible and environmentally friendly [23,24]. Most significantly, the process can be appropriately elevated up for bulk production of NPs. Many plants such as *Medicago polymorpha* [25], *Diospyros lotus* [26], *Lysimachia foenum-graecum* [27], *Convolvulus cneorum* [28], *Theobroma cacao* [29], *Duranta erecta* [30], *Olea ferruginea* [31], *Bridelia retusa* [32], and *Cucumis melo* [33] have shown the potential of formation of AgNPs.

Silver nanoparticles' antibacterial activity is the most promising in the drug delivery field, where they are the most interesting materials in clinical research due to their increasing microbial resistance to metal ions, antibiotics, and the development of resistance of various strains [34]. Owing to their large surface area, silver nanoparticles (AgNPs) are very significant compared to their volume. AgNPs were added because of their exceptional antibacterial properties in different applications, such as antibacterial devices, fibre-reinforced composites, food storage, drug delivery, cosmetic products, gas sensors, superconductive materials, electronic cryogenic parts, coatings, and other environmental applications [11,35].

Currently, many pathogenic bacteria have acquired resistance to a large number of antibiotics; *Candida albicans* (*C. albicans*) developed resistance against fluconazole and likewise *Staphylococcus aureus* (*S. aureus*) show resistance against methicillin. Similarly, *Salmonella typhi* showed resistance against a range of drugs including chloramphenicol, ampicillin, and trimethoprim. *Escherichia coli* (*E. coli*) acquired resistance to many antibiotics including tetracycline, streptomycin, ticarcillin, and ampicillin. This is an

alarming situation and a great challenge for researchers to develop new therapeutic ways to treat pathogenic bacteria [25]. Nevertheless, as the noble metal catalysts are costly and scarce, their large-scale production and utilization for antimicrobial activity get limited due to high cost. Recently, silver nanoparticles have established superiority on other metal catalysts due to significantly high stability as well as lower cost. The use of silver NPs possesses germicidal properties and plays an important part in the fight against drug-resistant microorganisms and different microbes [36]. AgNPs also showed remarkable activity against bacteria [37], fungi [38], viruses [39], and tumoricidal effects against diverse cell lines [40]; therefore silver colloids have shown potential in the area of nanomedicine.

Water pollution has recently become a major concern throughout the world. Many methods are used to treat sewage, such as photocatalysis, adsorption, catalysis, etc. Most industries like textiles, cosmetics, and plastics are manufacturing various synthetic dyes and using them for the colouring of their products [41]. For example, dyes with azo groups like MO are highly toxic and cancer-causing, resulting in a severe threat to aquatic organisms. Therefore, the catalytic reduction of these azo dyes is a key issue in the removal of pollutants. Currently, due to their superior catalytic efficiency, noble metal nanoparticles such as Ni, Cu, Pt, Au, Pd, and Ag have been widely used as catalysts [42]. Industrial wastewater contains a variety of toxicants such as dyes and other pollutants. These lethal compounds are commonly discharged into the ecosystem. Many scientists have paid great attention to these pollutants because they have prominent bad effects on life. Most of the pollutants, especially synthetic azo dyes, have a complex aromatic structure with azo groups and show a toxic effect when inhaled. Dyes retard sunlight penetration into water and decrease the levels of oxygen, which affect the growth of aquatic life due to decreased photosynthetic action. That is why, azo dyes are banned in several countries [43–45]. Thus, dyes remediation before their discharge into ponds and river is a necessary and concerned subject to make natural water free from these toxic dyes.

We report herein the synthesis of the silver nanoparticles through the reduction of aqueous Ag^+ and with ginger rhizome extract. Ginger is renowned for its medicinal values such as for curing skin diseases, colorectal cancer, arthritis, heart disease, antibacterial properties, and antioxidant activities [46]. Thus, during the current study, the greener cost-effective and entirely biogenic strategy was developed to synthesize silver nanoparticles and its properties were checked against various pathogenic bacterial strains. Additionally, the prepared

AgNPs were tested for their catalytic abilities via azo dye reduction.

2 Experimental

2.1 Materials

Silver nitrate (AgNO_3 , 99.99%), methyl orange, sodium borohydride, and 1-1-diphenyl-2-picrylhydrazyl were acquired from Sigma Aldrich (USA). Muller-Hinton and Sabourad Dextrose agar (SDA) media used were of ThermoFisher Scientific Canada. Fresh ginger was purchased locally and utilized for the study.

2.2 Extract preparation

The fresh ginger rhizome of 5 g was washed with distilled water followed by crushing. Afterwards, 100 mL of double-distilled H_2O water was added to it and simmered for 30 min, filtered, and kept at 4°C . This filtrate was later used for reduction in silver nanoparticles synthesis.

2.3 Biosynthesis of AgNPs

In phyto-synthesis protocol, 10 mL extract of ginger rhizome was added to the 90 mL of AgNO_3 (1 mM) solution and stirred at ambient temperature. The silver ions reduced to AgNPs soon after 10 min, indicated by the change in colour to reddish-brown. The mixture was centrifuged at 10,000 rpm and the resulting pellets were dispersed in double-distilled water and again centrifuged at 10,000 rpm and dried overnight and further stored for characterization. The kinetic study was carried out by optimizing the temperature and pH of the synthesized nanoparticles.

2.4 Characterization of AgNPs

The AgNPs were confirmed by different techniques. UV-Visible (UV-Vis) spectrum of the prepared materials was obtained by UV-Vis Carry 7000 spectrophotometer within a wavelength range of 200–800 nm. Transmission electronic microscopy (TEM) micrographs were recorded on

FEI Tecnai G2 Spirit Twin TEM instrument. The X-ray diffraction spectroscopy was employed to examine the phase variety, crystallinity, and grain size of AgNPs and spectra were obtained using RigakuUltima IV X-ray diffractometer equipped with a $\text{Cu K}\alpha$ sealed-tube source operating at a voltage of 45 kV and a current of 40 mA in a scattering range (2θ) of 20° – 84.9° with a step size of 0.04° and a counting time of 1 s per step.

2.5 Antibacterial activities

Antimicrobial assessment of the biogenic AgNPs was checked against the different microbial isolates such as *Pseudomonas aeruginosa* (*P. aeruginosa*), Methicillin-resistant *S. aureus*, and *C. albicans* on Muller-Hinton agar (MHA) plates and SDA by using agar well diffusion method. SDA was especially used for *C. albicans*. The bacterial culture were evenly spread on MHA plates with the help of sterilized cotton swabs. Cork borer was used for making wells in the plates. Afterwards, 50 μL of 100 ppm of prepared AgNPs was added in the respective wells [47]. Gentamycin disc was used as control and incubated for 24 h at 37°C .

2.6 Antioxidant activity

The antioxidant activity of prepared AgNPs was assessed by observing the quenching DPPH free radical to their stability. A reaction mixture has 2 mL of DPPH (40 mM) and also 2 mL of the ginger-mediated silver NPs solution. Three different concentrations of 50, 100, and 200 $\mu\text{g/mL}$ of freshly prepared AgNPs were used during the assay, then incubated and monitored in the dark at room temperature for 30 min. The activity of the AgNPs was measured at 517 nm using UV-Vis spectrophotometer (Carry 7000 spectrophotometer). For positive control, ascorbic acid was used. Radical scavenging activity (RSA) in percentage inhibition was calculated by using Eq. 1:

$$\% \text{RSA} = \left[\frac{(A_{\text{DPPH}} - A_{\text{solution}})}{A_{\text{DPPH}}} \right] \times 100 \quad (1)$$

where A_{DPPH} and A_{solution} indicate the absorptions of DPPH at 517 nm before and after adding AgNPs correspondingly. IC_{50} ($\mu\text{g mL}^{-1}$) was also evaluated, which denotes the concentration of antioxidant material required to reduce the initial concentration of DPPH radical by 50%.

2.7 Catalytic reduction of dye

The catalytic ability of AgNPs was evaluated for organic azo dye methyl orange (MO) reduction. In the reduction reaction, 2.5 mL of (0.04 mM) MO was added in a UV quartz cuvette followed by the addition of 0.5 mL of 0.1 M aqueous sodium borohydride and reading was taken. Then add 5 mg of AgNPs. The reaction rate was monitored with an interval of 1 min using a UV-Vis Shimadzu-1800, Japan, spectrophotometer.

3 Results

The maximum absorbance was shown by UV-Vis spectra at 430 nm, which is the characteristic of AgNPs. The effect of different temperatures (25°C, 45°C, 65°C, and 85°C) on the synthesis of AgNPs was also examined (Figure 1a). The synthesis of AgNPs was also studied at various pH (5, 7, 9, and 11). The results of the UV-Vis spectroscopy indicated that at acidic pH of 5, no significant SPR peak of AgNPs was observed and it remained light brown (Figure 1b).

However, at pH 7, the reaction solution of AgNPs turned light yellow with strong SPR peak at 430 nm at 45°C (Figure 1b). On the other hand, at pH 9 and 11, the solution of AgNPs developed intense dark red colour with strong SPR peaks at 430 nm within a few minutes (Figure 1c and d).

The crystallinity of AgNPs was confirmed by XRD and peaks detected at 2θ value of 38.02, 45.85, 64.32, and 77.8 can be indexed to the planes of (111), (200), (220), and (311), respectively, as shown in Figure 2. The size was calculated by Scherrer's equation [48]. The average crystallite size calculated from the (111) peak was 7 nm (± 3 nm). The small peak obtained at 31.69 may correspond silver oxide indicated by (111*) (JCPDS No.: 75-1532). The additional peaks seen at 2θ range of 27.68 and 43.76 may be due to the crystallization of inorganic compounds of extract.

TEM analysis revealed the primarily spherical and well-dispersed silver nanoparticles. Some of the nanoparticles showed an irregular shape with a small level of aggregations (Figures 3 and 4). Mostly, the nanoparticles (as shown in the TEM histogram Figure 4c) were polydispersed with spherical and face-centred cubic structure ably supported

Figure 1: UV-Vis spectra of ginger-synthesized AgNPs at different pH and temperature (a) at pH 5, (b) at pH 7, (c) at pH 9, and (d) at pH 11.

Figure 2: Powder XRD pattern of AgNPs.

by XRD pattern also. The average size of the AgNPs calculated from the TEM histograms was ~ 6.5 nm.

Elemental analysis was performed by EDS (energy dispersive spectroscopy) method. Generally, metallic AgNPs demonstrate characteristic optical absorption

peak at 3 keV (Figure 4). Besides, other peaks for oxygen and carbon were also observed.

FT-IR study was performed for the detection of reducing and stabilizing agents of prepared Ag NPs. FTIR spectra revealed an absorption peak in the region of $3,178\text{--}3,264\text{ cm}^{-1}$ as revealed in Figure 5. Besides the peak at $3,200\text{ cm}^{-1}$, some other peaks were observed at $2,890$, $1,589$, $1,305$, $1,016$, and 541 cm^{-1} in the FTIR spectra of AgNPs.

The antimicrobial efficacy of the AgNPs against the selected microbial strains, namely, *P. aeruginosa*, Methicillin-resistant *S. aureus*, and *C. albicans*, was assessed by well diffusion method. For antibacterial activity, $50\text{ }\mu\text{L}$ of 100 ppm of freshly prepared AgNPs solution was added in each of the respective wells and antibiotic disc of gentamycin was used as a control followed by incubation at 37°C for 24 h.

The antibacterial activities of the AgNPs were demonstrated in Figure 6. AgNPs were more successful against Gram-positive [49]. This dissimilarity can be due to cell wall diversity. Gram-positive bacteria contain peptidoglycan, while Gram-negative cells contain lipopolysaccharides [50]. It can be assumed that AgNPs act quickly on naked peptides on the wall of Gram-positive bacteria

Figure 3: (a and b) Low magnified; (c and d) high magnified TEM images of AgNPs.

Figure 4: (a and b) Low to high magnification TEM images; (c) histogram of AgNPs; (d) EDS investigation of AgNPs.

Figure 5: FTIR spectra of synthesized AgNPs.

and their uptake within the cell have also been reported for *P. aeruginosa* and *Salmonella typhus* [51]. AgNPs exhibited a zone of inhibition against all microbes, 5 mm *C. albicans*, 4 mm *P. aeruginosa*, and 7 mm *S. aureus*. All strains showed resistance against gentamycin.

Free radical scavenging capacity of ginger extract-mediated AgNPs was elevated through DPPH assay. The

antioxidant activity of aqueous ginger extract-mediated AgNPs is shown in Figure 7. Our prepared AgNPs show superior radical inhibitory DPPH activity, suggesting a source of an antioxidant. For antioxidant activity, various concentrations of 10, 50, and 100 $\mu\text{g/mL}$ of the ginger extract synthesized AgNPs and ascorbic acid-positive control was utilized to react with the 50 mM DPPH solution of equimolar concentrations. However, the DPPH radical scavenging abilities of the AgNPs ($64.20\% \pm 0.12\%$) were higher to a small extent than those of the standard ascorbic acid, which showed that the AgNPs have the proton-donating ability and could serve as free radical inhibitors or scavengers, acting possibly as primary antioxidants. A linear regression analysis was applied to determine IC_{50} value for the sample [52]. The value of IC_{50} for AgNPs was $68 \mu\text{g mL}^{-1}$. The relative order of DPPH radical scavenging activity was found to be: AgNPs > ascorbic acid > extract.

Dyes are the organic colouring chemical substances which are widely used in the plastic, paper, cloth, paint, pharmaceutical, meat, leather, and cosmetic industries. Throughout the world, their existence in water bodies poses a serious threat to mankind. Their complete eradication

Figure 6: Antimicrobial activity of AgNPs and gentamycin (G) on (a) *C. albicans*, (b) *P. aeruginosa*, and (c) methicilne-resistant *S. aureus*.

Figure 7: DPPH free radical scavenging activity of prepared AgNPs.

from ground water is therefore very necessary and also a matter of wide-ranging study [53,54]. The catalytic efficiency of the prepared Ag nanocatalyst was evaluated using the catalytic reduction of MO dye on an aqueous medium. For the reduction study, the dye solution was taken in a 4 mL UV quartz cuvette and sodium borohydride solution was used as a reducing agent. The colour of the dye solution MO quickly disappeared with the addition of catalyst AgNPs, indicating the reduction of the dye in solution. Figure 8a shows the catalytic reduction of MO dye by sodium borohydride in the presence of AgNPs catalyst and their possible products to hydrazine derivative.

In the current study methyl orange dye was used as a model organic synthetic pollutant. The catalytic efficiency of the synthesized AgNPs catalyst was examined in the reduction of MO dye. The UV-Vis spectroscopic time-dependent measurement of the MO reduction by sodium borohydride is shown in Figure 8a. It is clear from Figure 8 that the MO reduction took place very quickly after the

AgNPs catalyst was added to the solution containing MO and NaBH_4 . These show that the AgNPs were to be responsible for the catalytic reduction of MO and the likely cleavage of the azo bond ($-\text{N}=\text{N}-$) present in the chemical structure of MO and thus decolouring the MO solution (Figure 8c). The adsorption peak change was monitored overtime at 465 nm for MO dye. Besides, the appearance of a new absorption peak at 252 nm over the reaction time demonstrates the formation of new aromatic compounds as demonstrated in Figure 8a.

It has been reported that aqueous NaBH_4 alone cannot reduce MO. The reduction of MO by NaBH_4 is thermodynamically favourable at the ambient condition as borohydride ions are strong reductant in aqueous medium but kinetically very slow. Reduction of MO was carried out with NaBH_4 alone as shown in Figure 8c. The results showed that NaBH_4 could not reduce MO and the reduction was very slow as reported in the literature.

Similarly, another reaction was carried out by using AgNPs alone in the reduction of MO. The chemical reaction of MO reduction by using AgNPs nanoparticles alone is shown in Figure 8d, which demonstrates that alone AgNPs could not reduce MO. Metal nanoparticles catalyze this reaction by facilitating relay of electrons from the donor NaBH_4 to the acceptor MO molecule to overcome the kinetic barrier. The reduction of MO thus cannot be proceeded favourably in the absence of catalyst as evident here experimentally.

4 Discussion

It has been reported in the literature that increases in temperature increase the formation of nanoparticles by

Figure 8: (a) Catalytic reduction of MO by AgNPs and NaBH₄, (b) $\ln A_t/A_0$ of the MO by AgNPs and NaBH₄ (inset show percent reduction of MO), (c) MO reduction by AgNPs alone, (d) MO reduction by NaBH₄ alone.

increasing the synthesis rate of AgNPs. In contrast to what previously reported by *Pulicaria glutinosa* plant extract AgNPs, the current study revealed that by increasing temperature, no significant increase in the SPR of AgNPs was observed [55]. This difference in temperature effect might be due to the synthesis effectiveness of ginger extract, as in the current study we obtained maximum yield of AgNPs at 45°C.

The reaction temperature had a considerable effect on the size and morphology of the manufactured AgNPs. Figure 1a–d indicate the difference in the absorption spectra of AgNPs synthesized at different temperatures using aqueous AgNO₃ and ginger extract. The reaction temperature was changed from 25°C to 85°C. It was perceived that with the rise in the reaction temperatures, the absorption peak shifted towards lower wavelength, i.e. from 430 to 411 nm, which recommended a decrease in particle size with an increase in temperature [56]. At 25°C, there was no significant peak in the absorption spectra, stating that there was no formation of AgNPs (Figure 1a). The shift in the absorption peak was due to the localization of surface plasmon

resonance of the AgNPs. This proposes that the size of the synthesized AgNPs decreases with increasing temperature, which was probably due to the faster reaction rate at a higher temperature. At high temperature, the kinetic energy of the molecules increases and silver ions get ingested faster leaving less possibility for particle size growth. Thus, smaller particles of nearly uniform size distribution are formed at higher temperature [57].

pH is another foremost component which affects the size, shape, and morphology of fabricated AgNPs. The considerable effect of the reaction pH is its potential to alter the electrical charges of the biomolecules present in ginger, which might change their reducing and capping ability and the upcoming growth of nanoparticles [58]. Figure 1b–d showed the effect of pH on the absorption spectra of AgNPs prepared using the ginger extract. It was observed that with an increase in pH, the absorption peak shifted towards higher wavelength from 430 to 411 nm, indicating an increase in the size of synthesized AgNPs. As the diameter of the particle increases, the energy required to excite the surface plasmon electrons decreases.

As a result, the absorption maximum shifted towards longer wavelength. In addition to the spectral shift, there was an increase in the absorption intensity with increase in pH. Moreover, it was observed that higher pH increases the rate of reduction as the colour of the solution turned colloidal brown more instantly as compared to a solution of lower pH. Hence, alkaline pH is fantastic for the synthesis of AgNPs [59].

The bio-reduction of Ag was assumed as capturing of Ag^+ ions on the phytochemicals surface due to electrostatic interactions between the silver ions and phytochemicals in the ginger extract. These phytochemicals reduce the Ag^+ ions, leading to their secondary structure change and fabrication of silver nuclei. These formed silver nuclei consecutively grow by the further reduction of Ag^+ ions and their build-up at the nuclei, leading to the synthesis of AgNPs. Moreover, it has been reported that the mechanism behind the plant-mediated synthesis of nanoparticles is a plant-assisted reduction due to phytochemicals as presented in Scheme 1. Thus, the diversity of secondary metabolites (like flavonoids, saponins, polyphenols, terpenoids, etc.) in the aqueous ginger extract is responsible for the reduction of AgNPs. A plausible mechanism of the reduction is given in Scheme 1.

The detected XRD peaks at 2θ value of 38.02, 45.85, 64.32, and 77.8 represent the indexes planes of (111), (200), (220), and (311), respectively, as shown in Figure 2 and confirmed structure as face-centred cubic structure of Ag-NPs [60].

The TEM size of AgNPs was distinctive as it was smaller than previously reported plant-mediated AgNPs [25,28,30], where AgNPs of 6.5 nm size range were synthesized as shown in Figure 3. The small size of AgNPs at basic pH 9 makes them more successful in its applications.

EDS pattern of AgNPs is presented in Figure 4. The strong peak at EDS pattern around 3 keV is because of the binding energies of Ag [41,61]. Results obtained coincide with the peak value obtained for AgNPs synthesized by using *Coleus aromaticus* [62]. Similarly signals correspond to C and O were also found, which point toward the occurrence of plant phytochemicals as capping elements on nanoparticles surface. The results confirm the formation of highly pure AgNPs.

Scheme 1: A plausible mechanism for synthesis of AgNPs by plant extract.

FTIR spectrum is presented in Figure 5. The strong band at 3,200 cm^{-1} range could be owed to the bending and stretching vibrations for H–O of AgNPs. The band at 2,890 cm^{-1} may be accredited to the C–H stretching vibration. Likewise, the moderate peak at 1,589 cm^{-1} corresponds to N–H of amide band. The peak at 1,305 cm^{-1} corresponds to the carboxylic acids C–O stretching vibration of and 1,016 cm^{-1} could be a representation C–N stretching vibrations of aliphatic amines. The small peak at 541 cm^{-1} corresponds to a metal–oxygen bond representing AgNPs.

Our prepared AgNPs revealed higher activity for Gram-positive Methicilne-resistant *S. aureus* as compared to the Gram-negative bacteria *P. aeruginosa* as presented in Figure 6c. A significant activity of AgNPs could be accredited to the severe damage of the bacterial cell membranes due to their minute size, of which lead amplifies the permeability of cell membrane and their destruction. The antibacterial mechanism of nanoparticles is still being searched and not understood to date completely; however, the action of AgNPs entails the development of irregular “pits” in the cell wall of microbes, which finally resulted in cell death. The bactericidal action of the AgNPs is originated by their contact with the bacterial cytoplasm; in brief, the silver ions enter through ion channels in the membranes. The ribosome denaturing results in the inhibition of expression of enzymes and therefore destroying the cell [25].

The free radical activity of our prepared AgNPs in comparison to control showed higher free radical activity. Most importantly, our prepared AgNPs showed admirable free radical activity with 66% DPPH scavenging capability (Figure 7), which are comparable to the recently reported *Asphodelus aestivus*-synthesized AgNPs [63]. The antioxidant activity of aqueous extract shows lower free radical activity than previously reported plant extract [64]. Our nanoparticles showed stronger antioxidant activity than the extract of ginger at all three concentrations. The reported activity was apparent not just because of the capping agents of ginger extract, but also because of the silver nanoparticles and may prove useful to treat many diseases due to oxidative stress and to deal with various infections.

Scheme 2: Proposed mechanism for the reduction of MO dye by AgNPs catalyst in the presence of NaBH₄.

The degradation of MO on the substrates decorated with Ag nanostructures is likely to proceed through the mechanism depicted in Scheme 2. The degradation process by catalysts relies on the electrons transfer from the donor (borohydride) to the acceptor (dye). The adsorption of both the MO molecules and BH₄⁻ ions on the surface of the nanoparticles is the initial step for the dye reduction. The silver nanocatalysts thus serve as electron relay systems which facilitate the transfer of an electron between the nucleophilic borohydride and electrophilic MO [65]. The high catalytic activity of our system could be related to the high surface coverage, large surface to volume ratio, and regular shape of the AgNPs, which make a possible transfer of electron and enable overcoming the kinetic barrier for the reduction reaction. Since the reaction occurs on the surface, increasing the area enhances the rate of degradation.

The new peak formation at 252 nm with increasing intensity is considered from the hydrazine derivatives compound of MO [66,67]. Equation 2 was used to calculate the percent reduction of MO from their UV-Vis spectra:

$$\% \text{Reduction} = 100 - (A_t \times 100/A_0) \quad (2)$$

where A_0 is the absorbance at an initial time at λ_{max} at 465 nm and A_t is the reaction absorbance at time t . Our AgNPs illustrated an exceptional catalyst in the reduction of MO dye and complete 94.3% reduction in 9 min by using 10 mg of Ag nanoparticles (Figure 8b). Catalytic reduction of MO dyes in the presence of sodium borohydride follows “pseudo-first-order” reaction. The pseudo-first-order equation can be expressed as follows:

$$r = \ln C_t/C_0 = \ln A_t/A_0 = -k_{\text{app}} t \quad (3)$$

The rate constant calculated from the slope of $\ln A_t/A_0$ was 3.9×10^{-3} (Figure 8d). Thus, our AgNPs showed superior catalytic reduction as compared to the recently reported study [68].

5 Conclusion

In this work, one-step approach of low cost and simple method was used to synthesize AgNPs. The prepared AgNPs show distinct surface plasmon resonance in the UV-Vis spectroscopy. Powder XRD confirms the crystalline nature of face central cubic (fcc) AgNPs. TEM confirms the formation of small size AgNPs with an average size of 6.5 nm. XRD and TEM images confirmed fcc structure of AgNPs and were predominantly spherical in structures; while EDS analysis showed elemental silver in nanoparticles. The impact of the pH and temperature on the synthesis of AgNPs was also investigated. The optimum temperature and pH were found at 65°C and 9, respectively. The AgNPs displayed effective results against selected pathogenic strains, *P. aeruginosa* (MTCC 424), Methicillin-resistant *S. aureus* (ATCC 43300), and fungus *C. albicans* (KACC 30003). AgNPs exhibited a zone of inhibition against all microbes, 5 mm *C. albicans*, 4 mm *P. aeruginosa*, and 7 mm *S. aureus*. AgNPs illustrated an exceptional catalyst in the reduction of MO dye and completed a 94.3% reduction of 9 min. As evidenced by antibacterial and antioxidant activities, our prepared AgNPs were more successful as compared to their precursor; on the whole, the strategy offered here for the preparation of AgNPs is a cost-efficient and simple way of nanomedicine that can be used in controlling various veterinary and human infections and also for the cure of various diseases due to oxidative stress.

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