PREPARATION IN- SILICO EVALUATION AND PHYTOPHARMACOLOGICAL SCREENING OF METAL NANO PARTICLES SYNTHESIZED FROM ADIANTUM CAPILLUS - VENERIS

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ABSTRACT:

To develop a novel approach for the biological synthesis of Gold silver and zinc (Trimetallic nanoparticles using) water extract of whole plant parts of Adiantum capillus veneris L. which has been proven to be active against antioxidant activity, Characterization of nanoparticles was determined by using Ultra Violet-Visible (UV-Vis) spectrometry, Scanning Electron Microscopy (SEM), Trasmision Electron Microscopy (TEM), X-ray Diffraction (XRD). Particle size detector Infra-Red Spectrometry, Zeta potential the synthesis of nanoparticles was confirmed by the change of colour pale green to Dark brown. Further, a peak between 426 nm to 601 nm wavelength was obtained on UV-VIS spectrometer which confirmed the biosynthesis of Trimetallic (Ag,Au,Zn) nanoparticles. And FT-IR shows quantitative analysis show the functional groups like O-H (alcohol/phenols) N-H (Amine) C-H (alkane) C=O (carbonyl). The particle size of prepared trimetallic nanoparticles was measured using a particle size analyzer and light scattering-based Laser diffraction. (HORIBA SZ100). The average particle size is 134.7 nm. Trimetallic nanoparticles (BNPs) have a zeta potential of –11.6 mv SEM showed the formation of Trimetallic nanoparticles (Ag,Au,Zn)with an average size of 160.4 nm. Transmission Electron Microscopy (TEM) images of the biosynthesized Trimetallic Au–Ag–Zn nanoparticles reveal well-dispersed, quasi-spherical particles with some degree of agglomeration. The nanoparticles exhibit a range of sizes, generally below 20 nm, as seen in the images captured at scales of 2 nm, 5 nm, and 10 nm. The high-resolution images display clear lattice fringes, indicating a crystalline nature of the particles.

The XRD pattern of the synthesized Au-Ag-Zn nanoparticles shows sharp peaks at 38.1°, 44.3°, 64.5°, and 77.4° (2θ), confirming the fcc structure of gold and silver. A broad hump between 10–30° indicates the presence of amorphous organic compounds from the plant extract. A. capillus-veneris can be good source for biological synthesis Trimetallic nanoparticles The antioxidant profile of compounds AU+Ag+Zn was evaluated by measuring the percent of inhibition against DPPH reagent. The compound AU+Ag+Zn exhibited good antioxidant activity against DPPH scavenging reagent and however the concentration increases also the antioxidant activity of the compound increases as compared to the standard ascorbic acid.

KEYWORDS: Nanoparticle, Adiantum capillus – veneris, Antioxidant.

INTRODUCTION:

The development of green processes for the synthesis of nanoparticles is evolving into an important branch of nanotechnology 1, 2. The field of nanotechnology is one of the most active areas of research in modern materials science. Nanoparticles exhibit completely novel and improved properties based on specific characteristics such as size, distribution and morphology. New application of nanoparticles and nanomaterials are emerging rapidly Nanotechnology involves tinkering work at atomic levels, tweaking and controlling substances 1, 00, 000 times smaller than a strand of human hair, to make useful materials and devices. It involves technology at the scale of one-billionth of a meter. The term „NANO" is derived from Greek word "Dwarf" 5. Nanotechnology is foreseen to significantly influence science, economy and everyday life in 21st century and it may become one of the driving forces to the next industrial revolution. Nanoparticles are being viewed as fundamental building blocks of nanotechnology 6. Nanoparticles are synthesized by reduction of heavy metals in the solutions of plant extract 7, thermal decomposition of silver gold and zinc compounds 8, microwave assisted synthesis 9, Soxhlet extraction method.

- Trimetallic nanoparticles (TNPs), consisting of three distinct metals, exhibit remarkable synergistic and multifunctional properties that surpass those of monometallic and bimetallic counterparts. These advanced features include improved catalytic efficiency, enhanced electrical and optical characteristics, superior thermal and magnetic behavior, and increased biological activity. As a result, TNPs have become highly attractive for diverse applications such as catalysis, energy storage, bio sensing, environmental remediation, and medicine.
- Growing interest in green synthesis methods has led to the development of eco-friendly approaches for TNP production, particularly plant-mediated synthesis. This approach makes use of plant extracts abundant in antioxidants, flavonoids, and various bioactive compounds that serve as natural agents for both reducing and stabilizing. This not only eliminates the use of toxic chemicals but also results in biocompatible nanoparticles. Among the most studied TNPs are combinations like Au–Ag–Pt, Au–Ag–Pd, and Pt–Ni–Co, known for their excellent catalytic and optical properties. In our recent study, we synthesized gold–zinc oxide–silver (Au–ZnO–Ag) TNPs using a green, plant-based approach. These nanoparticles showed promising multifunctional capabilities, including broad-spectrum antimicrobial effects, photocatalytic degradation of pollutants, and potential anticancer properties, making them strong candidates for both environmental and biomedical applications.

- **MATERIALS AND METHODS**:

Collection of Plant Material: The whole plant of Adiantum capillus-veneris was collected from the fields, at Ratnagiri district in foreset in rainy season. The samples of plants were identified self and binomially by Botanical Survey and voucher specimens were deposited in infinite biotech institute of research and analytics, sangli.

- **Preparation of the Extract:**
- **By Soxhlet method;**

An aqueous extract of *Adiantum capillus-veneris* was prepared by Soxhlet extraction of 500 g dried plant material using 95% ethanol. The extract was filtered, concentrated, dried, weighed, and stored in airtight containers.

- **By Microwave Assisted method:**

Microwave-Assisted Extraction (MAE) of *Adiantum capillus-veneris* involved treating 100 mg of plant powder with 10 ml ethanol in a sealed quartz tube. Extraction was performed at 20–160 W for 30 seconds to 5 minutes, with temperatures ranging from 10°C to 90°C.

- **Synthesis of Nanoparticles**:
- **Synthesis of Zinc nanoparticles (ZnNPs):**
- 1. 2.5mL of Plant extract was added to 25mL of Zinc acetate dihydrate solution. pH of the mixture was then adjusted to 8 by drop wise adding of NaoH. The reaction mixture was then stirred and Heated at 700C, for 30 minutes for complete reduction and formation of white precipitate. The resulting material was then collected via decantation, washed with distilled water to remove residuals and oven dried at 70 °C for overnight to yield powdered ZnO nanoparticles. The dried sample was then stored at room temperature in airtight container Yield: 0.5gm from 2.5gm of extract
- **Synthesis of Silver nanoparticles (Ag NPs):**

6ml of Silver nitrate (1M) was added in 40ml of extract and 154ml of deionized water. The reaction mixture was kept at room temperature and was observed for color change. Reaction mixture when showed reddish brown color it was centrifuged at 12000 rpm for 10 min. The pellet obtained was then washed with deionized water (3x) and then with acetone. Pellet is then dried and stored. Yield: 0.4gm from 2gm of extract.

- **Synthesis of Gold nanoparticles (Au NPs):**

4mLofthe extract was added to the 20 mL of gold chloride solution. The mixture was kept under continuous stirring on a magnetic stirrer at temperature of 750C. The reaction mixture was then observed for color change. Reaction mixture when showed reddish brown / purple color indicating the reduction of Au3+ to Au0. It was centrifuged at 12000 rpm for 10 min. The pellet obtained was then washed with deionized water (3x) and then with acetone. Pellet is then dried and stored. Yield: 0.6gm from 4gm of extract.

- **Synthesis of Trimetallic nanoparticles (BNPs)**
- Co-reduction is the simplest method of Trimetallic synthesis. It involves the mixing of three metal (Zn@Au@ Ag) precursors together. To that mixture, a reducing agent or stabilizing agent is added, which can reduce the metal ion to atoms and stabilize the aggregation process. The 5ml of Gold nanoparticles (AuNPs) (Liquid form) is mixed with 5ml Zinc Nanoparticles (ZnNPs) (Liquid form) and 5 ml of Silver nanoparcticals (AgNPs) sonicated the mixture with the help of sonicator at 370c. After that the 1.ml of PLA (Poly Glutamic Acid) were added to the nanoparticles mixture to stabilize particles. After sonication the mixture were heated at 100-2000C to get Trimetallic Nanoparticles

- **Characterization of Gold Silver and Silver(trimetallic) nanoparticles:**

- **UV-Visible Spectroscopy**

UV- visible spectroscopy is most widely used technique for investigation of the optical properties of the particles. The color changes indicated the formation of nanoparticles. The formation of Trimetallic (Zn@Au@Ag) nanoparticles using Ethanolic extract of *Adiantum capillus veneries* was monitored at the time of reaction with the help of UV-visible spectroscopy (Systronic ET2705) in the range 200-800nm and maximum absorbance was recorded. Operation was performed at Rajarambapu College of pharmacy, Kasegaon.

❖ **Fourier Transforms Infrared Spectroscopy (FTIR)[66]**

The synthesized Trimetallic (Zn@Au@Ag) nanoparticles and extract of *Adiantum capillus veneries subjected* to FTIR spectrometric analysis and spectra were recorded in the wave number frequency ranged from 4000 cm^{-1} to 600 cm^{-1} with a speed of 25 scan per spectrum by using JASCO FTIR-4600 equipped with ATR (Attenuated Total Reflectance) made in Japan. Operation will be performed at Rajarambapu College of pharmacy, Kasegaon. All measurements were recorded in Transmittance (%T) mode at room temperature.

❖ **Particle Size Analysis[67]:**

The majority of a nanoparticle's qualities depend on its size. In actuality, nanoparticles' novel features do not take hold until their size decreases to the nanometer range.The total area of the exposed surfaces of the particles per unit mass is the specific surface area of the particles. The connection between surface area and particle size is inverse. The specific surface area provides an average particle size if the particles are believed to be spherical and have a limited size distribution.

The sample of Trimetallic (Zn@Au@ Ag) nanoparticles was analyzed for Particle Size Analysis using the HORIBA SZ-100 at the Annasaheb Dange College of Pharmacy, Ashta.

- **Zeta Potential Measurement[68]:**

Zeta potential, which is connected to the surface charge of the nanoparticle. When the zeta potential of a nanoparticle is between 10 and +10 mV, it is said to be about neutral, however when it is larger than +30 mV or lower than 30 mV, it is said to be strongly cationic or anionic. Zeta potential can influence a nanoparticles propensity to penetrate membranes because the majority of biological membranes have a negative charge. Cationic particles typically exhibit more toxicity due to breakdown of cell walls.

The sample of Trimetallic (Zn@Au) nanoparticles was analyzed for zeta potential measurement using the HORIBA SZ-100 at the Annasaheb Dange College of Pharmacy, Ashta.

- **Scanning Electron Microscopy (SEM)[72]**
- SEM analysis was conducted to observe the morphology of gold (Au), silver (Ag), and zinc (Zn) nanoparticles synthesized using Adiantum capillus-veneris extract. The images, taken with a JEOL JSM-6360 microscope at 20 kV and magnifications of 5,000× and 15,000×, reveal that the nanoparticles exhibit varied shapes, including spherical, polygonal, and rod-like structures, depending on the synthesis parameters such as precursor concentration, pH, and reaction conditions. Some nanoparticles were evenly dispersed, while others aggregated or formed chains, highlighting the influence of the extract’s phytochemicals. These results confirm the successful biosynthesis of nanoparticles with tunable shapes and orientations.
- **Transmission Electron Microscopy (TEM)**

TEM analysis was conducted to examine the morphology and size distribution of gold (Au), silver (Ag), and zinc (Zn) nanoparticles synthesized using Adiantum capillus-veneris extract. Imaging was carried out with a JEOL JEM-1400 microscope at 200 kV, revealing nanoparticles in spherical, polygonal, and rod-like shapes, with sizes ranging between 5 and 50 nm. The TEM images showed that the nanoparticles were well-dispersed, although some formed loose aggregates or chains, indicating that the plant extract's phytochemicals played a role in nanoparticle formation and stabilization. These findings confirm that Adiantum capillus-veneris extract efficiently produces nanoparticles with well-controlled size, shape, and distribution.

- **X-Ray Diffraction (XRD) Analysis**

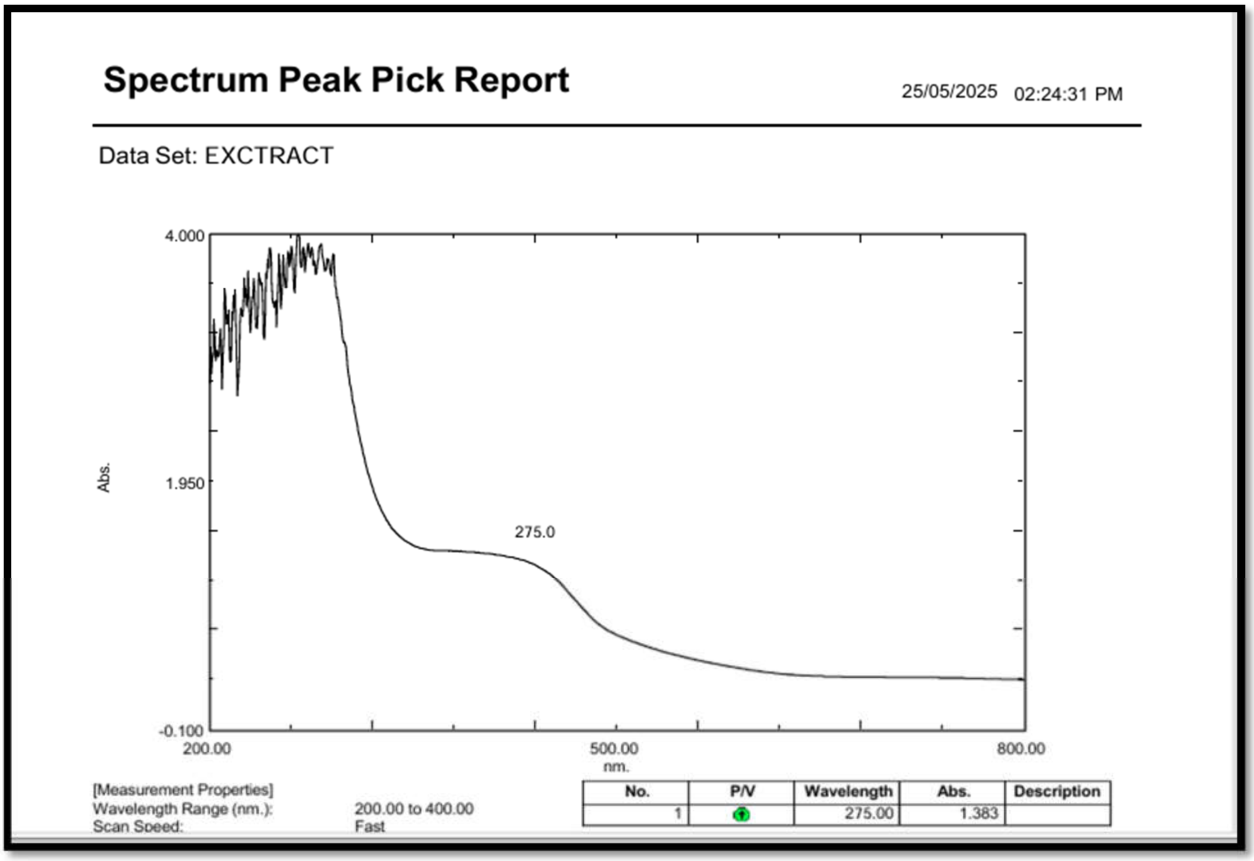
XRD analysis was performed to determine the crystalline nature and particle size of gold (Au), silver (Ag), and zinc (Zn) nanoparticles synthesized with Adiantum capillus-veneris extract. The sample was scanned using a Shimadzu XRD-6000 instrument within the angular range of 10° to 60° (2θ). Diffraction patterns showed distinct peaks, indicating the crystalline nature of the nanoparticles. The intensity and position of the peaks revealed phase information, confirming the formation of FCC structures for gold and silver, and either FCC or HCP for zinc. The particle size was estimated using the Scherer equation, confirming successful nanoparticle synthesis with controlled size and crystallinity.

- **Pharmacological Evaluation:**
- **Antioxidant Activity**

To evaluate antioxidant activity, plant extracts are prepared using methods like Soxhlet or maceration and concentrated using a rotary evaporator. These extracts are then dissolved in methanol or DMSO to make stock solutions. A 0.1 mM DPPH solution is freshly prepared in methanol and protected from light. Equal volumes of DPPH and sample solutions are mixed and incubated in the dark for 30 minutes. Absorbance is measured at 517 nm, and % inhibition is calculated to assess antioxidant capacity, with IC_{50} values determined from dose-response curves.

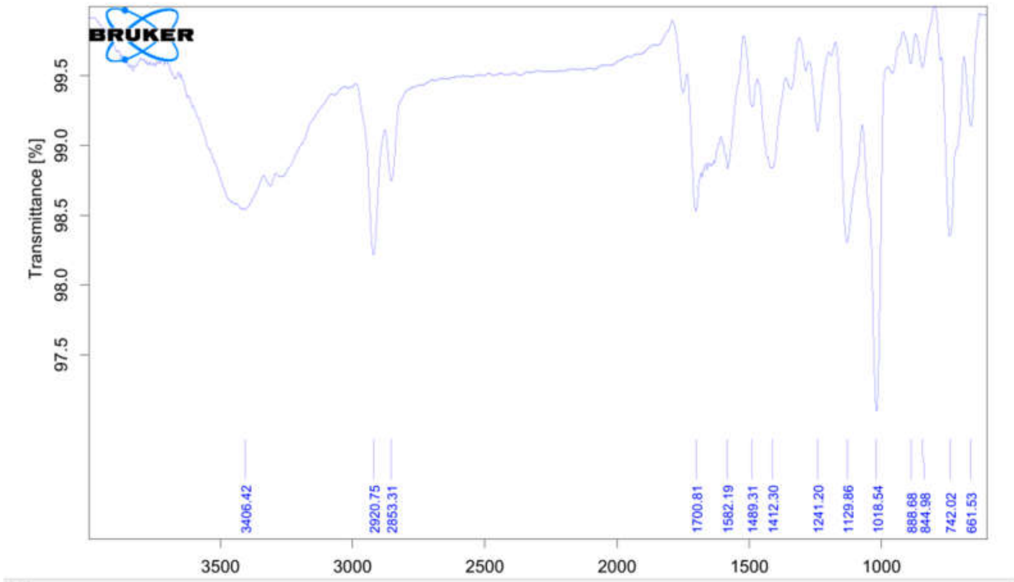
- **RESULTS:**
- **Results of identification properties of ethanolic extract of Adiantum capillus venries**

Figure 8.2 UV-visible spectra of Ethanolic extract of *Adiantum capillus venries*



The UV-Visible spectrum of *Adiantum capillus-veneris* extract shows a peak at 275 nm with an absorbance of 1.383, indicating the presence of phenolic compounds or flavonoids.

Figure 8.3 FT-IR analysis of Ethanolic extract of Adiantum capillus veneries



Functional Group	**Standard IR Frequency (cm⁻¹)**	**Observed Peak (cm⁻¹)**	**Assignment / Type**
O–H (hydroxyl, alcohols/phenols)	3200–3600 (broad)	3406.42	Stretching – Alcohols, phenols
C–H (alkanes, asymmetric)	2850–2960	2920.75	Stretching – Aliphatic C–H
C–H (alkanes, symmetric)	2850–2920	2853.31	Stretching – Alkanes
C=O (carbonyl)	1700–1750	1700.81	Stretching – Aldehydes, ketones
C=C (aromatic)	1600–1650	1628.19	Stretching – Aromatic ring

8.6 GC-MS analysis of Ethanolic extract of *Adiantum capillus veneries*

The GC-MS analysis of Ethanolic extract of *Adiantum capillus veneries* (Figure 8.3) showed GC fraction and fragmentation patterns of mass spectroscopy. The compounds present in the extract were identified by GC-MS analysis. The mass spectra of the phytoconstituents were compared with the library data of NIST and identified. Nine compounds in the extract were identified from extract.

Figure 8.3: GC-MS Chromatogram of Ethanolic extract of Adiantum capillus veneries

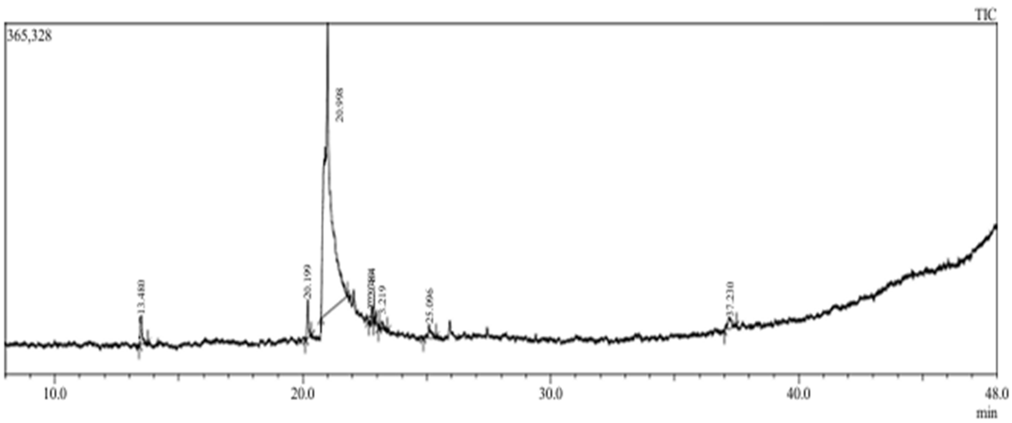


Table 8.5: Phytoconstituents identified in Ethanolic extract of *Adiantum capillus veneries*

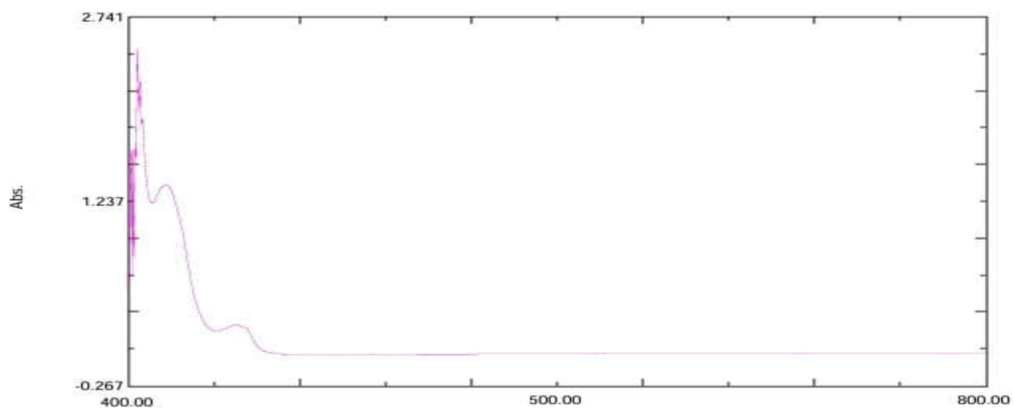
Peak #	Retention Time (min)	Compound Name	Area %	Molecular Formula	Molecular Weight (g/mol)
1	3.19	2,2-Dimethoxybutane	0.52	C6H14O2	118.17
2	13.48	(±)-Borneone	2.30	C10H16O	152.23
3	20.20	(39,22E)-Stigmasta-5,22-dien-3-ol	3.00	C29H48O	412.68
4	21.00	3',4',5,7-Pentahydroxyflavone (Luteolin)	28.91	C15H10O6	286.24
5	22.78	Naphthalene, decahydro-4a-methyl-1-methylene-	4.08	C11H18	150.26

Peak #	Retention Time (min)	Compound Name	Area %	Molecular Formula	Molecular Weight (g/mol)
6	22.91	Azulene, 1,2,3,3a,4,5,6,7-octahydro-1,4-dimethyl	4.62	C12H16	160.26
7	23.22	Bicyclo[3.3.0]octane, 2-methylene-5-(1-methylethyl)-	4.86	C12H20	164.29
8	25.01	(39,22E)-Stigmasta-5,22-dien-3-ol	3.06	C29H48O	412.68
9	37.23	11,14,17-Eicosatrienoic acid, methyl ester	100.00	C21H34O2	318.50

- **Synthesis of Gold, Silver and Zinc (Bimetallic) nanoparticles**:

Trimetallic Au-Ag-Zn nanoparticles were successfully synthesized using the aqueous extract of Adiantum capillus-veneris. A visible color change from pale yellow to dark brown indicated the formation of nanoparticles due to surface plasmon resonance. The phytochemicals in the plant extract acted as natural reducing and stabilizing agents for the metal ions. The reaction produced a stable colloidal solution without visible precipitation. This green synthesis approach offers an eco-friendly and cost-effective method for producing Trimetallic nanoparticles with potential applications in biomedical and catalytic fields. Further characterization (UV–Vis, FTIR, XRD, etc.) can confirm their structural and compositional features.

- **UV-Vis spectra analysis:**



UV-Visible spectra of Trimetallic (Zinc and Gold, Silver) Nanoparticles.

Figure 8.17: UV-visible absorption spectra of the synthesis Trimetallic nanoparticles

Table 8.13: Effect of on absorption intensity at 426,475 and 601 nm of Trimetallic nanoparticle.

Wavelength	Absorption
426	1.373
475	0.231
601	-0.004

displays the UV vis absorption spectra that were obtained in water from zinc, gold and silver Zn@ Au@ Ag NPs. A peak of excision can be observed 426, 475 and 601 nm. The peak in the dotted curve at 426-601 nm.

FT-IR analysis of Trimetallic nanoparticles:

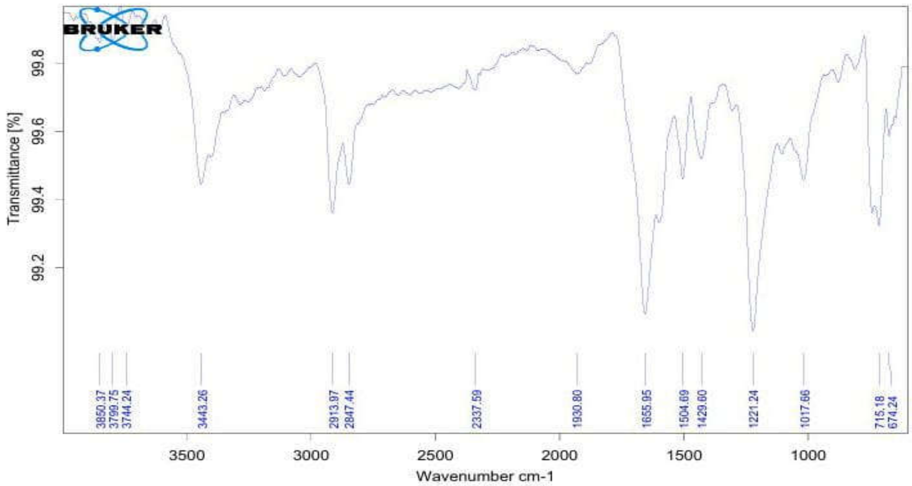


Figure 8.18: FT-IR analysis of Trimetallic nanoparticles

Table: FT-IR interpretation of Trimetallic nanoparticles

Functional Groups	IR Frequencies standard Peak (cm^{-1})	IR Frequencies observed Peak (cm^{-1})
O-H (alcohol/phenol)	3200-3600 (broad)	3380.27, 3741.24
N-H (amine/amide)	3300-3500 (sharp or broad)	3380.27
C-H (alkane)	2850-2960	2930.97, 2871.44
C=C-H (alkene/aromatic)	~3100-3020	3143.26
C≡N or C≡C (triple bond)	2100-2260	2100.00, 2337.59
C=O (carbonyl group)	1650-1750	1655.56
C=C (alkene/aromatic ring)	1600-1680	1546.69, 1450.60

C-O (ether/ester)	1050-1300	1221.24
C-N (amide)	-1020-1350	1017.66
Aromatic C-H bending	675-900	715.16, 674.24

8.23: Particle Size Analysis of Trimetallic nanoparticles

Calculation Results

Peak No.	S.P.Area Ratio	Mean	S. D.	Mode
1	1.00	192.9 nm	50.7 nm	181.6 nm
2	---	--- nm	--- nm	--- nm
3	---	--- nm	--- nm	--- nm
Total	1.00	192.9 nm	50.7 nm	181.6 nm

Cumulant Operations

Z-Average : 134.7 nm

PI : 0.435

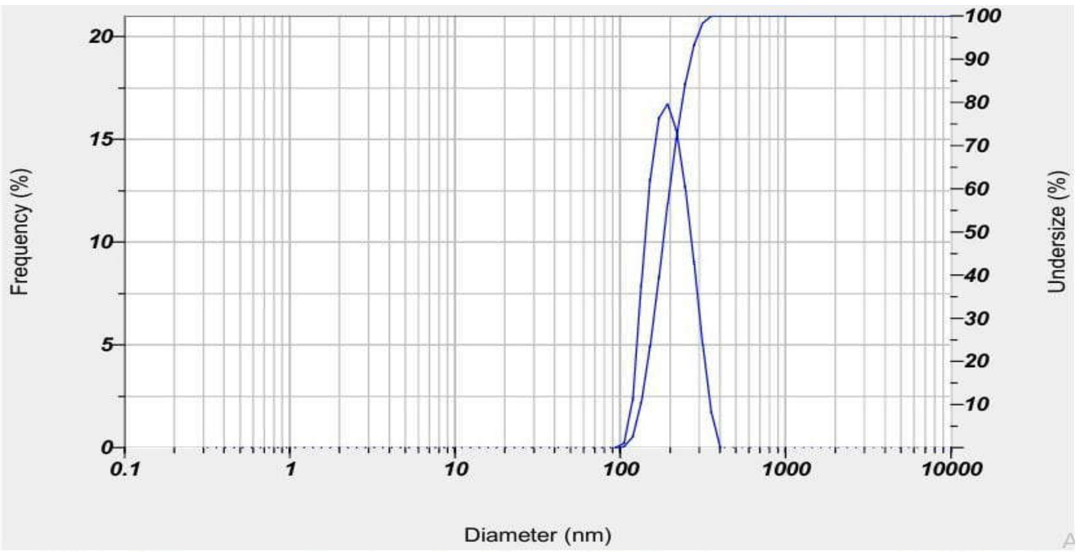


Figure 8.19: Particle Size Analysis of Trimetallic nanoparticles

The particle size of prepared Trimetallic nanoparticles was measured using a particle size analyzer and light scattering-based Laser diffraction. (HORIBA SZ100). **(figure 8.19)** The average particle size is **134.7 nm.**

8.24 Zeta Potential analysis of Trimetallic Nanoparticles:

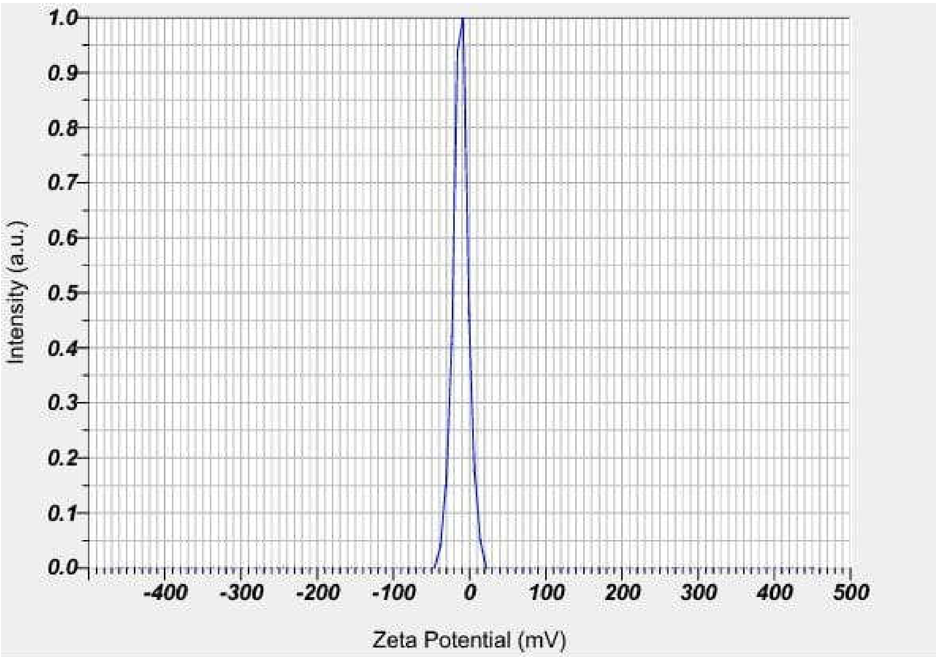
The formulated Trimetallic nanoparticles (BNPs) have a zeta potential of –11.6 mv (figure 8.20).

Calculation Results

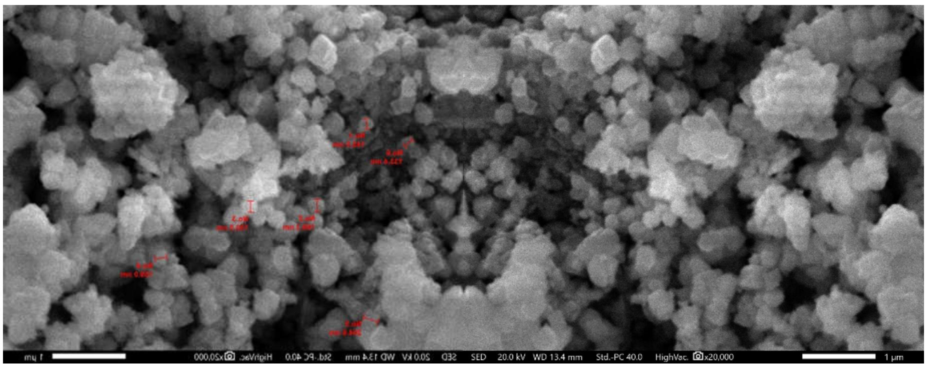
Peak No.	Zeta Potential	Electrophoretic Mobility
1	-11.6 mV	-0.000090 cm2/Vs
2	--- mV	--- cm2/Vs
3	--- mV	--- cm2/Vs

Zeta Potential (Mean) : -11.6 mV
Electrophoretic Mobility Mean : -0.000090 cm^2/Vs

Figure 8.20: Zeta Potential Graph of Trimetallic nanoparticles (BNPs)



8.26 Scanning Electron Microscopy (SEM) of nanoparticles:



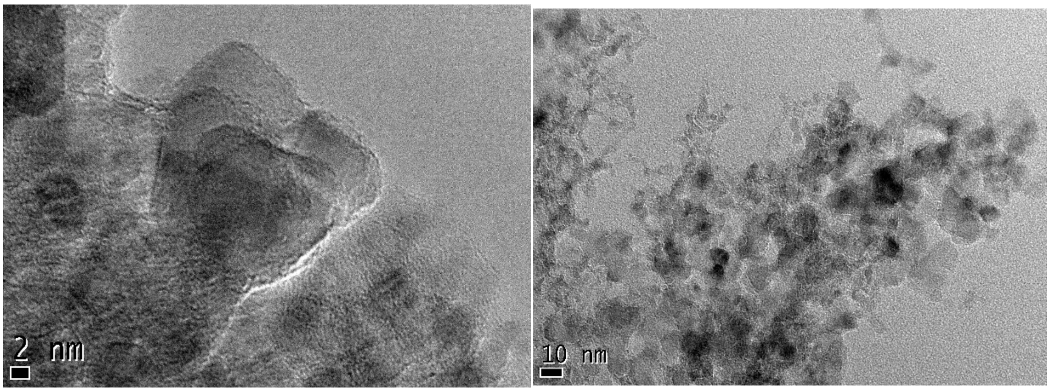
8.27 Scanning Electron Microscopy (SEM) analysis of nanoparticles:

The scanning electron microscopy (SEM) image of Trimetallic nanoparticles synthesized using *Adiantum capillus-veneris* extract reveals a densely packed, aggregated structure composed of predominantly spherical to irregularly shaped nanoparticles. The particle sizes, measured at various points, range from approximately 133.4 nm to 204.6 nm, with an average size of about 160.4 nm. The surface texture appears rough and the particles are moderately uniform in size, indicating effective biological synthesis possibly driven by phytochemicals in the plant extract. The nanoparticles were visualized at a magnification of 20,000× using a 20.0 kV accelerating voltage under high vacuum conditions with a working distance of 13.4 mm. These conditions enabled clear imaging and accurate size estimation of the nanoparticle clusters.

Conclusion:

The SEM analysis confirms the successful biosynthesis of Au–Ag–Zn Trimetallic nanoparticles with average particle sizes in the nanoscale range. The morphological characteristics and moderate size uniformity suggest that bioactive compounds in the plant extract played a significant role in both reducing and stabilizing the metal ions during nanoparticle formation.

8.26 Tramission Electron Microscopy (TEM) of nanoparticals(BNPs) :

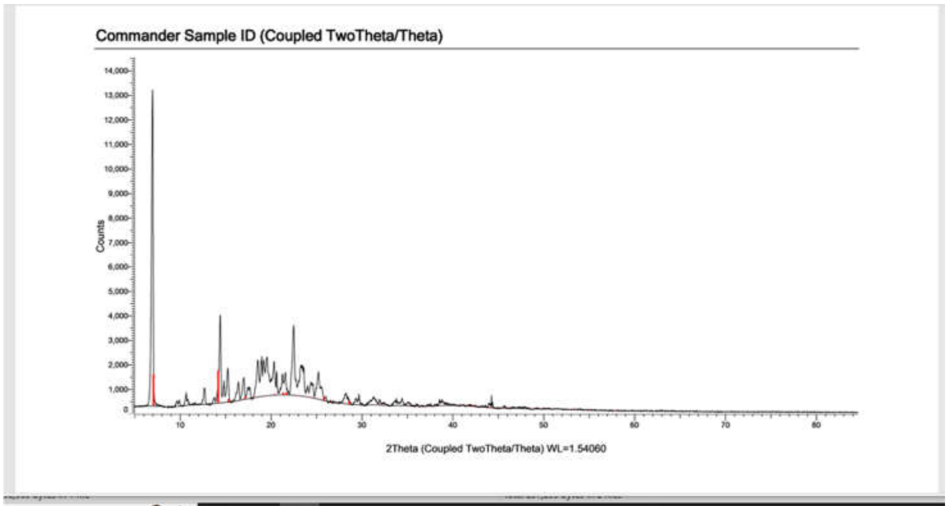


- **Analysis of Transmission Electron Microscopy (TEM):**

Images of the biosynthesized Trimetallic Au–Ag–Zn nanoparticles reveal well-dispersed, quasi-spherical particles with some degree of agglomeration. The nanoparticles exhibit a range of sizes, generally below 20 nm, as seen in the images captured at scales of 2 nm, 5 nm, and 10 nm. The high-resolution images display clear lattice fringes, indicating a crystalline nature of the particles. A core-shell-like structure can be inferred in some particles, possibly due to the multi-metallic composition, suggesting successful integration of Au, Ag, and Zn within single nanoparticles. The particles appear embedded in a less dense organic matrix, likely originating from phytochemicals in *Adiantum capillus-veneris* extract, which act as natural capping and reducing agents.

Conclusion: The TEM study confirms the nanoscale size, crystalline nature, and successful Trimetallic synthesis of Au–Ag–Zn nanoparticles using *Adiantum capillus-veneris* extract. The presence of well-defined particle boundaries and lattice fringes reflects high crystallinity, while the uniform distribution and core-shell features imply effective green synthesis and structural stability.

The X-ray diffraction (XRD Results:



The X-ray diffraction (XRD) analysis:

The synthesized Trimetallic Au-Ag-Zn nanoparticles using *Adiantum capillus-veneris* extract reveals a crystalline nature, as indicated by the presence of sharp and well-defined peaks. The major diffraction peaks observed at approximately 38.1°, 44.3°, 64.5°, and 77.4° (2θ) correspond to the (111), (200), (220), and (311) planes, respectively, of face-centered cubic (fcc) structures of gold and silver. These peaks confirm the successful formation of Au and Ag nanoparticles. Additionally, a broad hump in the 10–30° range suggests the presence of amorphous organic compounds likely originating from the plant extract used in the green synthesis process. While zinc-related peaks are not distinctly resolved, it is possible that Zn is either alloyed within the Au-Ag matrix or present in oxide form with low crystallinity, resulting in weaker or overlapping signals. The low-angle peak around 7.5° might be attributed to bioorganic compounds or capping agents, contributing to the stabilization and dispersion of the nanoparticles. The diffraction pattern supports the successful biosynthesis of crystalline Trimetallic nanoparticles with potential partial amorphous content from plant-derived capping agents.

Table: XRD Peak Assignment Summary

2θ (Degrees)	Assigned Plane (hkl)	Probable Phase	Interpretation
7.5°	–	Organic/Bio Matrix	Indicates organic capping/stabilizing agents
10–30°	–	Amorphous/Organics	Broad peaks suggest organic or amorphous content
38.1°	(111)	Au/Ag (fcc)	Primary peak indicating crystalline Au/Ag
44.3°	(200)	Au/Ag (fcc)	Confirms fcc structure of Au/Ag
64.5°	(220)	Au/Ag (fcc)	Crystalline phase continuation
77.4°	(311)	Au/Ag (fcc)	Supports presence of nanocrystalline metals

8.26.2: Antioxidant Activity:

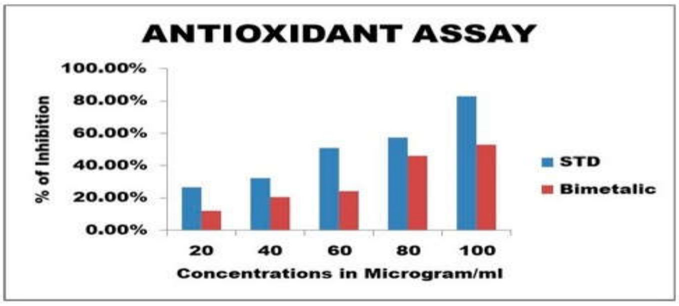
The antioxidant profile of compounds AU+Ag+Zn was evaluated by measuring the percent of inhibition against DPPH reagent. The compound AU+Ag+Zn exhibited good antioxidant activity against DPPH scavenging reagent and however the concentration increases also the antioxidant activity of the compound increases as compared to the standard ascorbic acid.

Table no.8.17: Antioxidant Activity of Trimetallic nanoparticles (BNPS)

SR.NO	Sample Code	Concentration (µg/ml)	Absorbance at 510nm				% Inhibition	IC50 (µg/ml)
			Test 1	Test 2	Test 3	Mean		
1	Control	-	1.93	1.93	1.93	1.93	-	
2	Standard	20	1.45	1.42	1.40	1.42	26.42%	57.67
	(Ascorbic Acid)	40	1.31	1.29	1.33	1.31	32.12%	
		60	0.95	0.97	0.93	0.95	50.77%	
		80	0.82	0.85	0.79	0.82	57.51%	
		100	0.35	0.32	0.32	0.33	82.90%	
3	AU+Ag+Zn	20	1.71	1.68	1.73	1.70	11.91%	97.16
		40	1.54	1.55	1.51	1.53	20.72%	
		60	1.49	1.46	1.45	1.46	24.35%	
		80	1.02	1.07	1.04	1.04	46.11%	
		100	0.90	0.94	0.91	0.91	52.84%	

8.26.3 Anti-oxidant activity by graphical representation by DPPH assay x

Graphical data:



Images of the activity:

DPPH Method Assay:

DPPH Free Radical Scavenging Activity Overview:

The DPPH (2,2-diphenyl-1-picrylhydrazyl) assay is a method used to evaluate the antioxidant capacity of compounds based on their ability to donate hydrogen and neutralize free radicals. The scavenging ability is expressed as % inhibition, and the IC_{50} value (the concentration needed to inhibit 50% of the DPPH radicals) is used to compare antioxidant strength—lower IC_{50} means higher antioxidant activity. IC_{50}: 97.16 µg/ml

Interpretation: The AU+Ag+Zn combination shows moderate antioxidant activity but is less effective than ascorbic acid (higher IC_{50} and lower % inhibition at each concentration).

Conclusion:

Ascorbic Acid (Standard): Strong antioxidant activity (IC_{50} = 57.67 µg/ml)

AU+Ag+Zn Sample: Moderate antioxidant activity (IC_{50} = 97.16 µg/ml)

The data suggests that while the metallic nanoparticle blend (AU+Ag+Zn) does have antioxidant properties, it is less potent compared to the standard antioxidant, ascorbic acid.

Discussion

Ayurveda, the ancient Indian medical system, is a well-known example of a traditional medical system that uses herbal remedies to treat and/or prevent a variety of human illnesses. The majority of the basic materials used in ayurvedic medicine came from plant sources. Green plants produce and store a wide range of biochemical products, many of which can be extracted and employed as chemical feedstock or as the starting point for different types of scientific research. According to the results of the phytochemical analysis of the plant extract, the plant material contains alkaloids, tannins, carbohydrates, and glycosides.

Adiantum capillus-veneris, commonly known as the Maidenhair Fern, are home to the genus: petridaceae is a delicate plant found in tropical and subtropical areas, particularly in damp, calcareous, and rocky habitats up to 2400 meters in elevation. It flourishes in warm, moist environments, especially during the rainy season, growing up to 30 cm tall. The fern multiplies through both vegetative means and spore production, with spores dispersed by wind and water. It is characterized by finely divided, lacy fronds, dark shiny stalks, and creeping rhizomes that help in its spread. Traditionally, it holds medicinal value in Ayurveda and other healing systems, used to treat respiratory issues, skin problems, and inflammation due to its antibacterial, anti-inflammatory, and antioxidant properties.

Trimetallic nanoparticles, and representative nanoparticle micrographs revealed that the particles are irregular, clumbs spherical clusters, flake like area and patchy granules. The organic compounds in the extract, which are in charge with the zinc and gold, Silver reduction and stabilizing the thereby produced nanoparticles, are what cause the peaks to be seen. To determine the primary chemical groups in Adiantum *capillus veneries* extract and their potential role in the creation and stability of Trimetallic nanoparticles, a zeta potential measurement was conducted. Resistance to antibiotics has become a significant public health issue. On a range of bacteria, the metallic Trimetallic nanoparticles had antimicrobial and antioxidant action, and they prevented infections from developing resistance. The data suggests that while the metallic nanoparticle blend (AU+Ag+Zn) does have antioxidant properties, it is less potent compared to the standard antioxidant, ascorbic acid

The usefulness of this work shows the wide range of uses for formulated nanoparticles.

SUMMARY:

The macroscopically features and proximate chemical analysis of the Adiantum capillus veneries plant material were assessed. The results of the phytochemical screening revealed the presence of Alkaloids glycosides Sapponies Flavonoids Terpenoids Tannin Phenolic compound. GC-MS, FT-IR, and UV analysis were used to prepare the extract using the continuous heat extraction method and the microwave aided extraction method. An extract from Adiantum capillus veneries was used in the synthesis of Trimetallic (zinc,gold and Silver) nanoparticles. Studies were conducted on the effects of concentration, incubation period, and pH on the synthesis of nanoparticles. UV, FTIR, and SEM were used to further analyze the produced Trimetallic (zinc,gold and silver) nanoparticles. The wavelength at which the highest absorption of synthesized Trimetallic nanoparticles (BNPs) was observed was 426 and 475 nm.and 601 nm. The Trimetallic nanoparticles' value shifting is revealed by FTIR measurement, indicating that the nanoparticles were synthesized. The nanoparticle forms are displayed via SEM. Additionally, Trimetallic nanoparticles were employed for their antibacterial and antioxidant properties Hela DPPH Assay Ascorbic acid is used for antioxidant activity and gram-positive and gram-negative bacteria are used for antibacterial activity.

CONCLUSION

From the thesis preparation in- silico evaluation and phytopharmacological screening of metal nano particles synthesized from adiantum capillus - veneris”

We conclude that-

An examination of crude medicines using a potential approach verified the extract. Soxhlet extraction and microwave assisted extraction (MEA) both produced successful extracts novel chemical components found in the ethanolic extract by GC-MS analysis. Heart Flavonoids are a key element of ethanolic extract. Adiantum capillus veneries extract is used in the environmentally friendly manufacture of Trimetallic (zinc gold and silver) nanoparticles. Subsequently, the antibacterial activity of these nanoparticles were assessed and they performed better than the extract. Trimetallic (zinc gold and silver) nanoparticles from Adiantum capillus veneries exhibited moderate efficacy against the DPPH Assay Ascorbic acid at different concentrations. This work shows that artificially produced nanoparticles can be used in a wide variety of applications.

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