

Biosynthesis of Silver Nanoparticles Using *Cinnamomum tamala* Leaf Extract and Revealing Its Antioxidant Activity

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ABSTRACT

Silver nanoparticles (AgNPs) possess distinctive properties. This study examines the biosynthesis of AgNPs with antioxidant properties using *Cinnamomum tamala* leaves through green chemistry. Silver nitrate (AgNO₃) was converted to AgNPs mediated *Cinnamomum tamala* leaf extract method, providing a cost-effective and eco-friendly alternative to conventional methods. X-ray diffraction (XRD) confirmed the crystalline structure of AgNPs with an average size of 21.92 nm. Scanning electron microscopy (SEM) and Transmission electron microscopy (TEM) analyzed the nanoparticles' morphology and size distribution. The AgNPs were found to range from 30 to 60 nm, establishing their sub-100 nm size. The presence of AgNPs was verified by a peak at 3-3.5 keV in the EDX spectrum. Ultraviolet-visible (UV-Vis) spectroscopy and Fourier-transform infrared spectroscopy (FTIR) were employed to examine the reduction of AgNPs and stable phytochemicals. Various radical scavenging activities were evaluated to determine antioxidant capacity. The antioxidant activity was assessed through DPPH, superoxide anion, hydroxyl, hydrogen peroxide, and nitric oxide scavenging assays. AgNPs exhibited substantial antioxidant activity with IC₅₀ values of 58.88µg/mL, 57.18µg/mL, 53.58µg/mL, 61.50µg/mL, and 56.02µg/mL respectively, indicating effective free radical scavenging. The results highlight the significant antioxidant properties of both the leaf extract and AgNPs. Synthesized AgNPs exhibited enhanced characteristics compared to the leaf extract. AgNPs from *Cinnamomum tamala* leaf extract show considerable potential as antioxidants for nutraceutical and pharmaceutical applications against oxidative stress-related diseases.

Keywords: Silver nanoparticles, *Cinnamomum tamala*, characterization, antioxidant activity, oxidative stress.

1. INTRODUCTION

The recent tremendous growth in technology in nanoscience have advanced the study of nanoparticles and the application of plants in the green biosynthesis of nanoparticles and have been the focus of an explosion of scientific research [1]. It is widely accepted that nanoparticles (NPs) are a vital element of nanotechnology. Silver nanoparticles (AgNPs) synthesis have attracted an enormous amount of curiosity because of their potential application in various domains, including biomedical applications, clean water technology, biological labelling, information storage, biological activity, optoelectronics and catalysis [2]. Organic materials such as extracts from plants and animals, elements like silver (Ag), gold (Au), platinum (Pt), nickel (Ni), zinc (Zn), copper (Cu) and compounds like titanium dioxide (TiO₂), ferrous oxide (FeO) and ferric oxide (Fe₂O₃) are utilized to synthesize nanoparticles. The size, shape, composition, crystallinity and structure of the NPs are the main factors that influence their applicability [3]. Recent research has shown that biosynthesised AgNPs made from plant extracts exhibit a wide range of biological functions including antioxidant properties [4], antibacterial [5], anti-rheumatoid [6], anti-inflammatory [7] and wound healing effects properties [8]. Plants with significant pharmacological characteristics are of great interest and have the potential to be used in the synthesis of green AgNPs [9]. Due to their eco-friendliness, biological ways of producing AgNPs employing enzymes, fungi, microbes and plant extracts have been suggested as an alternative to

chemical and physical approaches. Because it lowers oxidative stress and inflammation, the synthesis of NPs using plants or plant extracts is far more accessible than other methods [10]. In a tropical nation country like India which is endowed with a wealth of natural resources, paves an ideal platform for producing nanoparticles through plants, since they are harmless to the environment and do not contain any toxic substances. Many different types of biomolecules found in plants have distinct functional groups that aid in the reduction of silver ions to AgNPs [11]. The present investigation employed the leaf extract of *Cinnamomum tamala*, commonly referred to as Indian bay leaf (Lauraceae family), to synthesize AgNPs. Indian cuisine often uses *Cinnamomum tamala* as a spice because of its flavour and aroma, which comes from an essential oil in the leaves [12]. The active phytochemical components of *Cinnamomum tamala* leaf extract such as polyphenols, flavonoids, terpenoids, steroids, alkaloids, tannins and other secondary metabolites exhibit vast biological properties such as anti-inflammatory, antioxidant and anti-arthritis effect against Rheumatoid arthritis [13,14]. This plant is widely available in India and because of its healing and alexiteric qualities, the leaves were suggested in ancient medical systems to treat a number of illnesses including piles, heart problems, scabies, lack of taste and other conditions [15]. The leaves have also been used medicinally to treat rheumatism, nausea, vomiting, diarrhea and colic problems [16]. In the present research, a hydroethanolic leaf extract of *Cinnamomum tamala* was used in an attempt to synthesize silver nanoparticles. Further, the biological activities such as free radical scavenging properties were also evaluated for *in-vitro* studies.

2. MATERIALS AND METHODS

Biosynthesis of silver nanoparticles (AgNPs)

Cinnamomum tamala leaves were collected from Thrikkakara North, Kerala, India and was identified authenticated by a botanist from the Department of Botany at “The Rapinat Herbarium and Centre for Molecular Systematics”, Tiruchirappalli, Tamilnadu. The remaining reagents and chemicals utilized in the study were of analytical grade. For the hydroethanolic leaf extract, the fresh leaves were cleaned with distilled water, sun-dried for 20 to 30 days, and ground into fine powder. About 10 g of leaf powder were subjected to cold extraction following the maceration method [17] using hydroethanolic solution [Ethanol and water (70:30)] for 24 h with intermittent shaking. The extract was further filtered through Whatman No. 1 filter paper and the filtrate was stored at 4°C for further analysis. In the biosynthesis of AgNPs, 5 mL of *Cinnamomum tamala* leaf extract was mixed with 45 mL of freshly made 1mM silver nitrate aqueous solution (AgNO_3) in a 250 mL Erlenmeyer flask and continuously stirred for 5 hours at room temperature in the dark with a rotation speed of 200 rpm. A control setup without the leaf extract was also maintained in parallel. After incubation, the leaf extract solution colour changed from pale yellow to brown, while the silver nitrate solution without the leaf extract remained colour unchanged [18]. The mixture was then incubated for 1 h and the AgNPs were isolated through centrifugation at 15,000 rpm for 1 h. The isolated AgNPs were further washed with deionized water, dried and stored in a refrigerator for further characterization studies [19].

Characterization techniques

An XPERT-PRO X-ray diffractometer that produced Cu K α radiation (at 1.544 Å of angular resolution) operated at a voltage of 40 kV with a 30 mA current was used to determine the crystalline structure of synthesized AgNPs. The Debye-Scherrer formula has been used to determine the average particle size using the equation $D = k\lambda/\beta \cos \theta$, where D is the particle diameter size. 'k' is a dimensionless shape factor (approximately 0.9), λ is the X-ray wavelength, β is the line broadening full width half the maximum intensity (FWHM) in radians and ' θ ' is the diffraction angle. The morphology of the samples and elemental composition were analysed using ZEISS SEM (Germany) with energy dispersive x-ray spectroscopy (EDS) instrument with a magnification (Max) of 300,000X and a resolving power (Max) of 2.3 nm. Further, morphology of the nanoparticles, were examined using a transmission electron microscope (TEM; Philips model CM200) with a 2,000,000X magnification and ultra-high resolution of 0.2 nm, the instrument was run at an accelerating voltage of 200 kV. The TEM has a grid size of 3 mm diameter, which was prepared by placing 5 μ L of the silver nanoparticle solutions on carbon-coated copper grids, drying under a mercury lamp and then examined. The UV-Vis Perkin Elmer photometer operated at room temperature with a 1nm resolution between 200-1000 nm scales was used to measure the optical properties of the biosynthesized AgNPs. The infrared spectra of the AgNPs were measured and recorded using the PerkinElmer FT-IR 1600 spectrophotometer (USA) in the wavelength ranging from 400–4000 cm^{-1} at room temperature [20,21].

Antioxidant Assays

DPPH radical scavenging activity

The DPPH (1, 1-diphenyl-2-picrylhydrazyl) radical scavenging assay is commonly employed to evaluate the antioxidant potential of the relevant samples and ascorbic acid (as a standard). The scavenging activity was determined by assembling the ability of the samples to donate hydrogen atoms or electrons, and the IC_{50} values were calculated. The DPPH radical scavenging assay was studied through discolouration technique. Upon the addition of an antioxidant to the oxidized form of DPPH, which is violet in colour, undergoes reduction thereby leading to a colour shift from violet to yellow. To test various concentrated solutions (20–100 $\mu\text{g/mL}$), 2 mL of 0.1 mM DPPH (standard solution) was added to 1 mL of synthesised AgNPs and *Cinnamomum tamala* leaf extract diluted in methanol. Additionally, ascorbic acid standard solutions were also included for comparison. After mixing the solutions were allowed to incubate for 30 minutes in the dark at room temperature. After

incubation period, the absorbance (A) of each solution was measured using UV-visible spectrophotometer at 517 nm to evaluate the radical scavenging activity. The percentage inhibition (I%) was calculated using the equation:

$$\text{Percentage Inhibition (I\%)} = ((AC - AS) / AC \times 100)$$

Where, AC is the = absorbance of the control and AS = is the absorbance of the sample [22].

Superoxide anion scavenging activity

Superoxide anion ($O_2^{\cdot-}$) are relatively less reactive free radical that is generated in vivo and can contribute to the formation of more reactive species when subjected to oxidative stress [23]. The reaction mixture consisted of 50 μ L of 16 mM Tris-HCl buffer (pH 8.0), 50 μ L of 0.3 mM nitroblue tetrazolium (NBT), 50 μ L of 0.936 mM NADH (β -nicotinamide adenine dinucleotide, reduced disodium salt hydrate) and various concentrations (20–100 μ g/mL) of AgNPs and *Cinnamomum tamala* leaf extract were used to evaluate the scavenging property of superoxide anions. The reaction was initiated by adding 50 μ L of 0.12 mM PMS (phenazine methosulfate) solution to the above reaction mixture. The superoxide anion is formed when NBT is reduced by the dissolved oxygen resulting from the PMS/NADH coupling process. After five minutes of incubation at 25 °C, the absorbance at 560 nm, was measured with ascorbic acid as a reference. The percentage of inhibition was calculated as described above in the Equation [24].

Hydroxyl radical scavenging activity

The hydroxyl radicals, which have a very short half-life is the most reactive free radicals and pose significant damage to cellular components. These radicals can damage almost all biomolecules and are particularly involved in lipid peroxidation, where the hydrogen atoms are being removed from unsaturated fatty acids, contributing to cell damage [25]. Various concentrations (20–100 μ g/mL) of AgNPs and *Cinnamomum tamala* leaf were mixed with 0.45 mL of 200 mM sodium phosphate buffer (pH-7), 0.150 mL of 10 mM H_2O_2 , 0.150 mL of 10 mM 2-deoxyribose, 0.150 mL of 10 mM $FeSO_4$ -EDTA and 0.525 mL of deionized water. The mixture was incubated at 37°C for 4 hours. After incubation, 0.750 mL of 2.8% trichloroacetic acid and 0.750 mL of 1% thiobarbituric acid (TBA in 50 mM NaOH solution) were added to stop the reaction. Further, the solution was heated in a boiling water bath for 10 minutes and the absorbance was measured at 520 nm using UV-Vis spectrophotometer. The percentage of inhibition was then calculated using the above mentioned Equation [26].

Hydrogen peroxide scavenging activity

Hydrogen peroxide, is a type of reactive oxygen species (ROS), which can accumulate within living cells, leading to the production of other harmful ROS such as hydroxyl and peroxide radicals. These species formed can severely damage the cell membranes [27]. A mixture of 0.3 mL of 50 mM phosphate buffer (pH 7.4), 0.6 mL of 2 mM hydrogen peroxide solution in phosphate buffer and various concentrations (20–100 μ g/mL) of AgNPs and *Cinnamomum tamala* leaf extract in 50 mM phosphate buffer (pH 7.4) was vortexed. After 10 minutes, the absorbance was measured at 230 nm using UV-Vis spectrophotometer. The percentage of inhibition was calculated again using as described above in Equation [28].

Nitric oxide scavenging activity

Nitric oxide (NO) is a crucial bioregulatory molecule involved in various physiological processes, including immune response, neurological function and cardiovascular health [29]. Nitric oxide (NO) radicals were generated from sodium nitroprusside. A mixture of 1 mL of 10 mM sodium nitroprusside, 1.5 mL of 0.2 M phosphate-buffered saline (pH 7.4) and various concentrations (20–100 μ g/mL) of AgNPs and *Cinnamomum tamala* leaf extract was incubated at 25°C for 150 minutes. Following incubation, 1 mL of the reaction mixture was treated with 1 mL of Griess reagent (1% sulphanilamide, 2% H_3PO_4 and 0.1% naphthyl ethylene diamine dihydrochloride). The absorbance was measured at 546 nm and the percentage of inhibition was calculated using the equation above [30]. Additionally, the antioxidant capacity was quantified by determining the IC_{50} value, which represents the concentration required to inhibit 50% of radical activity, calculated by comparing with a standard ascorbic acid linear plot.

3. RESULTS AND DISCUSSION

Material characterization

On addition of silver nitrate solution, the solution containing the colourless leaf extracts of *Cinnamomum tamala* turned dark brown in a few minutes (Fig. 1), confirms the reduction of $AgNO_3$ and the successful formation of *Cinnamomum tamala*-mediated AgNPs [31].

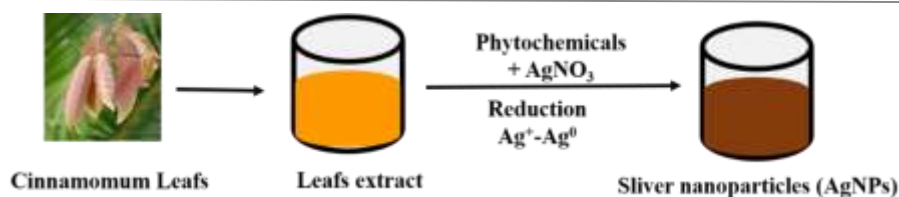


Fig.1. Leaf extract when combined with AgNO₃ changes into dark brown colour indicating the formation of silver nanoparticles.

Silver nanoparticles (AgNPs) were synthesized through an eco-friendly approach using silver nitrate (AgNO₃) solution and *Cinnamomum tamala* leaf extracts (as both capping and reducing agent) was characterized by various techniques. X-ray Diffraction (XRD) analysis was performed to characterize the synthesized AgNPs and to evaluate their purity and crystalline structure. The XRD pattern of the AgNPs displayed five distinct diffraction peaks at 38.08, 43.92, 56.23, 65.13 and 75.54 2θ values corresponding to the planes (111), (200), (211), (220) and (311) representing Bragg's reflections of metallic silver [32]. This results clearly indicates the formation of crystalline cubic face-centred structure, as identified in the standard powder diffraction card of JCPDS, silver file No. 04-0783 (**Fig. 2(a)**). The average crystallite size was determined using Debye-Scherrer formula, $D = 0.9 \lambda / \beta \cos \theta$, where 'λ' is the wavelength of X-ray (0.1541 nm), 'β' is the FWHM (Full Width at Half Maximum), 'θ' is the diffraction angle and 'D' is the particle diameter size [33]. The average crystalline size of AgNPs was found to be 21.92 nm.

The morphology of the synthesised AgNPs was analyzed by Scanning Electron Microscopy (SEM). The SEM images showed the dispersion of agglomerated clusters spread across the surface, with a significant amount of random unoccupied space [34]. The structure revealed the crystalline nature of the polydispersed spherical silver nanoparticles, with sizes ranging from 18 to 96 nm. Only a small fraction of the gathered nanoparticles was found to be polydispersed and spherical under the SEM. The average mean particle size was determined to be 60.43 ± 19.59 nm. The overall distribution of silver nanoparticles was well dispersed within the range of 30 to 60 nm, confirming that the synthesised AgNPs are less than 100 nm in size (NPs) (**Fig. 2(b)**) [35]. Energy dispersive X-ray (EDX) analysis was performed to examine the composition of elements present in the sample. The EDX spectra showed a strong signal in the silver region, supporting the hypothesis that the biomolecules binding to the AgNPs contributed to their formation. The elemental analysis revealed that silver made up the largest proportion by weight, approximately 54.54%, followed by carbon at 22.02% and oxygen at 23.57%. This demonstrated the significant silver content is present in the synthesized nanoparticles. Additionally, the presence of AgNPs was confirmed by a prominent peak in the EDX spectrum at around 3-3.5 keV (**Fig. 2(c)**) [36].

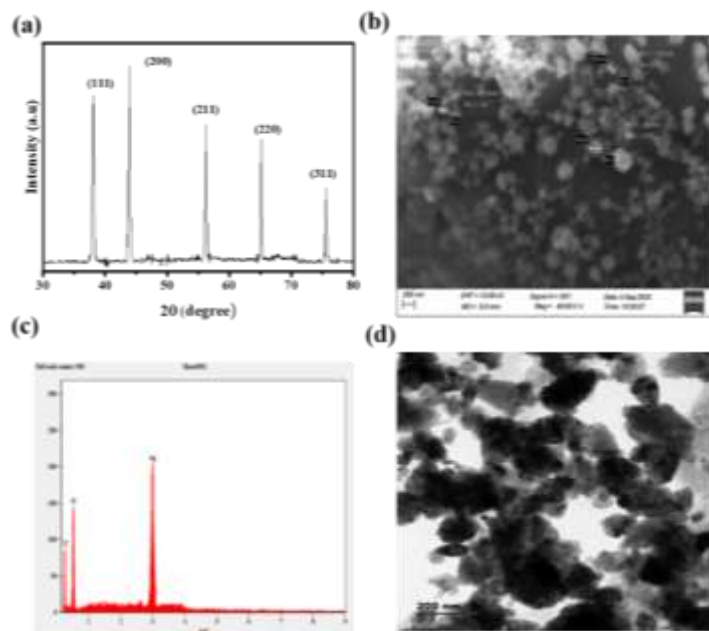


Fig. 2. (a) XRD pattern, (b) SEM image, (c) EDX spectrum and (d) TEM image of biosynthesized AgNPs

The size and morphology of the synthesized nanostructures were further analyzed using Transmission electron microscope (TEM) analysis. Most of the AgNPs exhibited a spherical morphology. Some noticeable variation in the size and shape of

the synthesized nanoparticles were also observed might be due to the physical interaction and aggregations with the present biomolecules. Although the morphology of the silver nanoparticles showed some inconsistency, the spherical morphology remained predominant (**Fig. 2(d)**) [37].

The formation of AgNPs was further confirmed by UV-Vis spectrophotometric analysis, which tracked the reaction progress during the reduction of Ag^+ ions. The reduction of Ag^+ to metallic Ag^0 was confirmed by measuring the absorbance in the range of 400–450 nm. A peak observed at 420 nm signifies surface plasmon resonance (SPR), caused by the excitation of free electrons in the metal during synthesis of AgNPs (**Fig. 3(a)**) [38]. This indicates that the bioactive components of the leaf extract altered the optical properties of the silver nitrate solution, facilitating the silver ion reduction to elemental silver and subsequently to silver nanoparticles. Several studies have also reported the AgNPs resonance peak within this wavelength range [39].

The FTIR spectrum of the AgNPs reveals the biofunctional groups involved in the bioreduction of silver. The absorption spectrum displays the peaks corresponding to components at higher concentrations, indicating the presence of various types of bonds and functional groups, such as amines, halides, ketones and alkanes, which absorb infrared light at different wavelengths [40]. The peak at 3436.49 cm^{-1} is attributed to O-H stretching, hydrogen-bonded, indicating the presence of hydroxyl groups such as alcohol and phenol compounds. The peak at 2986.94 cm^{-1} corresponding to O-H stretching, indicating the presence of carboxylic acids. Further, the observed peak at 1638.01 cm^{-1} is assigned to the N-H bending, indicates the presence of 1° amines. The peak at 1406.40 cm^{-1} corresponding to C-C stretching (in-ring), indicating the presence of aromatic compounds. The peak at 1250.46 cm^{-1} assigned to C-N stretching, suggesting the presence of aromatic amines, while the peaks observed at 1047.57 , 1066.09 and 1076.09 cm^{-1} are associated with the aliphatic amines. The peak at 878.65 cm^{-1} is attributed to N-H wagging, indicating the presence of both 1° and 2° amines (**Fig. 3(b)**) [41, 42]. The spectra, with slight variations in peak positions, suggest that the leaf extract used as a capping agent for the synthesis of silver nanoparticles was retained in the sample.

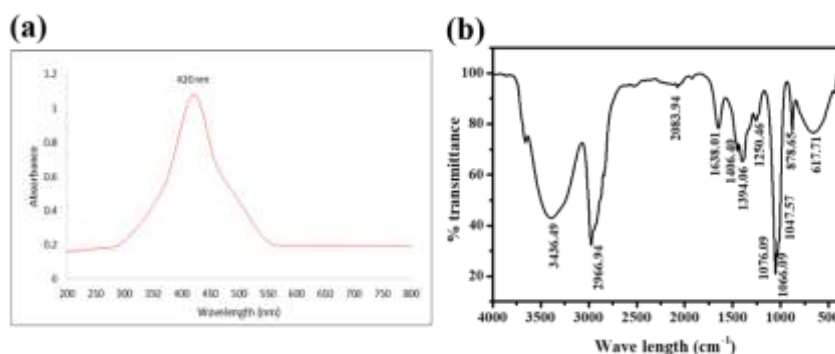


Fig.3. (a) UV-Vis absorption spectrum of the biosynthesised AgNPs at 420nm and (b) FTIR Spectra of the biosynthesised AgNPs

In-Vitro Antioxidant Activity

DPPH radical scavenging activity

The leaf extract and the AgNPs had a notable ability to reduce DPPH free radicals. The rate of scavenging was found to increase with the increase in concentration of the tested sample [43]. Specifically, at concentrations of 20, 40, 60, 80 and $100\text{ }\mu\text{g/mL}$, the *Cinnamomum tamala* leaf extract exhibited $16.62 \pm 0.03\%$, $27.95 \pm 0.11\%$, $43.32 \pm 0.23\%$, $60.45 \pm 0.28\%$ and $79.34 \pm 0.41\%$, scavenging activities, respectively. While the scavenging activities AgNPs were found to be $20.65 \pm 0.09\%$, $32.49 \pm 0.15\%$, $48.86 \pm 0.32\%$, $65.99 \pm 0.42\%$ and $86.64 \pm 0.55\%$ respectively (**Fig. 4(a)**). Additionally, the highest antioxidant activity was observed for bio-synthesized AgNPs employing *Cinnamomum tamala* leaf extract, with an IC_{50} value of approximately $58.88\text{ }\mu\text{g/mL}$, compared to the leaf extract which is about $65.65\text{ }\mu\text{g/mL}$. This observed reports suggests that the AgNPs exhibited higher antioxidant activity, potentially due to the unique chemical composition of the sample. The biosynthesis of AgNPs using *Cinnamomum tamala* leaf extract involves several secondary metabolites, such as flavonoids, tannins, carotenoids, lycopene, carbohydrates, triterpenoids, glycosides, phenolic compounds, steroids and alkaloids. These compounds, particularly their ability to donate hydrogen atoms, likely contribute to the enhanced free radical scavenging capacity of the synthesized AgNPs. This ability to scavenge free radicals plays a crucial role in the stability of the AgNPs [44].

Superoxide anion scavenging activity

In the present study, both synthesized AgNPs and *Cinnamomum tamala* leaf extract demonstrated notable scavenging activity against superoxide anions. At various concentrations, 20, 40, 60, 80 and $100\text{ }\mu\text{g/mL}$, the scavenging activities were found

to be $17.24 \pm 0.10\%$, $33.00 \pm 0.26\%$, $53.69 \pm 0.35\%$, $71.42 \pm 0.42\%$, $87.19 \pm 0.53\%$, respectively for AgNPs and $15.76 \pm 0.08\%$, $29.55 \pm 0.22\%$, $49.26 \pm 0.31\%$, $67.48 \pm 0.36\%$, $82.75 \pm 0.44\%$, respectively for the leaf extract (**Fig. 4(b)**). The IC_{50} values for the synthesised AgNPs and leaf extract were found to be $57.18 \mu\text{g/mL}$ and $61.20 \mu\text{g/mL}$, respectively. Both the leaf extract and AgNPs exhibited concentration-dependent increase in superoxide radical scavenging activity. The enhanced antioxidant properties of the AgNPs can be attributed to the presence of various bioactive compounds in the *Cinnamomum tamala* leaf extract, including polyphenols, flavonoids and tannins. These phytochemicals might contribute to the observed significant antioxidant activity, effectively neutralizing free radicals and protecting cells from oxidative stress-induced damage [45].

Hydroxyl radical scavenging activity

In this present study, the hydroxyl radical scavenging activity of the AgNPs and leaf extract was evaluated at various concentrations of 20, 40, 60, 80 and $100 \mu\text{g/mL}$. The observed reactivity was found to be 20.57 ± 0.23 , 36.21 ± 0.39 , 57.61 ± 0.45 , 74.48 ± 0.55 , 89.30 ± 0.73 for AgNPs and 18.51 ± 0.15 , 31.68 ± 0.26 , 52.67 ± 0.37 , 70.78 ± 0.53 , 85.59 ± 0.69 for the leaf extract. The average IC_{50} values being 53.58 and 57.86 , respectively for the AgNPs and the leaf extract (**Fig. 4(c)**). The lower IC_{50} value observed for the biosynthesized AgNPs indicates a superior antioxidant activity compared to that of *Cinnamomum tamala* leaf extract [46]. This enhanced activity can likely be attributed to the presence of various bioactive compounds in the leaf extract, which might contribute to the overall effectiveness of the AgNPs.

Hydrogen peroxide scavenging activity

In this study, hydrogen peroxide scavenging activity of the synthesized AgNPs and *Cinnamomum tamala* leaf extract was assessed. At concentrations 20, 40, 60, 80 and $100 \mu\text{g/mL}$, the scavenging activities of synthesised AgNPs ranged from $16.32 \pm 0.06\%$, $28.97 \pm 0.12\%$, $47.95 \pm 0.20\%$, $69.18 \pm 0.34\%$, $80.61 \pm 0.49\%$ and while the leaf extract exhibited the following activities: $13.67 \pm 0.03\%$, $25.10 \pm 0.09\%$, $43.26 \pm 0.16\%$, $65.51 \pm 0.28\%$, $78.57 \pm 0.34\%$, respectively. The IC_{50} value of the *Cinnamomum tamala* leaf extract and the AgNPs was found to be $65.61 \mu\text{g/mL}$ and $61.50 \mu\text{g/mL}$, respectively (**Fig. 4(d)**). Although both samples demonstrated hydrogen peroxide scavenging activity, it was notably lower than their DPPH scavenging abilities. This difference in scavenging potential is likely due to the bioactive compounds present on the surface of the AgNPs, which may have a more pronounced effect on other free radicals but exhibit a relatively lower capacity against hydrogen peroxide [47].

Nitric oxide scavenging activity

In this study, the nitric oxide scavenging activity of the synthesized AgNPs and leaf extract was evaluated at various concentrations viz., 20, 40, 60, 80 and $100 \mu\text{g/mL}$. The scavenging activities of the AgNPs were found to be $19.93 \pm 0.08\%$, $37.76 \pm 0.26\%$, $51.39 \pm 0.32\%$, $68.88 \pm 0.46\%$, $88.81 \pm 0.71\%$, respectively, while the activities of the leaf extract was found to be $17.13 \pm 0.05\%$, $32.86 \pm 0.19\%$, $45.45 \pm 0.26\%$, $63.28 \pm 0.35\%$, $83.56 \pm 0.62\%$, respectively. The average IC_{50} values was found to be $56.02 \mu\text{g/mL}$ and $61.89 \mu\text{g/mL}$, respectively (**Fig. 4(e)**). The nitric oxide scavenging activity observed was lower than both the DPPH and hydrogen peroxide scavenging activities. However, the results still suggest that the synthesized AgNPs possess potential antioxidant properties. Given their significant scavenging capacity, these AgNPs may have applications in future antioxidant formulations [48].

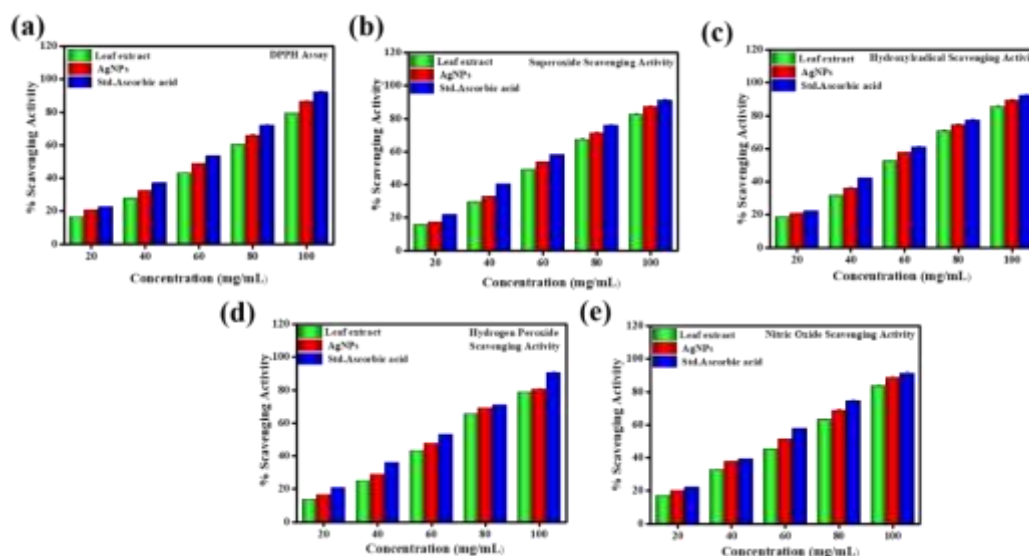


Fig.4. *In-vitro* antioxidant studies of leaf extract, AgNPs and std. ascorbic in (a) DPPH radical scavenging activity, (b) Superoxide anion scavenging activity, (c) Hydroxyl radical scavenging activity, (d) Hydrogen peroxide scavenging activity and (e) Nitric oxide scavenging activity.

4. CONCLUSION

The biosynthesized AgNPs through the reduction of silver nitrate using *Cinnamomum tamala* leaf extract exhibited a significant antioxidant radical scavenging activity, with the effect increasing in a dose-dependent manner. The AgNPs displayed superior antioxidant activity, approaching that of the standard ascorbic acid. Hence, this green eco-friendly synthesis method provides a promising alternative to traditional physical or chemical approaches for the production of nanoparticles, in-particular the *Cinnamomum tamala* leaf extract. The leaf-mediated synthesis process inherently have bioactive phytoconstituents which are biocompatible and offer a range of potential applications such as anti-inflammatory, antiarthritic and anti-oxidant properties. Given the potent antioxidant properties of the biosynthesized AgNPs which also holds great potential as drug carriers for targeting cancer cells.

DECLARATION OF COMPETING INTEREST

The authors claim that no known conflicting financial interests or personal relationships appeared to have an impact on the work that was published in this paper.

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CREDIT AUTHORSHIP CONTRIBUTION STATEMENT

Shireen Farhana S: Writing-Original draft. Chandrasekar Shobana: Investigation, Writing-review & editing. Boopathy Usharani: Writing-review & editing. D. Rohini: Supervision.

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