Biosynthesis of silver nanoparticles from Rhaphidophora pertusa leaves extract and its characterization, antimicrobial, anti-oxidant and cytotoxicity activities

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Research Article

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Abstract

The current study investigated the bio-synthesis of AgNPs (silver nanoparticles) through the use of an extract from the leaves of *Rhaphidophora pertusa (R.pertusa)*. Visual confirmation of the formation of biosynthesized *R.pertusa*silver nanoparticles (RP-AgNPs) came from observing the colour difference between them. Additionally, it has been examined through FT-IR, HR-SEM, EDAX, XRD, HR-TEM and DLS. At 424 nm the presence of AgNPs has been confirmed by UV-vis spectra. FT-IR reveals the secondary molecules of *R.pertusa* which are bound with AgNPs contain OH and NH functional groups. The synthesised particles' sizes are validated by HR-TEM which is approximately 19 nm, whereas EDAX investigations have substantiated the existence of AgNPs. The FCC crystallinity of the synthesised RP-AgNPs is confirmed by the XRD report. The size distribution of the particles with a range of 177 nm is discovered using DLS analysis. Significant antioxidant properties are displayed by the bio-synthesised RP-AgNPs for DPPH. Furthermore, it was found that RP-AgNPs are extremely receptive to both thick peptidoglycan and thin peptidoglycan microbes. The MCF-7 cell line is used to test the cytotoxicity of the synthesised RP-AgNPs and the research shows that the MCF-7 cell proliferation is effectively inhibited by the synthesised RP-AgNPs. As a result, the present research findings have enormous potential for the delivery of medication.

1. Introduction

Nanotechnology is a burgeoning and stimulating scientific and innovation technology. Nanotechnology has considerable advancement in the departments such as physiochemical, medicine, biotechnology, engineering and space industries too [1]. The nanoparticles have various dimensions and size ranges between 1 nm and 100 nm. A lot of techniques have been made in the particular study. It is attainable to order these techniques into classes namely physical, chemical and biosynthesis. The outcome of physical synthesis seems to be of excellent calibre with no unique structural flaws. However, the expense of these techniques is high and a lower quantity is delivered in each time. Nanotechnology has improved living conditions in chemical synthesis; however it has increased pollution, which contaminates the air. Nano contamination is the term for pollution brought by nanotechnology. Therefore green synthesis is a methodology that is safe for the environment, eco-friendly, non-toxic, and non-harmful methodology [2–4].

Nowadays, green synthesis techniques for nanoparticles have been recommended over traditional techniques due to their advantages in terms of cost, energy use, non-toxicity, and safety. In the bio-synthesis method, a living organism acts as a capped and reductant agent for synthesizing process [5]. The plant extract's biological by-products such as terpenoids, tannins, alkaloids, saponins, tannins and flavonoids are the response to the synthesis of AgNPs (from Ag^{+} ion to Ag^{0} metal) during the process. In this research, bio-synthesis of AgNPs was done with medicinal plant leaves extract in bottom-up approach [6, 7].

Rhaphidophora Pertusa is a perennial, robust, climbing plant which belongs to the *Araceae* family cultivated throughout India, Malaysia and Australasia. In some parts of Kerala the tribal peoples used to prepare medicated oil by using *R.pertusa* plants for curing some skin diseases. Also, it is used for treating ascites, liver and spleen-oriented disease [8, 9].

As indicated by the WHO (World Health Organization) report, in the year 2020, 2.3 billion women are found to have breast cancer growth and there are 685,000 passing worldwide. Towards the end of the year 2020, there are 7.8 million living women will be diagnosed with breast cancer malignant growth in the 5 years, making it the most common disease on the planet [10]. Chemotherapy is the only treatment which is used to kill the cancer cell. There are many types of chemotherapy treatment are available with respect to the disease. Even though chemotherapy kills the cancer cells, it also damages the DNA (Deoxyribonucleic acid) [11, 12]. Furthermore, chemotherapeutics are very expensive and have numerous side effects. As a result, it is critical to develop low-cost, beneficial cancer therapies. Hence the standard cytotoxicity *in-vitro* is a significant advantage of employing AgNPs for cancer therapy [13].

The researchers used decoction of *R.pertusa* leaf to synthesis non-toxic, eco-friendly, and economically affordable AgNPs, and their final products are evaluated by UV-Vis spectra, X-Ray diffraction, High Resolution- Scanning Electron Microscope, EDAX, High Resolution–Transmission Electron Microscope, DLS and ZETA potential. The disc diffusion method is used to conduct antimicrobial research on thick peptidoglycan layer and thin peptidoglycan layer microorganisms. Furthermore, the efficacy of anti-oxidant and cytotoxicity against MCF-7 is being scrutinized.

2. Materials And Methods

2.1 Materials

The *Rhaphodophora pertusa* (Araceae family) leaves used in the study are obtained in Namakkal, Tamil Nadu, India, at Kolli hills (11.2485° N, 78.3387° E). Merck provides silver nitrate ($AgNO_3$) with a purity of 99.5%. Double Distilled Water (DDW) is used for all experiments.

2.2 Preparation of RPLE

To eliminate undesirable dust and debris, the collected *R.pertusa* leaves are washed numerous times with double distilled water. The washed leaves are left to dry in the shade for an extended period of time. The moisture desiccated leaves are then turned into fine particles. Two gram of pulverized *R.pertusa* leaves are dissolved in 100 mL of DDW and soaked for 15 min. The soaking mixture is held at 70°C in a heating mantle until it vaporises and leaves a distinct fragrance of leaves. The mixture is cooled to room temperature before being filtered using Whatman filter paper and the leaf extract is preserved for future use.

2.3 Bio-synthesis of AgNPs

By dissolving 17×10^{-3} g of $AgNO_3$ with 1 L of DDW to make 1 mM of $AgNO_3$ Solution. A 200 mL beaker is filled with 45 mL $AgNO_3$ solution (1 mM). The mixture is then stirred with 5 mL of RPLE for an hour using a magnetic stirrer, while the colour difference in the mixture is observed. The RPLE and the $AgNO_3$ solution (1 mM) are mixed with a ratio of 1:9 respectively. The colour changes are visible in the synthesized solution which has a pH of 7.8. The bio-synthesized $AgNO_3$ solution is centrifuged many times at 12,000 rpm for 15 minutes using a Remi 12C machine. Finally, the reduced RP-AgNPs solution is dried in a hot air oven at 200°C for 15 minutes and powdered for future use. Afterwards, the dried RP-AgNPs underwent UV-Vis spectroscopy analysis and the peak at a specific nm scale confirmed the presence of the AgNPs.

2.4 Characterization studies of bio-synthesized AgNPs

The bio-reduction of $AgNO_3$ to AgNPs using *R.pertusa* is monitored on a regular basis using a UV-Vis Spectrometer with a wavelength range of 190–1100 nm. Perkin Elmer FT-IR is used to investigate the functional group and associated bonds present in the synthesised sample. The bio-products in the leaf extract might also be examined using FT-IR. The DLS method is used to examine the hydrodynamic dimension of the NPs in the material. Malvern PAnalytical Nano Zs is utilized to examine the Zeta potential of the synthesised AgNPs. X-ray diffraction investigations utilising PAnalytical Xpert Plus are used to determine the crystallinity of the biosynthesized AgNPs. XRD is used to diagnose the powdered AgNPs with a CuK radiation $\lambda = 1.54$ nm and a scanning angle of 2θ ranges from 10° to 80°. The morphological structural examinations are carried out using a HR-SEM (FEI Q250 FEG) equipped with an EDAX setup. HR-SEM studies were done in IIT Madras, Tamil Nadu, India. The synthesized AgNPs dry powder underwent analyse of HR-TEM (JEOL-2100+) in Alagappa University, Karaikudi, Tamil Nadu. The antimicrobial and cytotoxicity test is experimented in BioTechno solutions, Trichy, Tamil Nadu, India.

2.5 Antimicrobial properties

2.5.1 Collection of test organism

The aim of the test is to break down the antimicrobial exercises of bio-synthesized silver nanoparticles towards chosen microbes, utilizing disc diffusion technique. The bactericidal efficacy of bio-synthesized AgNPs is investigated with thick peptidoglycan pathogens such as *Staphylococcus aureus (MTCC 25923)*, *Bacillus subtilis (MTCC 2451)*, *Streptococcus aureus (MTCC 273)* and thin peptidoglycan strains such as *E.Coli (MTCC 25922)*. MTCC in Chandigarh, India provide the bacterial species. These bacterial species are grown at 37°C and stored at 4°C on supplement agar slant (Difco, USA).

2.6 Antifungal properties

2.6.1 Inoculum and fungal strains collection

Agar growth medium containing peptones was utilised for the antifungal test. The infectious strains are injected autonomously for 6 hour through agar growth/stock and the solution is tested for 10^5 CFU/mL.

Aspergillus flavus (MTCC 356) and *Aspergillus niger (MTCC 227)* are used in the clinical tests for the live organisms under consideration, which are procured from the NCL, Maharashtra, India.

2.6.2 Antioxidant analysis

The antioxidant properties of the bio-synthesized AgNPs are investigated using the DPPH free radical scavenging action (A.Braca et al.,79 (2002)). A 250 µL Ethanolic solution of DPPH (0.05 mL) is introduced to 1000 µL of bio-synthesized NPs with varies doses (100–500 g/mL). The DPPH solutions are newly produced then stored at 4°C inside the darkness. The combination runs through for 5 minutes and the absorbance is measured spectrophotometrically at 517 nm. To set the absorbance to zero, ethanol is used. A clear example of having a comparable amount of ethanol and DPPH are also ready. All conclusions are repeated three times, communicated as the level of hindrance is determined by the accompanying condition (Yen, G.C. and Duh, P.D. 1994).

DPPH activity (Inhibition %) = [(A - B) / A] x 100

Here, A and B are the absorbance advantages of test and prominent example correspondingly.

2.6.3 Cytotoxic activity

The pattern of bio-synthesized RP-AgNPs is tested for *in-vitro* cytotoxicity and the usage of MCF-7 cells using the 3-(4,5-dimethylthiazol-2-yl)-2, 5-diphenyltetrazolium bromide (MTT) assay. Briefly the cultivated MCF-7 cells are collected by enzymes secreted and pooled in a 15 mL vial. The cells are then plated on a 96-well tissue subculture plate at a density of $1x10^5$ cells/mL cells/well (200 µL) in Dulbecco's Modified Eagle's Medium (DMEM) containing 10% FBS and 1% antibiotic solution for 24 to 48 hour at 37°C. All wells were rinsed with sterile PBS and incubated with various concentrations of the RP-AgNPs pattern in serum-free DMEM media. All patterns are repeated three times and the cells have been cultured for 24 hour at 37°C in a humidified 5% CO_2 incubator. Following the incubation time, MTT (20 µL of 5 mg/mL) is poured to all well and the cells are incubated for another 2 to 4 hour or until the pink precipitates are clearly visible beneath an inverted microscope. Finally, the medium containing MTT (220 µL) aspirated from the wells and rinsed with 1X PBS (220 µL). Furthermore, a 100 µL of DMSO is added and the dish is shacked for 5 minutes to dissolve formazan crystals. The absorbance recorded at 500 nm for each well using a micro plate reader and the percentage of cell viability and IC_{50} value are computed using Graph Pad Prism (6.0) software (USA).

3. Results And Discussion

3.1 UV-Vis analysis of bio-synthesized RP-AgNPs

UV-vis spectral analysis is used to evaluate AgNPs synthesised from *R.pertusa* leaves extract. The optical quality of a metal nanoparticle is one of its most essential characterizations. Fig.2 depicts the formation of AgNPs after *R.pertusa* exposure, which is visible as a colour shift from light to dark brown. The colour change attributed to SPR [14]. UV-visible spectrum characterization is done for $AgNO_3$ solution of 1 mM,

raw leaves extract of *R.pertusa* and the solution of bio-synthesized RP-AgNPs. The researchers discovered a prominent SPR peak at 424 nm in the synthesised RP-AgNPs which is the characterization peak of AgNPs [15].

3.2 Powder X-Ray Diffraction analysis of RP-AgNPs

The phase angle and crystalline structure of the bio-synthesized AgNPs are validated using a powder XRD profile. The RP-AgNPs XRD pattern is shown in Fig.3. The researchers detected five strong peaks in this pattern at the 2q angles of 32.40°, 38.12°, 46.22°, 64.46° and 77.02°, these peaks are associated to the planes (110), (111), (200), (220) and (311) respectively. The powder XRD patterns are corresponds to the FCC (face centered cubic) of the standard silver metal (JCPDS No. 04-0783). The formation of the XRD crystallographic profile revealed that the element is silver and crystalline and no phase other than FCC is formed. Some of the little-noticed peaks (*) and other lower-degree value peaks occur as a result of phytochemicals present in the leaf extract. The average size of the crystalline is 23.37 nm found by using Debye-Scherer's formula [16-19].

3.3 *FT-IR profile analysis*

FT-IR is the most significant analyze for identify the biomolecules contribution to the reduction of silver ions and stabilizing the biogenic AgNPs. The Fig.4 shows the FTIR spectrum of the *R.pertusa* leaves extract and the bio-synthesized RP-AgNPs. The FTIR spectroscopy of RP-AgNPs have showed the peak at 3433 cm^{-1} which is responsible for water H=O bonding [7]. The absorption peak at 2920 cm^{-1} and 2850 cm^{-1} are responsible for C-H stretch alkane group [20]. The peak at 1631 cm^{-1} is responsible for N-H bond for primary amines, the peak at 1383 $cm^{-1}$and 1353 cm^{-1} are C-C stretch (in-ring) for aromatics [21]. The absorption peak at 1112 cm^{-1} represents C-O stretching (easter). The peak at 1081 cm^{-1} corresponds to –C-O-C and C-OH stretching [22]. The peak appeared at 875 cm^{-1} is due to alkyl halides [23]. The peaks at 780 $cm^{-1}$and 668 cm^{-1} are corresponding to C-H stretching [24,25]. From the above mentioned FT-IR combination of the crude extract and synthesized RP-AgNPs, some of the peaks are in the synthesised RP-AgNPs are vanishes, which are comparing with the *R.pertusa* leaf extract. It could be concluded that some of the biomolecules found in the leaf extract may be involved in the transformation of Ag^{+} ions into Ag^{0} metal nanoparticles [26,27].

3.4 HR-SEM analysis

The morphological representation and Energy Dispersive X-ray spectra of the synthesized AgNPs reduced by *R.pertusa* leaves extract are shown in Fig.5. The bio-synthesized metal nanoparticle is in spherical and has a staunch peak at 3.2 KeV in EDX spectrum, indicating the presence of Ag^{0} [28]. Additional peaks like Si, Cl are apparently associated with the glass substrate coatings. Oxygen might be present due to bio-synthesized AgNPs reduced from *R.pertusa* [29]. Furthermore, the EDX spectrum report shows bio-synthesized nanoparticles with Ag of 61.4 Wt %, 26.1 Wt % of O, 6.7 Wt % Cl, and 5.8 Wt % Si.

3.5 HR-TEM analysis

HR-TEM micrographs of the synthesized *R.pertusa* AgNPs are displayed in the Fig.6. The image shows that the synthesized RP-AgNPs are spherical and dispersed. HR-TEM picture displays the presence of cross-section borders demonstrating crystallinity nature of the AgNPs with "d" spaced value. Furthermore, the SAED image revealed the heterogeneous character of the AgNPs which corresponds to the cross-section planes of Bragg's reflection (110), (111), (200), (220) and (311). Based on the Histogram, the particles size ranges between 14 and 24 nm. The average size of RP-AgNPs is nearly 19 nm which has close value with XRD crystallite size.

3.6 Zeta potential and DLS analysis

The hydrodynamic size of the bio-synthesized AgNPs from DLS is seen at 134.1 nm and the PDI (Polydispersity Index) value of 0.322 shows that the particles are monodispersed with particle sizes ranging from 16.5 nm to 280.8 nm with a mean particle size of 134.1 nm. Furthermore, the interval between the biggest and smallest particles is 117 nm [30]. The zeta potential is taken into a significant parameter to decide the particle sizes and stability. The electro kinetic potential of the bio-synthesized RP-AgNPs is -17.9 mV indicating a high negative charge on the nanoparticles' surface. The negatively charged particles create a powerful repulsive contact amongst particles, preventing aggregation and providing valid stability [31,32].

3.7 Antibacterial analysis of bio-synthesized AgNPs (disc diffusion method)

The disc diffusion technique is used to assess the antimicrobial activities of synthesized silver nanoparticles. Muller Hinton Agar is utilized to make the petri dishes (width 60 mm) which are then injected with tested microorganisms. Purified discs of 6 mm wide are treated with 10 µL of obtained samples. The prepared discs are put on top of the agar discs and kept at ambient temperature for 30 minutes to allow compound diffusion. A positive control is made using 10 µL of Amoxicillin as the typical bacterial colonies. The plates are maintained at 37 °C for 24 hour and zone of inhibiting is measured in millimeters. The experiment is carried out twice for reducing the error.

Table.1

Antibacterial analysis of bio-synthesized RP-AgNPs with varies bacteria.

Samples	Concentrations (µL/mL)	Bacteria / Inhibition Zone (mm)			
		Staphylococcus aureus	*Bacillus subtilis*	*E.Coli*	*Streptococcus aureus*
A (Amoxicillin)	10	8.4±1.82514	8.1±1.6329	8.2±2.58199	6.6±1.82574
B ($AgNO_3$)	10	0±0	0±0	0±0	0±0
C (*R.Pertusa* Leaf extract)	10	2.2±0.8165	0±0	2.2±0.8165	1.1±0.8165
D (RP-AgNPs)	10	5.1±1.82574	4.2±1.82574	3.1±1.8257	2.3±0.8165

(The population means differs considerably at the 0.05 level)

The antimicrobial efficiency of biosynthesized RP-AgNPs is investigated by disc diffusion method and results of these tests are as shown in the Fig.8. From the result, it is seen that the raw leaves extract of the *R.pertusa* alone has an antimicrobial effect on *Staphylococcus aureus*, *E.Coli* and *Streptococcus aureus* which do not have an effect on *Bacillus subtilis*. The zone of inhibition diameter of the bacteria is given in the Table.1. When it is compare with the inhibition diameter, it is seen that the *Streptococcus aureus* has the least inhibition diameter of 2.3±0.8165 mm. *Staphylococcus aureus*, *Bacillus subtilis* and *E.coli* are very much affected by bio-synthesized RP-AgNPs of 5.1±1.82574 mm, 4.2±1.82574 mm, 3.1±1.82574 mm respectively. In addition, it is seen that the synthesized RP-AgNPs have a brilliant effect on a thick peptidoglycan and thin peptidoglycan bacteria. Furthermore, the research done with the stem of *R.pertusa* plant compared to AgNPs, the reaction done with AgNPs gives the better antibacterial effect. And this is the first AgNPs synthesized from *R.pertusa* leaf extract [33]. Based on this, the bio-synthesized RP-AgNPs showed the significant antimicrobial activity, they could be the potential to be broadly in medication applications.

3.8 Antifungal analysis of bio-synthesized AgNPs

The disc diffusion approach is used to analyse the sample's antifungal activity. SDA (Sabouraud's dextrose agar) is used to make the petri dishes (60 mm of width) which are then inoculated by tested microorganisms. The 6 mm width of sterile discs are treated with 10 µL of various samples. The arranged discs are put on upper layer of the agar media then kept at ambient temperature for 30 minutes to allow additive penetration. The positive control is made using 10 µL of Fluconazole as the typical antimicrobial agent. The discs are incubated for a 24 hour period at 37 °C and the zone of inhibitory is measured in mm. The results of the antimicrobial tests on the RP-AgNPs are given in Fig.9. Hence, it is seen that the bio-synthesized RP-AgNPs is more successfully responded with *Apergillus niger* (4.3±1.82574 mm) than *Aspergillus flavus* (3.2±0.8165 mm). Additionally, the crude plant extract of the *R.pertusa* alone

responded with *Aspergillus niger* (3.1±1.82574 mm) and not for *Aspergillus flavus*. Fluconazole is taken control over here. The inhibition zones are recorded in Table No.2.

Table.2

Inhibition zone representation of different fungal samples treated with *R.pertusa* AgNPs

Samples	Concentration of the solution (µL/mL)	Organisms /Inhibition zone (mm)	
		Aspergillus flavus	*Aspergillus niger*
A (Fluconazole) B ($AgNO_3$)	10 µL	9.1±2.94392	8.1±1.82574
	10 µL	0±0	0±0
C (*R.pertusa* L. extract)			
D (RP-AgNPs)	10 µL	0±0	3.1±1.82574
	10 µL	3.2±0.8165	4.3±1.82574

(The population means differs considerably at the 0.05 level)

3.9 Anti-oxidant analysis (DPPH free radical scavenging)

Many research studies have investigated the antioxidant properties of AgNPs and found that they have excellent antioxidant activity [34]. The bio-synthesized AgNPs using *R.pertusa* leaf crude extract have been shown to have anti-oxidative activity compared to $C_6H_8O_6$ (ascorbic acid) as a control. The antioxidant assay results for 100, 200, 300, 400, and 500 µg/mL of RP-AgNPs obtained 64.38, 69.57, 71.34, 75.49 and 77.53% respectively. When the concentration of RP-AgNPs increases which also increases the amount of antioxidant [35].

Table.3

DPPH free radical Antioxidant analysis of bio-synthesized RP-AgNPs with Ascorbic acid

S.No	Concentration	Anti-oxidant effect by DPPH (%)	
		RP-AgNPs	Ascorbic acid
1	100 (µg/mL)	64.38±0.82061	76.63±1.85131
2	200 (µg/mL)	69.57±1.8331	79.25±4.79601
3	300 (µg/mL)	71.34±1.80091	80.38±4.41213
4	400 (µg/mL)	75.49±1.84039	83.16±2.95752
5	500 (µg/mL)	77.53±1.84039	85.67±1.84036

3.10 Cytotoxicity testing

Silver nanoparticles have a high anticancer potential and have been widely employed in medicine. Bio-synthesized AgNPs have the potential to disrupt cell proliferation and cause cell death. [36].The anticancer impact of RP-AgNPs on MCF-7(Human Breast Cancer cell) is investigated in this work. The bio-synthesized RP-AgNPs destroy the DNA (Deoxyribo nucleic acid) and induce cell death. The viability of the cancer cell decreased when the dosage of RP-AgNPs increased from 10 µg/mL to 100 µg/mL. The minimum inhibitory concentration (IC_{50}) is seen at 102.4 µg/mL. Besides, the decrease in MCF-7 cell practically exhibits the counter disease impact of synthesized RP-AgNPs. A huge morphology like shrinkage of cell, loss of cell membrane and the volume of the cells shows the expected anticancer impacts of RP-AgNPs which is explicated in the Fig.11 [37,38].

Conclusion

The researchers successfully synthesized non-toxic, non-chemical, non-hazardous eco-friendly and economically sound bio-synthesized AgNPs reduced from *Rhaphidophora pertusa* leaf extract. The results are interpreted with different analytical instruments like UV-vis spectrometer, HR-SEM, HR-TEM, XRD, DLS, Zeta potential FTIR and EDAX spectra. The UV-Vis spectra show the peak at 424 nm which confirms the presence of silver. Besides, characterization studies of FTIR, EDAX and powder XRD also confirm the presence of AgNPs. From HR-TEM, the sizes of the particles of the bio-synthesized AgNPs are 19 nm. The stability of the bio-synthesized AgNPs is examined by zeta potential is -17.9 mV which indicates a strong negative charge on the surface of the nanoparticles. Also, RP-AgNPs are undergone anti-microbial and anti-oxidant assays which show good potential. The bio-synthesized AgNPs are at lower concentrations do not exhibit cytotoxicity on MCF-7 cells, however, the higher concentration (IC_{50} value: 102.4 µg/mL) shows the brilliant cytotoxic activity. Hence, the bio-synthesized *Rhaphidophora pertusa* AgNPs could be a good potential alternative for human pathogenic bacterial disease and breast cancer.

Abbreviations

$AgNO_3$ – Silver Nitrate

AgNPs – Silver nanoparticles

CFU/mL – Colony Forming Unit/ milli liter

DDW – Double Distilled Water

DLS –Dynamic Light Scattering

DMEM – Dubecco's Modified Eagle Medium

DMSO – Dimethyl sulfoxide

DPPH - 2,2-diphenyl-1-picrylhydrazyl

EDAX – Energy Dispersive X-ray micro analysis

FCC – Face Centered Cubic

FT-IR – Fourier Transform –Infra Red

HR-SEM – High Resolution – Scanning Electron Microscope

HR-TEM – High Resolution Tunneling Electron Microscope

JCPDS – Joint Committee on Powder Diffraction Data

KeV – Kilo electron Volt

µL - micro Liter

mM – milli Molar

MTCC – Microbial Type Culture Collection

NCL – National Chemical Laboratory

RP-AgNPs – Rhaphidophora pertusa- Silver nanoparticles.

RPLE – Rhaphidophora pertusa Leaf Extract

SAED – Selected Area Electron Diffraction

UV-Vis – Ultra Violet visible spectra

XRD – X-Ray Diffraction.

Declarations

Ethical Approval

We also declare that the study was performed according to the international, national and intuitional rules considering animal experiments, clinical studies and biodiversity rights. And also no animals / humans were harmed in this research work.

Competing interests

The authors declare that they have no competing interest.

Author's contributions

Authors

We confirm that the manuscript has been read and approved by all named authors and that there are no other persons who satisfied the criteria for authorship but are not listed. We further confirm that the order of authors listed in the manuscript has been approved by all of us.

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Availability of date and materials

The data that support the findings of this study are available from the corresponding author [J TJ Prakash] upon reasonable request.

Corresponding Author

We understand that the Corresponding Author is the sole contact for the Editorial process. She is responsible for communicating with the other authors about progress, submissions of revisions and final approval of proofs

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We declare that this manuscript is original, has not been published before and is not currently being considered for publication elsewhere.

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Figures

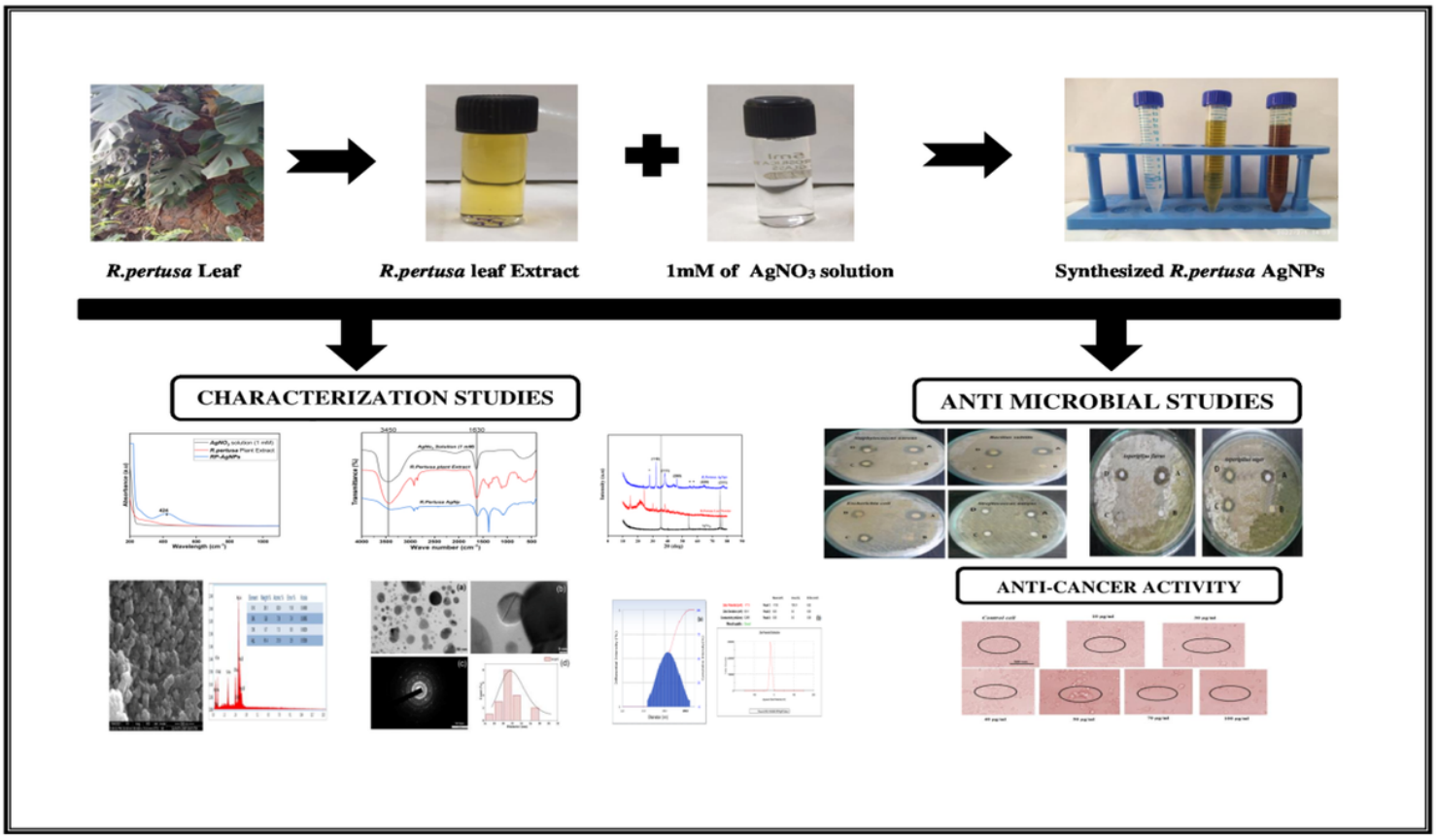

Figure 1

Graphical illustration of synthesis and characterization of *R.pertusa*AgNPs

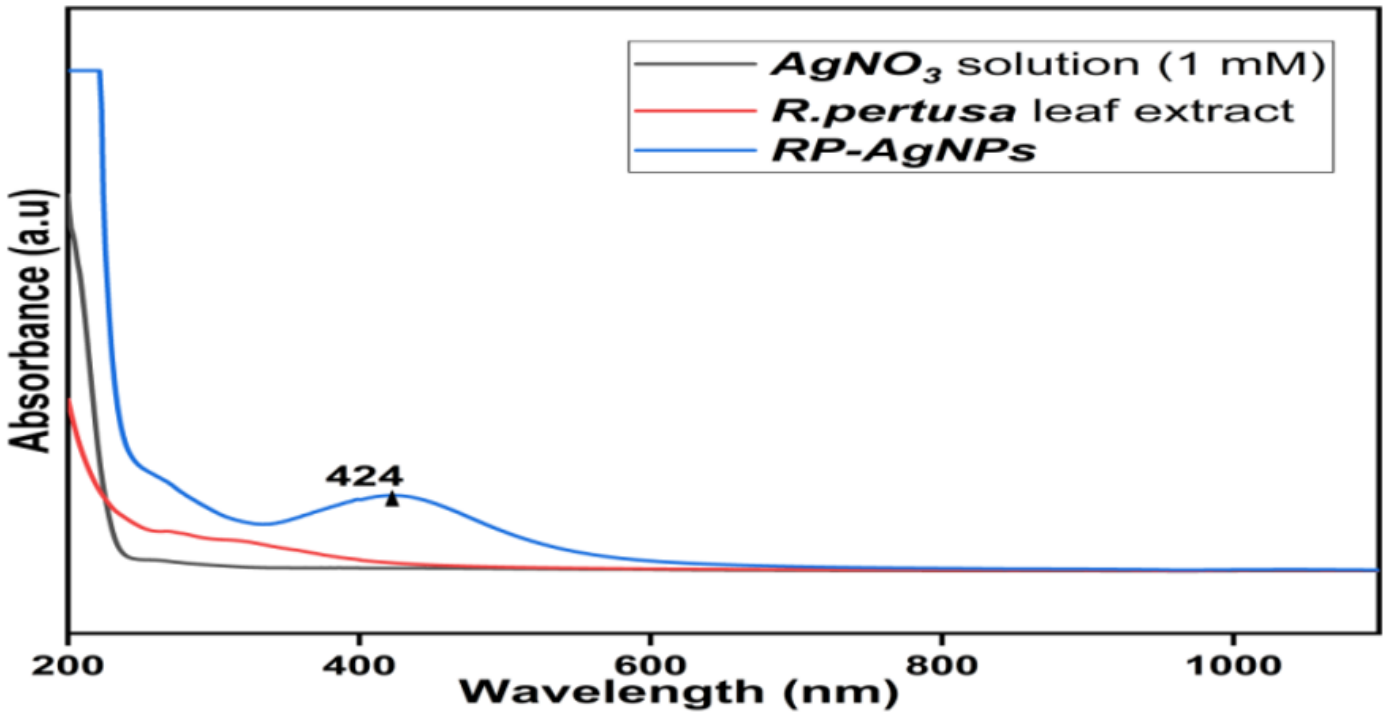

Figure 2

UV-vis spectra of 1 mM AgNO3solution, *R.pertusa* leaf extract and bio-synthesized RP-AgNPs

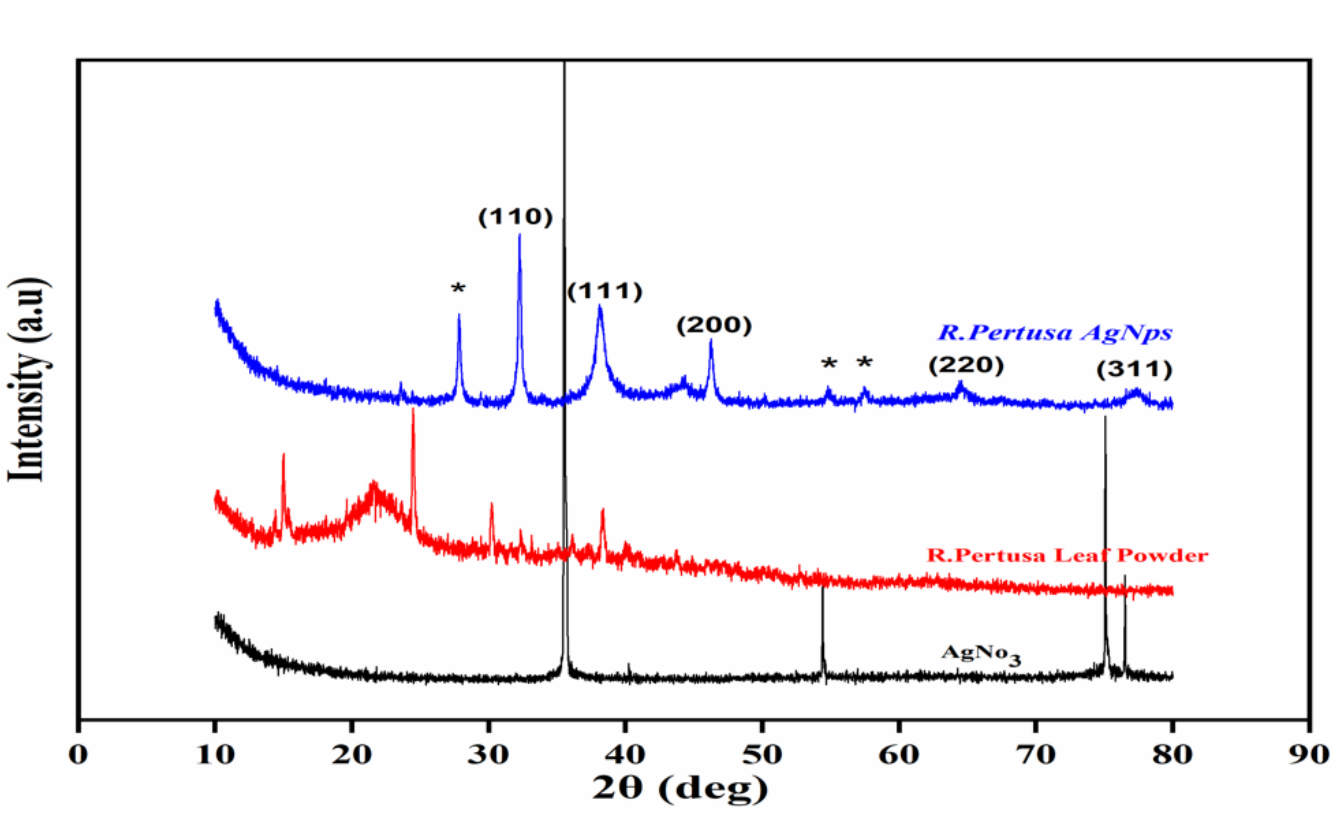

Figure 3

Powder XRD pattern of AgNO3, *R.pertusa* leaf powder and bio-synthesised

RP-AgNPs

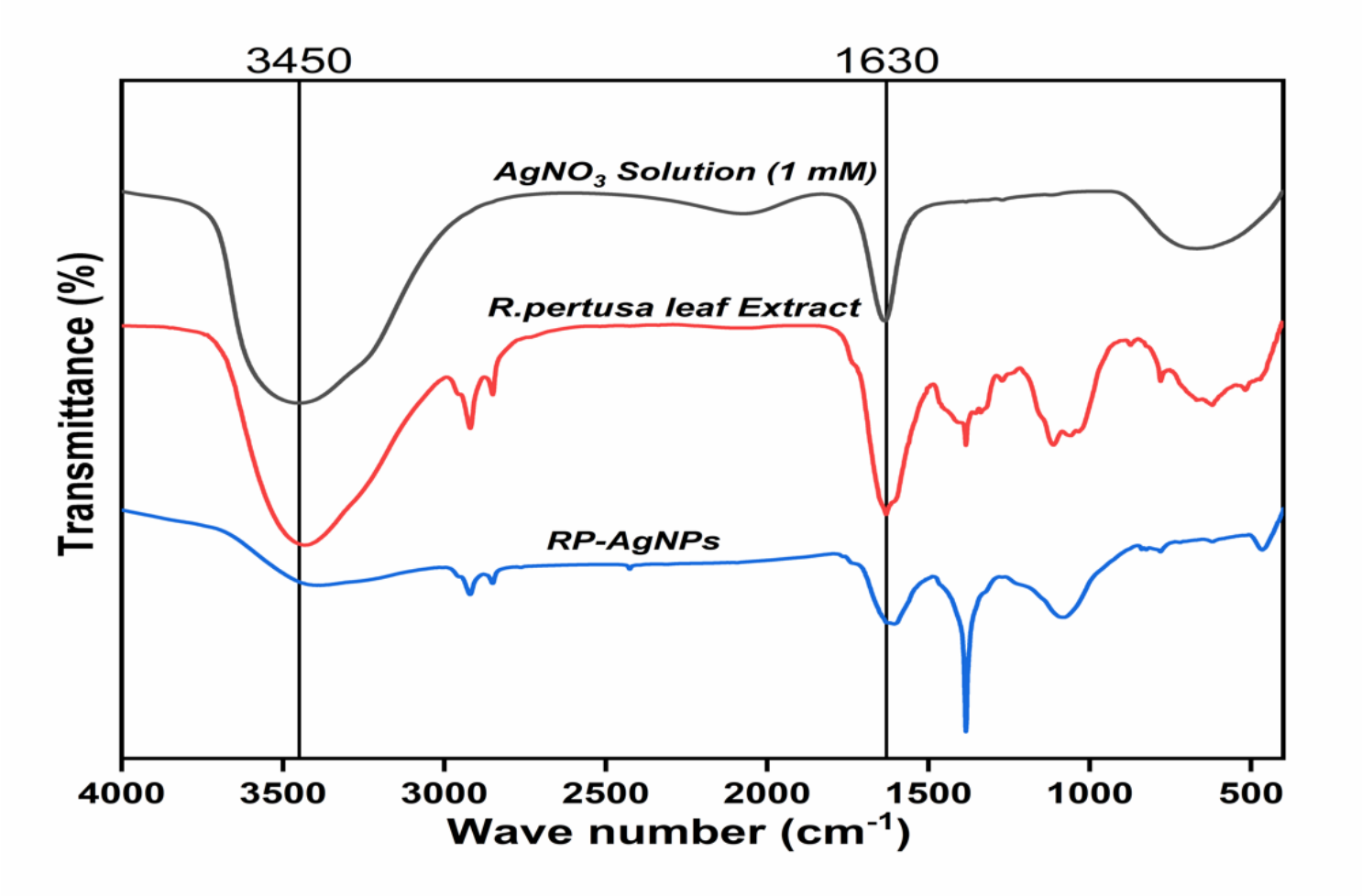

Figure 4

FT-IR profile of AgNO3solution, *R.pertusa* leaf extract and bio-synthesized RP-AgNPs

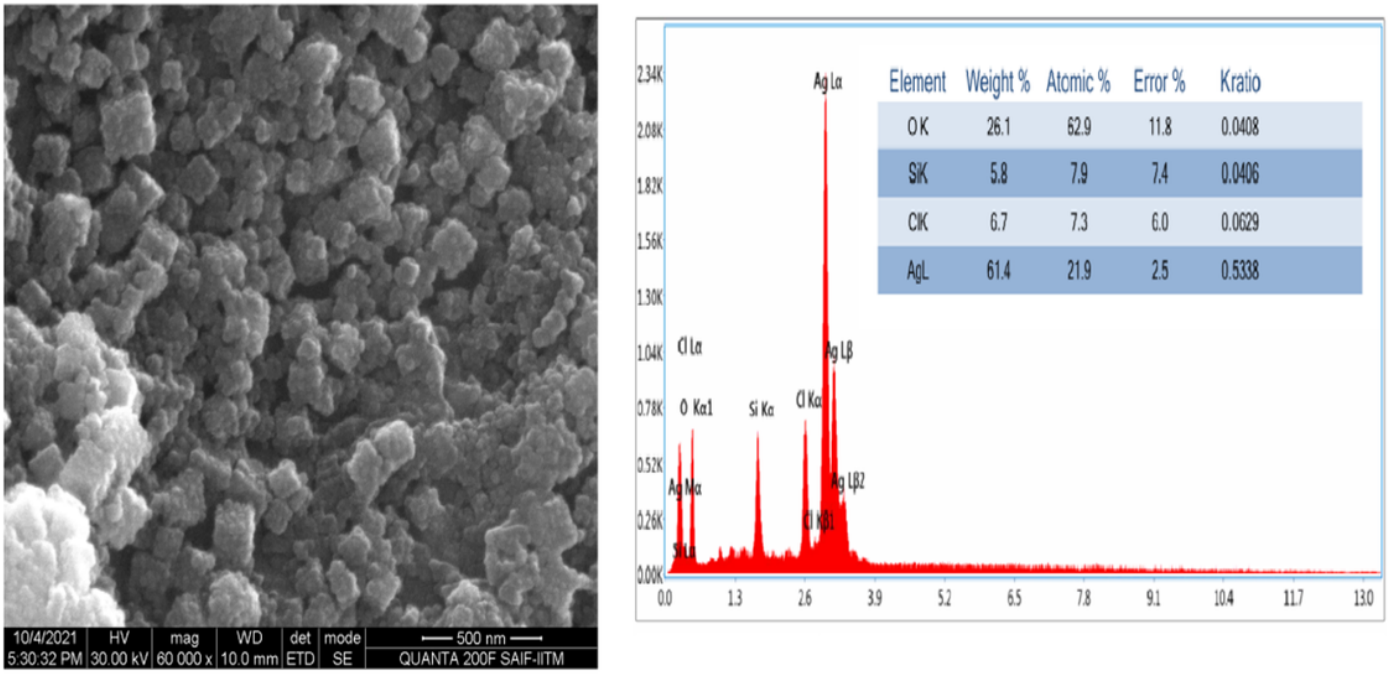

Figure 5

HR-SEM and Enery Dispersive X-ray spectrum of bio-synthesized RP-AgNPs

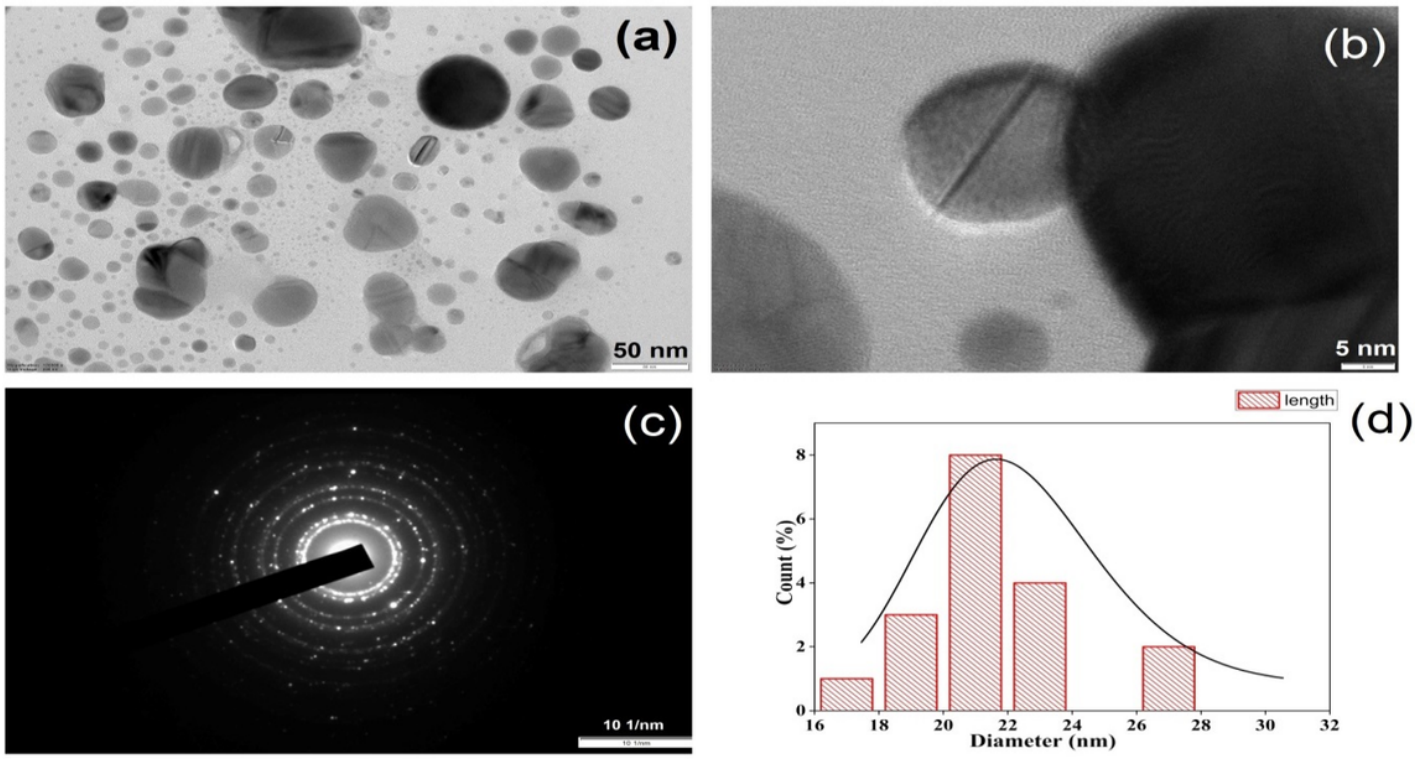

Figure 6

(a-d) HR-TEM images, SAED and size distribution histogram graph of RP-AgNPs

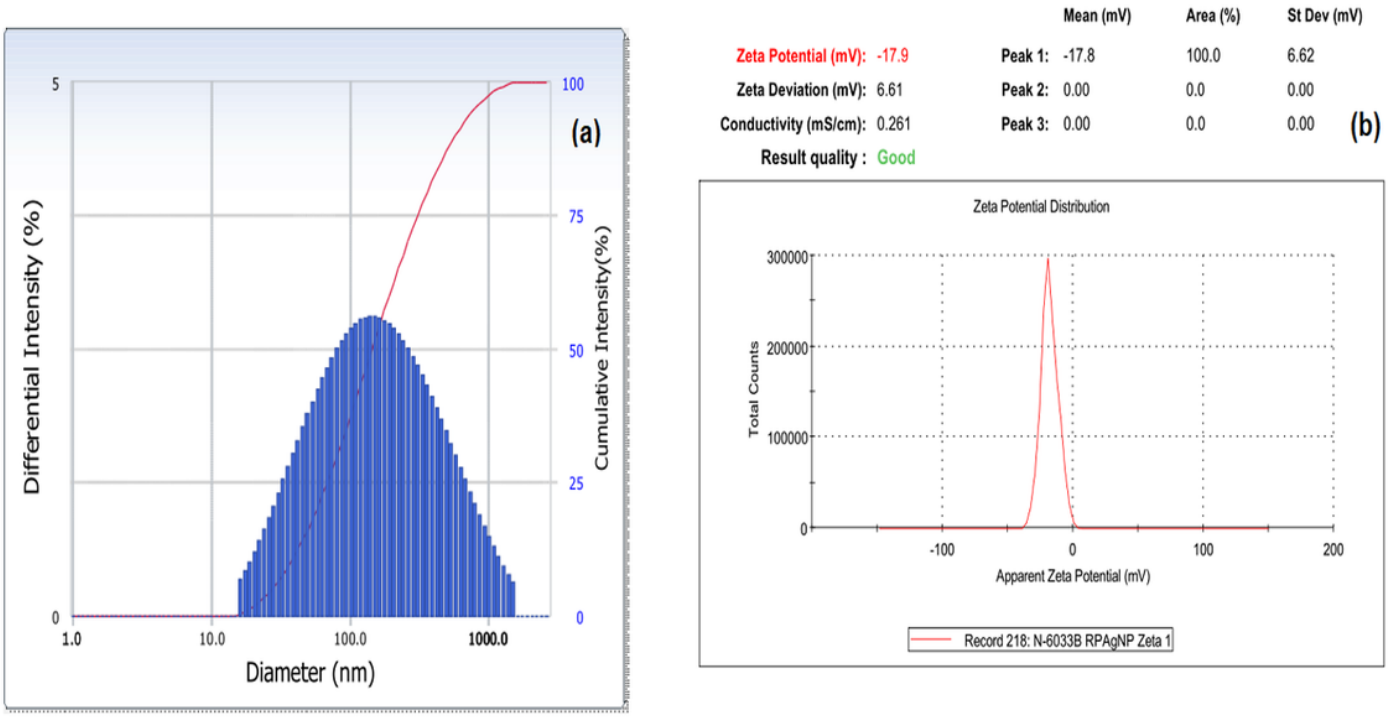

Figure 7

(a) hydrodynamic diameter (size) of RP-AgNPs, (b) the elector kinetic (Zeta) potential graphical representation of RP-AgNPs.

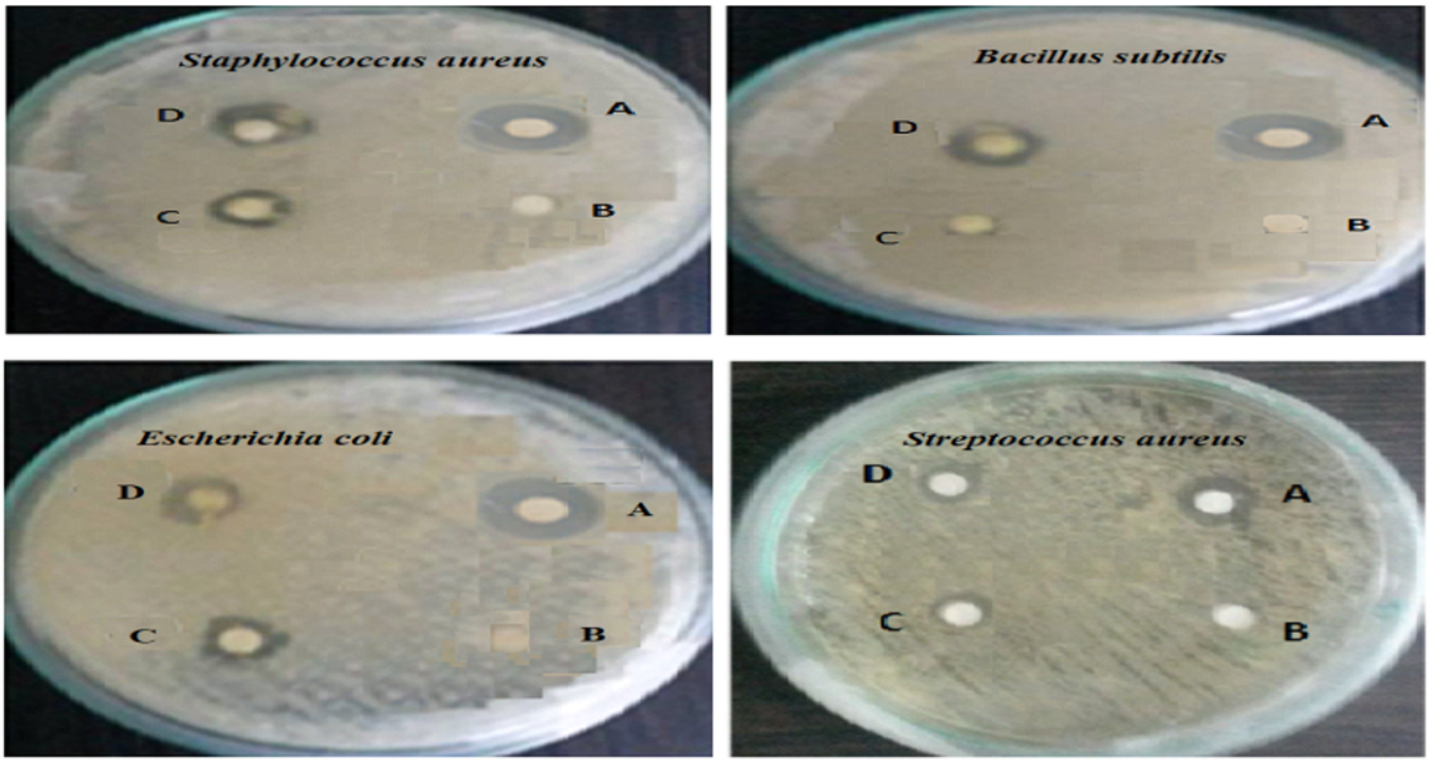

Figure 8

Inhibition zone representation of different bacterial samples treated with *R.pertusa* AgNPs

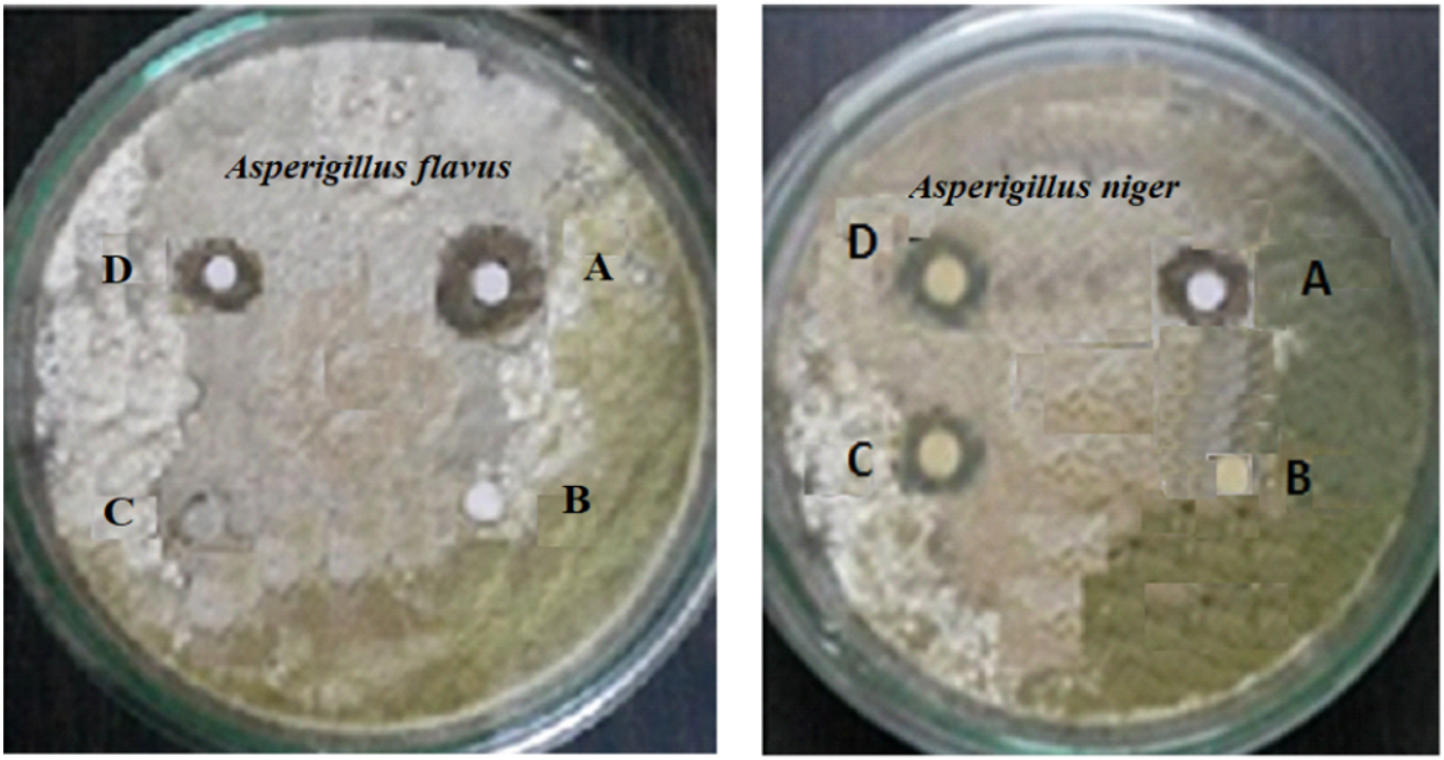

Figure 9

Inhibition zone of RP-AgNPs treated with *Aspergillus flavus* and *Aspergillus niger* by disc diffusion method

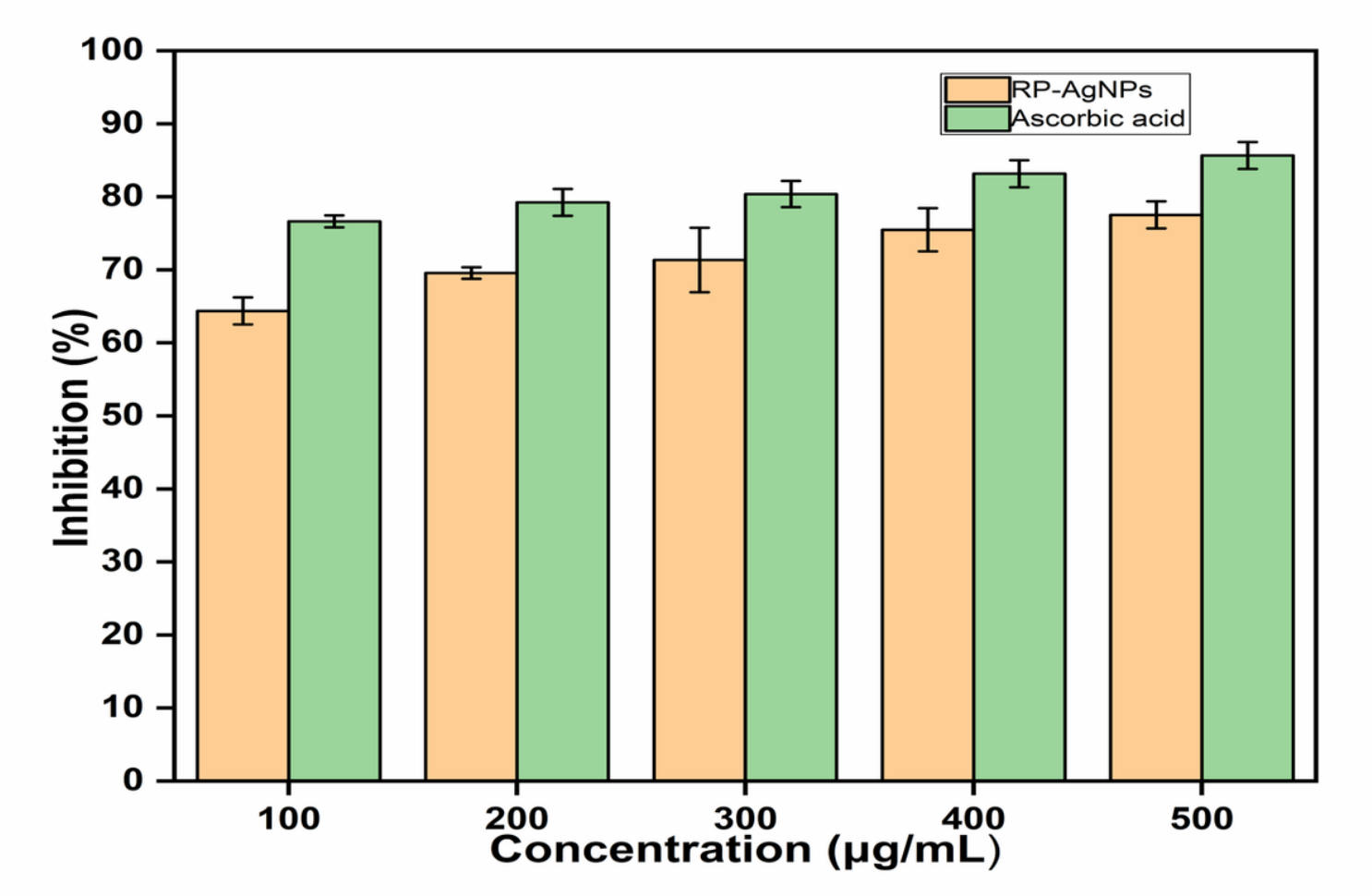

Figure 10

DPPH anti-oxidant analysis of bio-synthesized RP-AgNPs against Ascorbic acid.

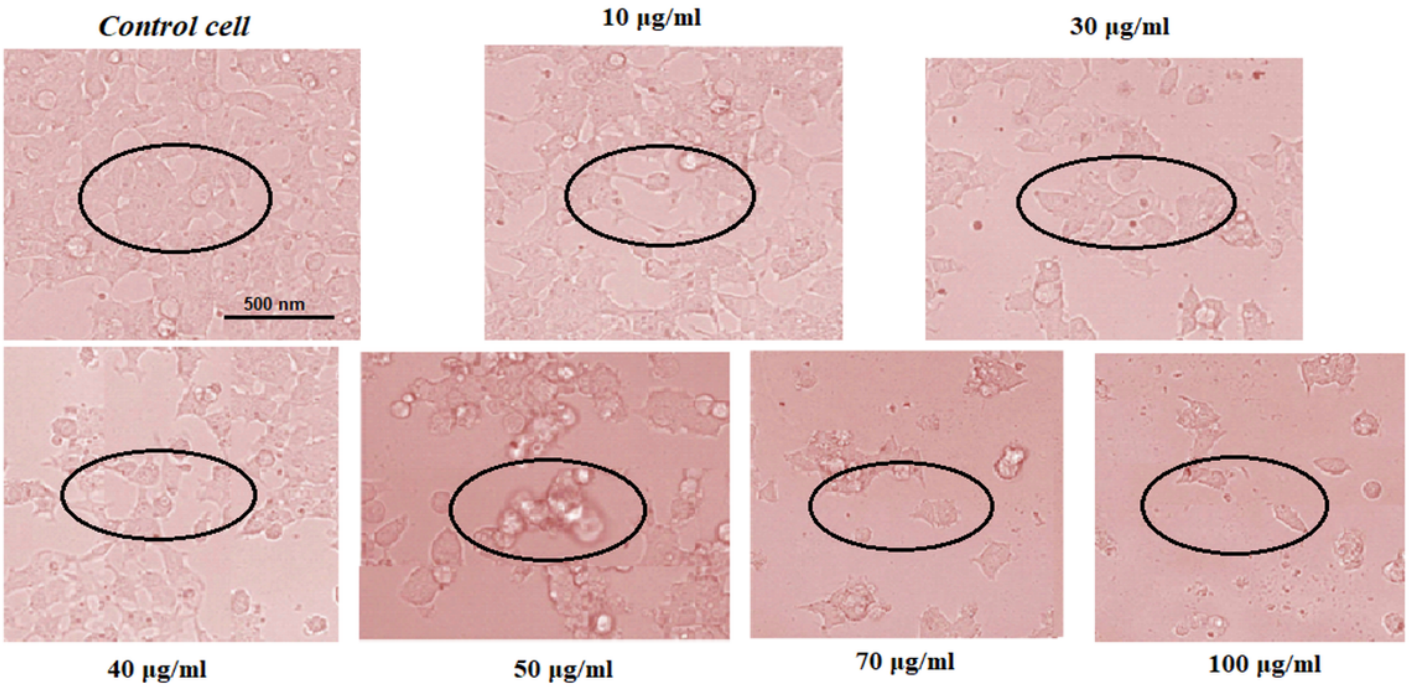

Figure 11

cytotoxicity analysis on MCF-7 exposed with different doses of bio-synthesized RP-AgNPs

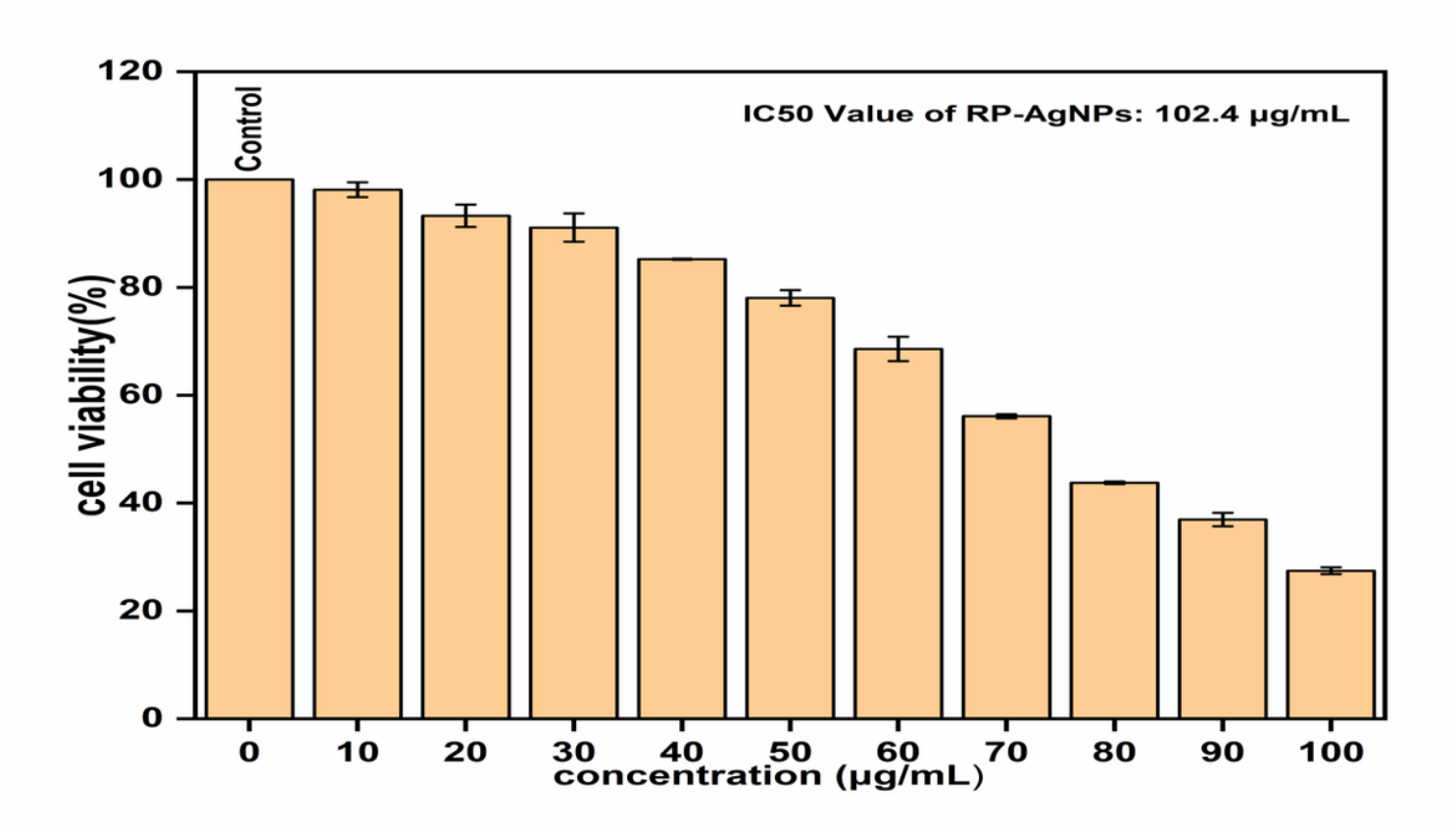

Figure 12

Cell viability analysis of MCF-7 cells treated with bio-synthesized RP-AgNPs.