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Plant-mediated green synthesis of Ag nanoparticles using *Rauvolfia tetraphylla* (L) flower extracts: Characterization, biological activities, and screening of the catalytic activity in formylation reaction

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Antimitotic assay.

Abstract. Various plant extracts have currently been used in the bioproduction of nanoparticles with enormous applications. In this study, *Rauvolfia tetraphylla* flower extracts were employed to obtain silver nanoparticles (Ag NPs) in bioproduction. The biologically produced nanoparticles were characterized by XRD, FTIR, UV-Vis, BET, SEM, EDXA, and TEM analyses. Phytochemical screening of the *Rauvolfia tetraphylla* flower extracts indicated presence of 9 different constituents. Bioreduction of Ag NPs by phytochemicals was revealed by FTIR analysis. The elemental composition of Ag NPs was reported by spectral EDXA. The Ag NPs exhibited anti-bacterial activity against *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Klebsiella aerogenes*, and *Escherichia coli*; anti-fungal activity against *Penicillium citrinum* and *Aspergillus flavus*; and antimitotic activity. The response of amines in formic acid in the presence of an Ag NPs catalyst in dissolvable free condition provided high yielded convention for the *N*-formylation to shape the comparing formamide derivatives. *N*-fromylation had the characteristics of incite recyclability, clean strategy, environmental friendliness under milder response conditions, and straightforward work-up with brilliant yield of the coveted items.

1. Introduction

Nanotechnology is a new-found and quickly developing field with myriad applications in science and innovation. Nanotechnology can be used to produce silver nanoparticles with unique optical, electrical, and magnetic properties based on their sizes. Nanoparticles are known for biological activities suitable for incorporation into various applications including biosensors, antimicrobials, cosmetic products, materials for cryogenic superconductors, composite filaments, and electronic components [1]. Ag NPs are endowed with distinctive properties, like chemical stability, conductivity, and catalytic and antibacterial activities [2] in colloidal

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state. Silver in different forms finds various applications and as a nanoparticle, it has been used in dental medicine, wound treatment, coating of stainless steel materials, water purification, and sunscreen lotions [3]. Nano catalysis is the fastest growing field which involves the use of nanoparticles as catalysts. Metal ions and noble metals such as Pt, Au, and Ag can catalyze the decomposition of H_2O_2 to oxygen [4]. Exposure of the luminol-H_2O_2 system to colloidal Ag solution causes chemiluminescence emission as an indication of nano catalysis [5]. Silver nanoparticles have extensive applications in integrated circuits [6]. Silver has long been recognized as an antiseptic and anti-biotic due to its inhibitory effect on many microorganisms [7].

The use of plants in the synthesis of nanoparticles has several advantages such as avoiding the complicated processes of maintaining cell cultures, easy scale-up for large-scale synthesis, and cost-effectiveness. The plant extracts may act as both reducing and stabilizing agents during the bioproduction of nanoparticles [8]. The use of plant extracts for the bioproduction of nanoparticles has drawn the attention of researchers, because the method is rapid, economical, and eco-friendly and it provides a single-step technique for such nano syntheses [9]. The chemical and physical methods of producing nanoparticles require high pressure, energy, and temperature as well as toxic chemicals [10]. In recent years, the use of plants and their extracts for bioproduction of nanoparticles has become popular as the method is cost-effective and environmentally friendly [11].

Many researchers used a variety of plants for the biological synthesis of silver nanoparticles. Also, plants and their extracts have been used for bioprodction of nanoparticles. The plant extracts of *Syzygium cumini*, *Solanum tricobatum*, *Citrus sinensis*, and *Centella asiatica* [12]; *Citrus sinensis*, *Solanum tricobatum*, *Syzygium cumini*, and *Ocimum tenuiflorum* [13]; *hedysarum* plant and the *Acanthe phylum bracteatum* [14]; *Azadirachta indica* extracts [15]; *Jasminum grandiflorum* and *Cymbopogon citrullus* [16]; *Trianthema decandra* [17]; *Cressa cretica* [18]; *Jamun* [19]; *Strawberry* [20]; *Euphorbia hirta* (Euphorbiaceae) [21]; *Avacado* [22]; and leaf powder from *C. asiatica*, *C. sinensis*, *S. tricobatum*, and *S. cumini* [23] are some instances.

Rauvolfia tetraphylla (Figure 1), belonging to Apocynaceae family, has many applications in traditional medicine. Its extracts are used to treat snake poisoning and mental illness. *Rauvolfia tetraphylla* powder has also demonstrated antimicrobial properties [24].

In this study, the bioproduction of Ag NPs is attempted with flower extracts of *Rauvolfia tetraphylla*. The silver nanoparticles obtained are characterized and evaluated for their antimicrobial and antimitotic

Figure 1. *Rauvolfia tetraphylla* plant material (inset: flower).

activities. Formamides are critical intermediates in manufactured natural science and in peptide blend, they are utilized as securing bunch for amines [25]. They are likewise utilized for amalgamation of pharmaceutically significant isocyanides [26] as well as nitrogen connected heterocylces [27]. Formamides are the impetuses, discover its applications in the hydrosilylation and allylation responses of carbonyl mixes [28,29]. Presently, formamides are combined from the accompanying techniques incorporates: acidic formic anhydride, actuated formic esters, imidazole in warm DMF have been utilized [30-32]. Numerous other helpful formylation impetuses, for example, AgO, silver metal and VB1 with formic corrosive have been accounted for formic acid has been reported [33-35].

2. Experimental

2.1. Collection and preparation of flower extracts

Rauvolfia tetraphylla plant was collected from the forest of Devarayanadurga in Tumakuru district, Karnataka, India. The plant was authenticated by taxonomist Dr. Y.N. Seetharam, co-ordinator from the Department of Botany at Tumkur University, Tumkur, Karnataka, India. Flowers from the twigs of *Rauvolfia tetraphylla* were collected and washed with tap water for removing the dirt and dust particles fallowed by double-distilled water. *Rauvolfia tetraphylla* flowers (20 g) were put in 100 mL double-distilled water placed on a heating mantle at 60°C for 30 min with stirring. Afterwards, the mixture was cooled to room temperature (30°C) and filtered by Whatman filter paper no. 1. The resulting pale yellow colored flower extract was used as reducing and capping agent in bioproduction of Ag NPs.

2.2. Phytochemical analysis

The flower extracts of *Rauvolfia tetraphylla* were assessed [37] for the existance of phytochemical com-

Table 1. Phytochemical analysis of *Rauvolfia tetraphylla* (flower).

Serial no.	Phytochemicals	Result
1	Flavonoids	+++
2	Alkaloids	+++
3	Phenols	++
4	Tannins	+
5	Cardiac glycosides	+++
6	Saponins	+++
7	Anthraquinones	–
8	Amino acids	++
9	Oxalate	–
10	Phlobatannins	+++
11	Terpenoids	+++

Note: +++ = Appreciable amount; ++ = Medium presence; + = Presence in trace amount;
- = Negligible amount or completely absent

ponents such as tannin saponins, phenols, terpenoids, tannins, flavonoids, anthraquinones, amino acids, phlobatannins, oxalates, cardiac glycosides, and alkaloids through the standard procedures. The obtained results are shown in Table 1 [36,38].

2.3. Synthesis of silver nanoparticles

Rauvolfia tetraphylla flower extract (10 mL) was added to $AgNO_3$ (90 mL) solution in a conical flask and mixed thoroughly at 30°C; then, the solution was placed on a magnetic stirrer for 10 min. The mixture was set aside for 24 h for complete bio-reduction to produce nanoparticles [39].

2.4. Characterization of silver nanoparticles

The synthesized Ag nanoparticles were characterized by XRD, UV-Vis, FT-IR, BET, SEM, EDXA, and TEM analyses [40].

2.5. Antimicrobial activity of Ag NPs

The antibacterial activity of biologically synthesized Ag NPs was determined by the disc diffusion technique. The bacterial strains such as *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Klebsiella aerogenes* and *E-coli*, were cultured in NB media for 24 hours at 37°C [41,42]. One mL of each bacterial broth culture was poured over the sterile NA media. Five-mm filter paper discs impregnated with silver nanoparticles suspension (10 μg/mL) were placed on nutrient agar medium. The filter paper discs dipped in double-distilled water served as negativ0065 control. The positive control discs contained Taxim (1 μg/mL) [43]. The test discs were also prepared by dipping in flower crude extracts (20%). The filer paper discs were placed over the surface of agar plates inoculated with test organism and incubated for 24 h at 37°C. The zones of inhibitions were measured in a measuring scale [44,45].

2.6. Antifungal activity of silver nanoparticles

Antifungal activity of Ag NPs was evaluated against selected plant pathogenic fungi, viz. *Penicillium citrinum* and *Aspergillus flavus*, by Kirby-Bauer disc diffusion method [46,47]. Ag NPs with the concentration of 20 μg/disc was impregnated on paper discs. Norfloxacin was maintained as a positive control at the concentration of 20 μg/disc and double-distilled water was used as negative control [48]. Fungal spore suspension was poured on Potato Dextrose Agar (PDA) plates and paper discs were placed on the medium. The plates, containing paper discs, were incubated at 28°C for 48-72 h. The inhibition zone was measured in the measuring scale.

2.7. Antimitotic assay

Antimitotic activity was determined using *Alium cepa* (onion) bulbs. *Alium cepa* was used to assess the disturbances in the mitotic cycle and chromosomal aberrations. *Alium cepa* has advantages over other short-term tests. Among the endpoints of *A. cepa* root chromosomal aberrations, detection of chromosomal aberration has been the most commonly applied technique to detect genotoxicity/antigenotoxicity for years. The mitotic index and chromosomal abnormalities were used to evaluate genotoxicity and micro nucleus analysis was used to verify mutagenicity of different chemicals. The effect of the Ag NPs synthesized by *Rauvolfia tetraphylla* flower extracts on cells showing different stages of mitosis, i.e., interphase, metaphase, telophase, and anaphase, was considered. The antimitotic index was calculated by the following formula [49]:

$$\text{Mitotic index} = \frac{\text{Number of dividing cells}}{\text{Total number of cells}} \times 100. \quad (1)$$

2.8. Synthesis of formamide derivatives of aromatic amines

Adopting the follow-up procedure, amine (1 mmol) was added to 98% formic acid (3 mmol) and Ag NPs (2 mol%) mixture. This reaction mixture was heated to 70°C with constant stirring. The reaction progress was observed by TLC. After the end of the reaction, EtOAc was added to the reaction mixture and the Ag NPs catalyst was removed through filtration process. The obtained organic solvent was clearly washed with deionised water and saturated brine solution. Then, it was dried over anhydrous Na_2SO_4. After the removal of the solvent, a pure product was obtained and no further purification process was needed.

3. Results and Discussion

3.1. Biosynthesis of Ag NPs using Rauvolfia tetraphylla flower extracts

About 10 mL *Rauvolfia tetraphylla* flower extract was added to 90 mL $AgNO_3$ solution at room temperature.

The mixture was stirred continuously for 10 min. The pale yellow color of the mixture changed to dark brown after 24 h, which indicated the biosynthesis of silver nanoparticles (Figure 2). Silver nanoparticles were purified by repeated centrifugation at 8,000 rpm for 15 minutes using cooling centrifuge (Remi C-24). The Ag NPs obtained was dried and stored.

3.2. X-ray diffraction studies

The XRD peaks at $2\theta = 38°$, 44°, 64°, and 77° were indexed with the planes (111), (200), (220), and (311) for the face centred cubic lattice of the attained Ag (silver) as per the JCPDS (Joint Committee on Powder Diffraction Standards). Card no. 04-0783 matched the database for Ag NPs synthesized by *Rauvolfia tetraphylla* flower extracts [50]. The calculated D (average size) value of synthesized silver nanoparticles was found to be 29.2 nm as calculated by the Debye-Scherer formula (Figure 3) [51].

3.3. FT-IR analysis

Strong infrared bands were observed at 3289, 2916, 1597, 1387, 1062, and 536 cm^{-1}. The strong broad band, which appeared at 3289 cm^{-1} alcohol O-H stretch; the bands at 2916 cm^{-1} Amine N-H stretch, 1597 cm^{-1} Cyclic alkene C=C, 1387 cm^{-1} Alkane C-H, and 1062 cm^{-1} Primary alcohol C-O; and the low band at 536 cm^{-1} corresponded to the halogen compound C-Br stretch (Figure 4).

3.4. BET studies

Pore size distributions of the synthesized Ag NPs were studied by N_2 absorption-desorption isotherms measured using static volumetric absorption analyzer. The results are presented in Figure 5. Perforated surface area of Ag NPs showed superior surface for catalytic properties as it enabled adsorption/desorption of reactant molecules [52]. The pore size distribution curves and specific surface area of the synthesized Ag NPs were obtained by BET gas sorption instrument. In Figure 5, it is obvious that the Ag NPs has mesoporous

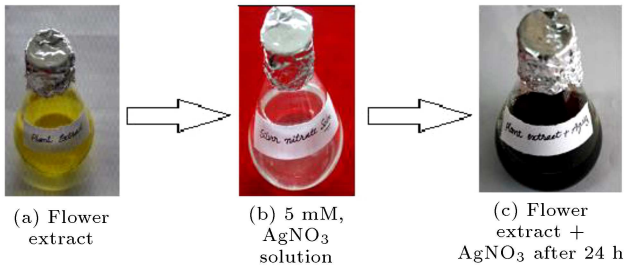


Figure 2. Bioproduction of Ag NPs from *Rauvolfia tetraphylla* flower extracts.

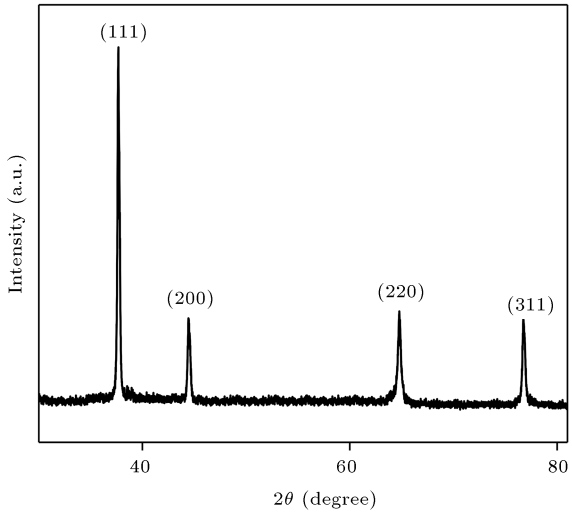


Figure 3. XRD pattern of Ag NPs from *Rauvolfia tetraphylla* flower extracts.

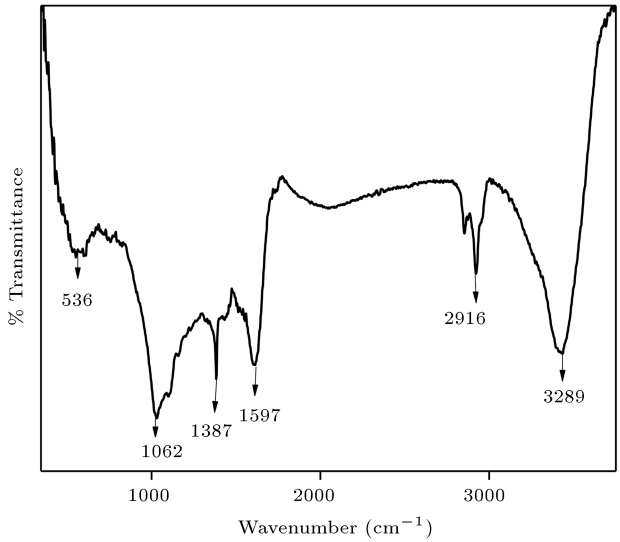


Figure 4. FT-IR spectrum of Ag NPs from *Rauvolfia tetraphylla* flower extracts.

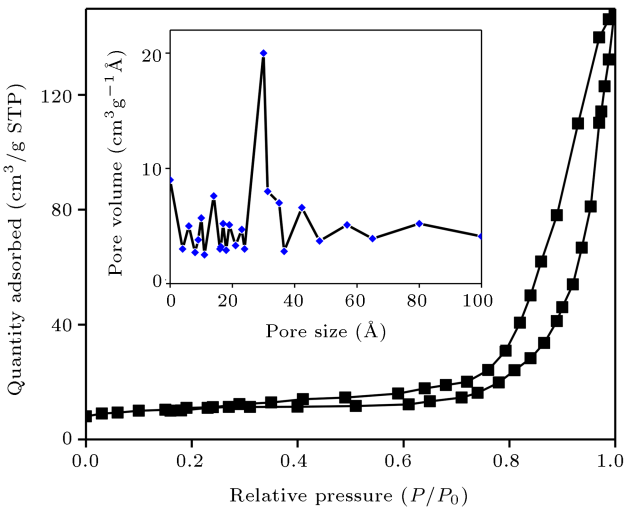


Figure 5. N_2 adsorption/desorption isotherms (inset: pore size distribution curves of Ag NPs).

nature at low pressure regions ($P/P_0 < 0.7$) with the typical IV adsorption type of H3 hysteresis loop [53]. After increase in the pressure beyond 0.7 (P/P_0), the isotherm rises suddenly and forms a large loop. Figure 5 shows the pore size distribution curves of the Ag NPs, in which it can be seen that the pore size probability for Ag NPs is about 20 nm with the surface area of 26.31 m^2/g. With reduction in the size of Ag, surface area increases, which enhances the catalytic properties of Ag NPs.

3.5. UV-Vis-spectroscopy analysis

UV-vis spectrum of silver nanoparticles biosynthesized by *Rauvolfia tetraphylla* flower extracts was 460 nm, which was peak broadening with an increase in absorbance due to increase in the number of Ag NPs formed as a result of reduction in Ag^+ ions present in the aqueous $AgNO_3$ solution (Figure 6) [54].

3.6. Scanning electron microscopy analysis

Scanning Electron Microscope (SEM) images of Ag NPs biosynthesized by the flower extracts *Rauvolfia tetraphylla* (Figure 7) showed separate as well as agglomerate silver nanoparticles [44]. The shape of the particles was spherical in morphology and particles were distributed uniformly.

3.7. Energy-dispersive spectroscopy analysis

Energy-Dispersive X-ray Analysis (EDXA) describes the elemental analysis of the mentioned silver nanoparticles. The spectrum of Ag NPs was measured at the energy of 3 keV for silver and some weaker peaks belonging to carbon, sodium, nitrogen, and oxygen were found (Figure 8) [55].

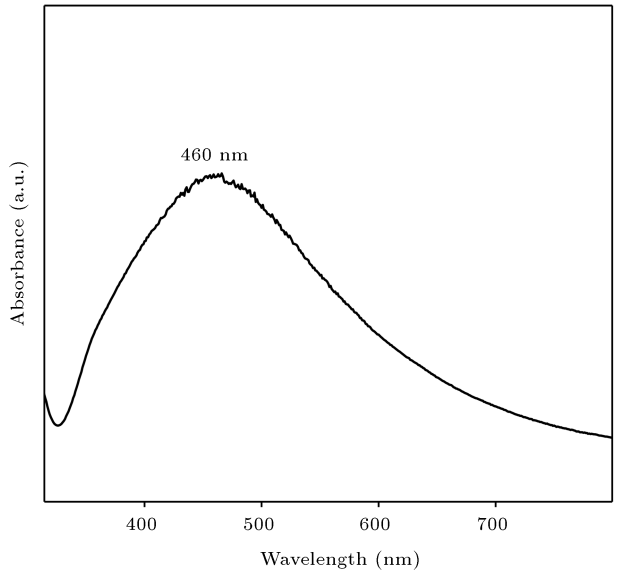


Figure 6. UV-vis spectrum of Ag NPs from *Rauvolfia tetraphylla* flower extracts.

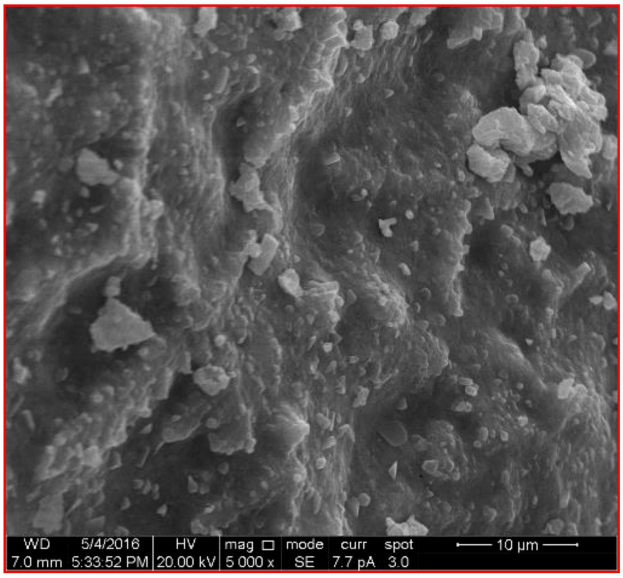


Figure 7. SEM micrograph of Ag NPs from *Rauvolfia tetraphylla* flower extracts.

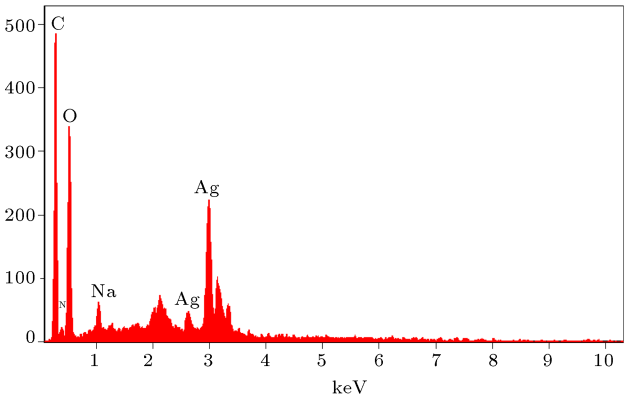


Figure 8. EDXA analysis of Ag NPs from *Rauvolfia tetraphylla* flower extracts.

3.8. Transmission electron microscopy analysis

Figure 9(a) shows the Transmission Electron Microscopy (TEM) image of the synthesized Ag NPs. It is clear that the synthesized materials are in the nano range with the spherical shaped structures. These particles have high surface area and prepare the site for the reaction on the surface of the catalyst. Figure 9(b) shows the particle size distribution of Ag NPs. It clearly reveals that the synthesized materials are maximum in the range of 20 to 25 nm size. This nano-size Ag NPs is proper to give good yield in lower time [55,56].

3.9. Antibacterial assay

The synthesized Ag NPs from flower extracts of *Rauvolfia tetraphylla* had significant antibacterial activity against *E-coli*, *Pseudomonas aeruginosa*, *Staphylococ-*

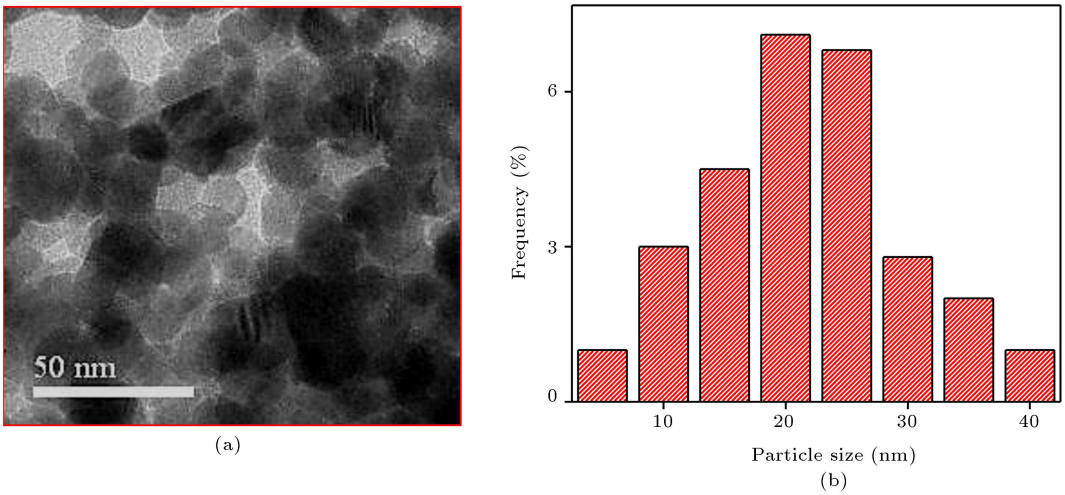


Figure 9. (a) TEM image and (b) graph of particle size distribution of Ag NPs.

Table 2. Antibacterial zone formation.

Zone of inhibition (mm)					
Serial no.	**Strain**	**Control[a]**	**Ag NPs[b]**	**Standard[c]**	**Flower extracts[d]**
1	*E-coli*	—	6.1	8.3	—
2	*Pseudomonas aeruginosa*	—	6.3	8.2	—
3	*Klebsiella aerogenes*	—	5.6	7.9	—
4	*Staphylococcus aureus*	—	5.4	7.6	—

[a]Control: Double-distilled water; [b]Ag NPs: Silver nanoparticles; [c]Standard: Taxim;
[d]Flower extracts: *Rauvolfia tetraphylla*.

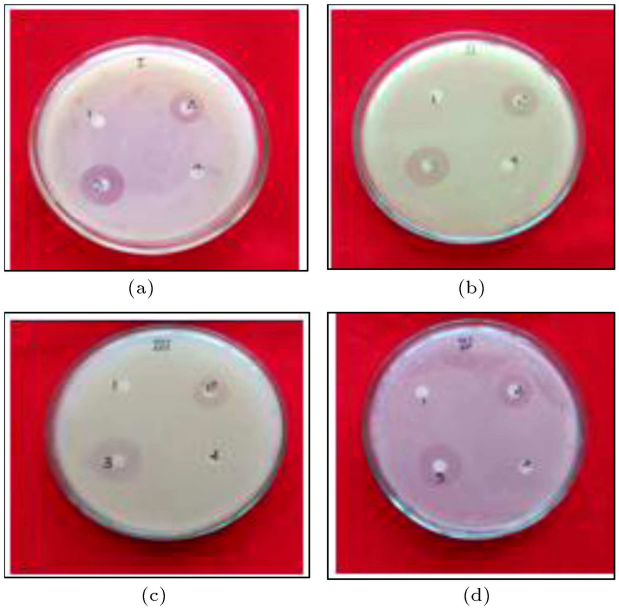


Figure 10. Zone of inhibition against (a) *E. coli*, (b) *P. aeruginosa*, (c) *K. aerogenes*, and (d) *S. aureus* in the presence of Ag NPs.

cus aureus, and *Klebsiella aerogenes* (Figure 10, Table 2) [51].

3.10. Antifungal activity

Antifungal study indicated that the Ag NPs from

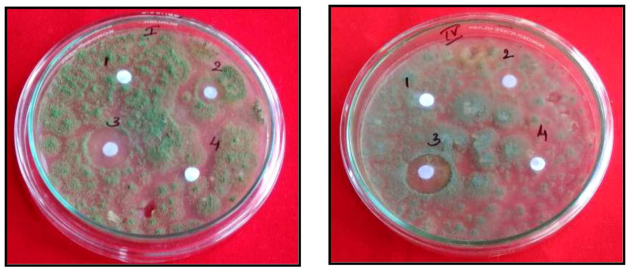


Figure 11. Antifungal activity of (a) *A. flavus* and (b) *P. citrinum* in the presence of Ag NPs.

Rauvolfia tetraphylla flower extracts had a broader zone of inhibition than the standard Norfloxacin antibiotic against *Penicillium citrinum* and *Aspergillus flavus* (Figure 11, Table 3) [57].

3.11. Antimitotic activity of Ag NPs at Allium cepa root tips

Antimitotic activity assessment was carried out using *Allium cepa* with control grown in tap water and Quercetin used as standard drug (Table 4, Figures 12(a), 12(b), and 12(c)) [58].

3.12. Formylation of aromatic amines in the presence of Ag NPs catalyst

Ag NPs is a vital nano metal oxide with an extensive variety of uses. A blend of HCO_2H and Ag NPs was

Table 3. Antifungal zone formation.

Zone of Inhibition (mm)					
Serial no.	**Pathogenic fungi**	**Control[a]**	**Ag NPs[b]**	**Standard[c]**	**Flower extracts[d]**
1	*Aspergillus flavus*	—	3.8	5.4	—
2	*Penicillium citrinum*	—	3.9	5.7	—

[a]Control: Double-distilled water; [b]Ag NPs: Silver nanoparticles; [c]Standard: Norfloxacin;
[d]Flower extracts: *Rauvolfia tetraphylla*.

Table 4. Antimitotic activity of Ag NPs.

Serial no.	Sample	Concentration	Mitotic index
1	Control	—	93.7
2	Ag NPs	10 mg/mL	21.5
3	Ag NPs	5 mg/mL	28.1
4	Quercetin (standard)	1mg/mL	14.9

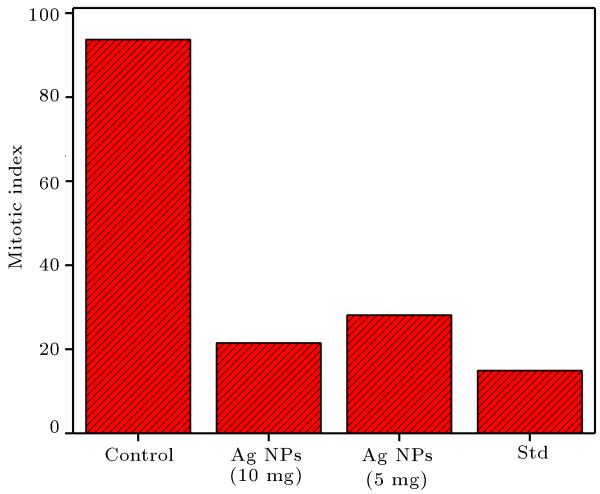
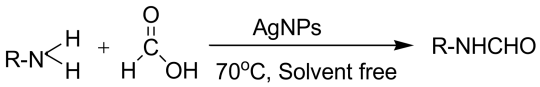


Figure 12a. Graphical representation of antimitotic activity of Ag NPs.

added to an amine and then, the reaction blend was refluxed at 70°C until fruition of the reaction (the advance of the response was judged by TLC). The reaction blend was brought to ambient temperature after culmination. By this system, a few formamides (6) were set up from the subsidiaries of fragrant amines. Then, the reaction blend was weakened by ethyl acetic acid derivation and Ag NPs was evacuated by filtration. The natural dissolvable was then washed with water and soaked arrangement of saline solution and dried over anhydrous Na_2SO_4. The solvent was evacuated with lower weight and the pure item was obtained (Table 5). It was additionally cleansed by recrystallization utilizing the reasonable solvent, diethyl ether. The integrated mixes were affirmed by ^{13}C NMR and ^{1}H NMR studies [59].



Mechanism of reaction on the surface of catalyst:

The possible mechanism for the activation of formic acid on the surface of Ag NPs catalyst during the formation of *N*-formamides is depicted in Schemes 1 and 2 [59].

Spectral data of the synthesized compounds

1. *N*-(3-chlorophenyl) formamide (**1**): ^{1}H NMR

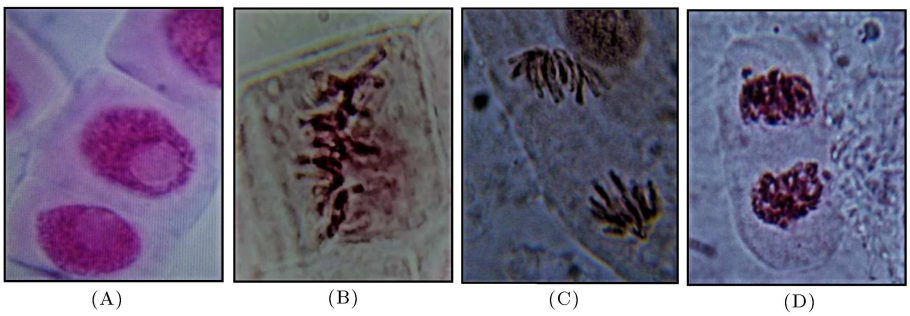


Figure 12b. Normal mitotic phases of *Allium cepa*: (A) interphase, (B) metaphase, (C) anaphase, and (D) telophase.

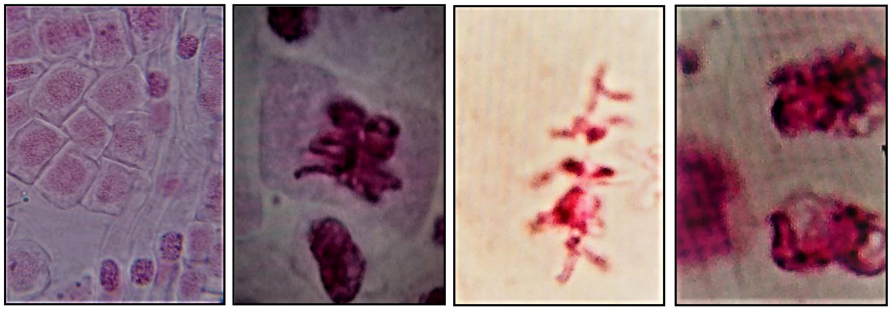


Figure 12c. Mitotic abnormalities of *Allium cepa*: (A) mega cells and cell shrinkage, (B) chromosomal clumping and cell shrinkage at metaphase, (C) vagrant chromosome at metaphase, and (D) chromosomal clumping at telophase.

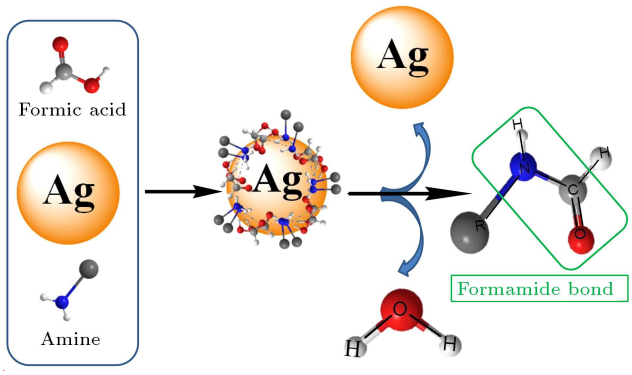
Table 5. *N*-formylation of amines with formic acid using Ag NPs under solvent-free condition.

Serial no.	R-NH_2	R-NHCHO	Yield (%)	Time (min)
1	3-chlorobenzenamine	*N*-(3-chlorophenyl)formamide	89	28
2	Aniline	*N*-phenylformamide	93	13
3	4-bromobenzenamine	*N*-(4-bromophenyl)formamide	88	19
4	3-aminophenol	*N*-(3-hydroxyphenyl)formamide	85	57
5	3-nitrobenzenamine	*N*-(3-nitrophenyl)formamide	87	60
6	4-aminobenzoic acid	*N*-formamidobenzoic acid	85	55

(250 MHz, $CDCl_3$) δ 8.95 (br, 1H), 8.57 (d, 1H, J = 11.17 Hz), 8.23 (s, 1H), 8.01 (br, 1H), 7.02-7.55 (m, 4H); ^{13}C NMR (62.9 MHz, $CDCl_3$) δ 118.7-132.3, 161.1, 164.1 ppm. HRMS: Calcd for $C_7H_6ClNNaO$ m/z: 178.00 $[M+Na]^+$, found 178.04;

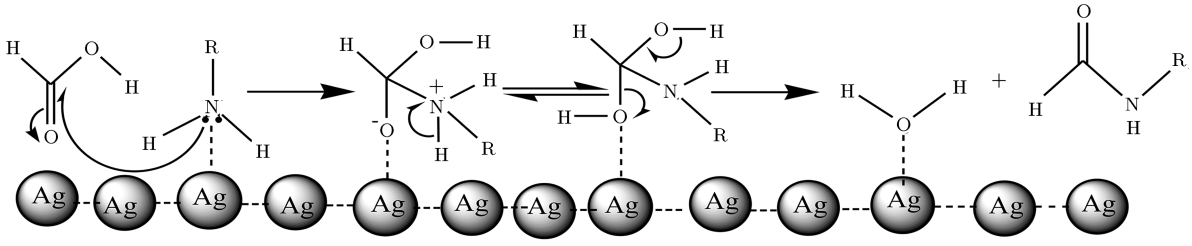
2. *N*-phenyl formamide (**2**): ^{1}H NMR (250 MHz, $CDCl_3$) δ 9.32 (brs, 1H), 8.7 (brs, 1H), 8.63 (d, 1H, J = 11.26 Hz), 8.11 (s, 1H), 6.97-7.57 (m, 5H); ^{13}C NMR (62.9 MHz, $CDCl_3$) δ 117.3-131.0, 137.0, 161.8, 164.7 ppm. HRMS: Calcd for C_7H_7NNaO m/z: 144.04 $[M+Na]^+$, found 144.07;

3. *N*-(4-bromophenyl) formamide (**3**): ^{1}H NMR (250 MHz, $CDCl_3$) δ 9.23 (brs, 1H, *trans*), 8.44 (d, 1H, *J=11.33* Hz), 8.23 (s, 1H), 8.13 (brs, 1H), 7.37-7.43 (m, 2H), 6.91-6.95 (m, 2H); ^{13}C NMR (62.9 MHz, $CDCl_3$) δ 115.3, 115.7, 116.1, 116.4, 120.7, 121.0, 132.5, 158.2, 163.1 ppm. HRMS: Calcd for $C_7H_6BrNNaO$ m/z: 221.95 $[M+Na]^+$, found 221.98;



Scheme 2. Graphical representation of the formation of formamide bond.

4. *N*-(3-hydroxyphenyl) formamide (**4**): ^{1}H NMR (250 MHz, DMSO) δ 10.01 (brs, 1H, *trans*), 9.95 (s,



R=3-chlorobenzenamine, Aniline, 4-bromobenzenamine, 3-aminophenol, 3-nitrobenzenamie, 4-aminobenzoic acid

Scheme 1. Plausible mechanism for the *N*-formylation of amines on Ag NPs surface.

1H), 9.47 (s, 1H), 8.65 (d, 1H, $J = 12.0$ Hz), 8.15 (s, 1H), 6.01-7.14 (m, 4H); ^{13}C NMR (62.9 MHz, $CDCl_3$) δ 104.6, 108.2, 110.7, 129.4, 138.5, 159.5, 162.3 ppm. HRMS: Calcd for $C_7H_7NNaO_2$ m/z: 160.04 $[M+Na]^+$, found 160.08;

5. *N*-(3-nitrophenyl) formamide (**5**): ^{1}H NMR (250 MHz, DMSO) δ 10.65 (s, 2H,), 8.91 (d, 1H, $J = 8.5$ Hz), 8.55 (s, 1H), 8.33 (s, 1H), 7.55-7.91 (m, 3H); ^{13}C NMR (62.7 MHz, DMSO) δ 111.5, 113.5, 122.0, 124.7, 130.2, 136.1, 147.6, 160.0, 162.4 ppm. HRMS: Calcd for $C_7H_6N_2NaO_2$ m/z: 189.03 $[M+Na]^+$, found 189.06;
6. *N*-formamidobenzoic acid (**6**): ^{1}H NMR (250 MHz, DMSO) δ 12.75 (brs, 1H), 10.63(s, 1H), 8.32 (s, 1H), 7.83 (d, 2H, $J = 6.21$ Hz), 7.68 (d, 2H, $J = 8.63$ Hz); ^{13}CNMR (62.5 MHz, $CDCl_3$) δ 116.5, 118.92, 126.06, 130.08, 142.52, 160.50, 167.22 ppm. HRMS: Calcd for $C_8H_7NNaO_3$ m/z: 188.03 $[M+Na]^+$, found 188.08.

3.13. Hot filtration method

Hot filtration is generally used for the recovery of catalyst in hot conditions. In this method, the catalyst is heated and washed with solvent to remove the impurities. In a final set of experiments, we further assessed the stability and reusability of the Ag NPs catalyst in the formylation reaction. It was crucial to confirm activity of the catalyst for recycling of Ag NPs. Figure 13 shows that there is no considerably lower yield in each cycle by reusing the catalyst. We observed nearly 10% loss of yield in 5 cycles of reaction [60].

3.14. Recyclability of the catalyst

The reusability of Ag NPs used as a catalyst was tested under identical reaction circumstances. The Ag NPs catalyst was effectively recovered from the reaction mixture by hot filtration. The obtained product was carefully washed by deionized water followed by ethyl acetate. Then, it was dried for 2 to 3 h under the vacuum condition. During each reaction, the Ag NPs catalyst was recovered from the reaction mixture, indicating a loss of around 13% after 5 times being reused. This loss seemed reasonable, as a result of washing and filtering of the heterogeneous catalyst, and was in coordination with the obtained yield after 5 cycles. On the other hand, the loss of catalyst is itself a reason for getting lower yield after 5 cycles (Figure 14) [61].

Catalytic properties of Ag derivatives synthesized by the green method were compared with those by the chemical methods. Only a finite number of publications are available on the Ag NPs catalyzed by organic reactions. To the best of our knowledge and based on the literature survey, this is the first manuscript reporting green synthesized Ag NPs as a catalyst for the formamide reaction. However, some other NPs like MgO and ZnO have been used for the formamide reactions. Therefore, we compared our results with different organic reactions catalyzed by Ag derivatives.

Javid Safari et al. synthesized AgI NPs with spherical shape and the size of around 10 to 20 nm by precipitation method. The NPs were used as catalyst for the A^3 coupling of benzofuran reactions. The time required for completion of the reaction was 4.5 h with the yield of around 52%, which seems very low efficiency in comparison with the present method [62]. Yuqing Zhou et al. synthesized Ag_2O NPs with spherical shape and the size of 18-20 nm by reflux method. The method was employed for the A^3 coupling reactions with the expense of 12 h and the yield of 29% [63].

Kushal D Bhatte et al. synthesized Ag NPs with the obtained size of 40 nm and spherical shape by chemical reduction method. Enamiones and enaminoesters were synthesized with 70% yield in 12 hours [64]. In the same way, Bharat A Makwana et al. synthesized

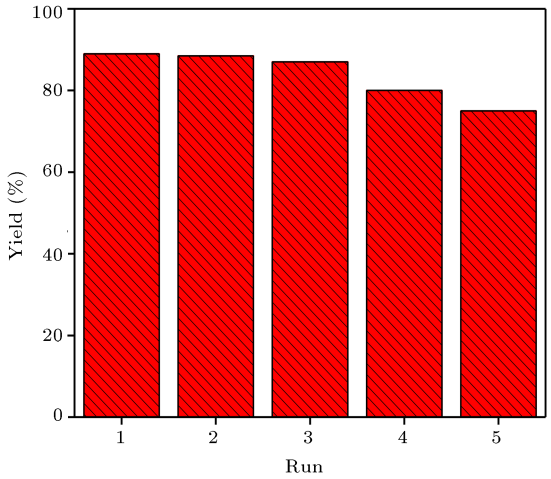


Figure 13. Recyclability of the Ag NPs catalyst by hot filtration method.

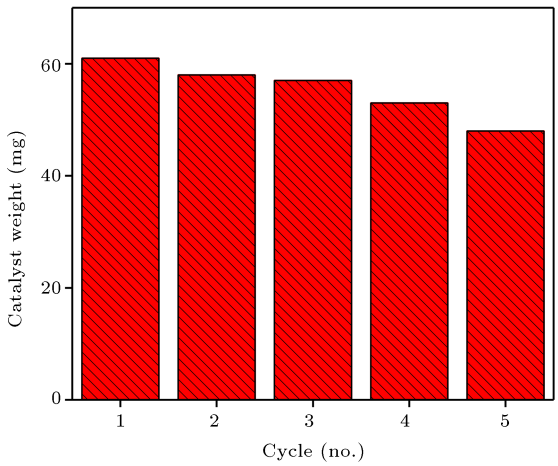


Figure 14. Recovery of Ag NPs catalyst after each reaction.

Table 6. Comparison of catalytic properties of Ag derivatives synthesized by different methods for different organic reactions.

Serial no.	Method of synthesis for Ag derivatives	Size and shape of Ag NPs	Name of the reaction carried out	Time for completion of reaction	Obtained yield (%)	Reference
1	Precipitation method (SDS surfactant)	Spherical shape, particle size of 10-20 nm	A^3 -coupling reaction	4.5 h	52%	[61]
2	Reflux method	Spherical shape, particle size of 18-20 nm	A^3 -coupling reaction	12 h	29%	[62]
3	Chemical reduction method	Spherical shape, particle size of 40 nm	Synthesis of enaminones and enamino esters using silver nanoparticles	6 h	70%	[63]
4	Chemical reduction method	Spherical shape, particle size of 3-7 nm	Synthetic route of octamethoxy resorcinarene tetrahydrazide	16 h	92%	[64]
5	Green synthesis method using *Rauvolfia tetraphylla*	Spherical shape, particle size of 20-25 nm	Formylation reaction	13 min	93%	[Present work]

Ag NPs with spherical shape and the size of 3 to 7 nm by chemical reduction method. They used Ag NPs as a catalyst in the octamethoxy resorcin arene tetrahydrazide reaction and obtained a yield of around 92% in 16 h [65]. In comparison with all the above-mentioned reactions, the present method dominates their time and yield, as tabulated in Table 6.

4. Conclusion

Silver nanoparticles were synthesized by the *Rauvolfia tetraphylla* flower extracts at room temperature. The Ag NPs have good antimicrobial activity against *Klebsiella aerogenes*, *Staphylococcus aureus*, *E-coli*, and *Pseudomonas aeruginosa* and antifungal activity against *Penicillium citrinum* and *Aspergillus flavus*. Also, the synthesized particles had consistent results in antimitotic assays. In the presence of Ag nano catalyst, incredible yield of formamide derivatives was achieved. It can be concluded that Ag NPs may find large applications to catalytic activity; drug delivery process; and anticancer, antibacterial, and antifungal practices in the medical field.

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