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Biosynthesis of Iron and Silver Nanoparticles Using Extract of Water Hyacinth (*Eichhornia crassipes*) for Antimicrobial and Antioxidant

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ABSTRACT

The biosynthesis of the iron and silver nanoparticles was extracted for antimicrobial and antioxidant activities in this research, Simple random sampling technique was employed to collect the plant samples within Adamawa metropolis. The collected samples were washed with 5% (v/v) nitric acid (HNO_3) and then rinsed with deionize water to remove the possible contaminant. The color of the reaction changes as the temperature increases 25°C to 75°C from yellow to brown, when 75 cm^3 of 1mM AgNO_3 solution extract was added to 25 cm^3 of the plant extract on stirring. The yellow-brown color was obtained from the formation of AgNPs. The color of the reaction changes as the temperature increases 25°C to 75°C from light red to brick red, when 75 cm^3 of 1mM Fe_3O_4 solution extract was added to 75 cm^3 of the plant extract on stirring. The fine brick red appears immediately after introducing the Fe_3O_4 Solution which shown the formation FeNPs. The absorbance increase has been associated with a higher concentration of metallic Fe, while the displacement of the maximum peak towards longer wavelengths is mainly owing to the increase in particle size. DPPH radical-scavenging activity (%) of EC 25/75 Nano was increasing as the concentration increases 10, 20, 30, 40 and 50 $\mu\text{l/ml}$ with 24, 39, 47 55 and 73 % respectively. (EC 50/50 Nano), 31, 32, 39, 47 and 63%, EC antimicrobial minimum inhibition concentration of EC25/75 AgNPs for *Klebsiella pneumoniae* and *S. aureus* showed that the extract had high inhibition on *Klebsiella pneumoniae* compared to *S. aureus*. Similar trends subsist for all the five-microorganism tested. The zones of inhibition increase with increasing concentration of the synthesized material concentration. EC silver 25/75 Antimicrobial has a maximum inhibition zone which I higher as compared to EC silver 75/25 Antimicrobial. The trend of Inhibition shows close inhibition for all the pathogens except *K. pneumonide* which has the least inhibition for all the 50/50 EC iron. The inhibition sequence shows close inhibition for all the pathogens except *S.typhi* and *K. pneumonide* which has the least inhibition for all the EC Fe. Inhibition is maximum at 20mm and 5mm minimum. Concentration of the material increase results in higher inhibition for four organisms while *k. pneumonide* shows a different trend with highest inhibition for 200 $\mu\text{g/l}$ concentration.

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1.0 Introduction

Nanoparticles is any microscopic particle mostly less than 100 nanometers (nm). Nanobiotechnology is an important emerging field of nanotechnology, which has been seriously used for different diverse applications in all the field of sciences.

Biosynthesis of different nanoparticles exhibits enormous growth in popularity from the beginning of twenty first century [1].

Silver nanoparticles (AgNPs) have potential applications in various fields such as bio-medicinal, catalysis, environment, and antimicrobial/ antibacterial activity. Compared to conventional method of heating, Microwave irradiation (non-traditional method) is known to have a faster heating rate. Due to the several disadvantages associated with the use of toxic chemicals and solvents for the synthesis of silver nanoparticles, a systematic biosynthesis method is adopted so as to a relatively inexpensive, biocompatible, eco-friendly and size-controlled nanoparticles.

Recently, environmentally friendly materials such as plant leaf extract, fruits, bacteria, and fungi are utilized for the synthesis of silver nanoparticles [2]

The synthesis of magnetic nanoparticles has gained significant attention due to their unique properties and potential applications in various fields. Silver and Magnetite (Fe_3O_4) nanoparticles, in particular, have demonstrated excellent magnetic properties and biocompatibility. In recent years, there has been a growing interest in developing environmentally friendly and cost-effective synthesis methods for magnetite nanoparticles.

Eichhornia crassipes, commonly known as Water hyacinth, belonging to the family of *Pontederiaceae*, is an aquatic plant. It has been listed as one of the worst weeds mostly in the tropics and subtropics and listed as Invasive Alien Species (IAS) in Sri Lanka.

Eichhornia crassipes is a wild freshwater fern. This plant is native to South America but has been naturalized in many tropical and subtropical regions of the world. Water hyacinth is considered the world's most rapidly growing and spreading aquatic plant with congested growth, which is also a source of biomass. Many efforts had been made to eradicate this species, but *Eichhornia* still shows

wide distribution, posing a tremendous threat to aquatic biota in many inland water systems.

Water hyacinth (*Eichhornia crassipes*) is a free-floating aquatic plant known for its abundant phenolic compounds and high antioxidant activity. Several studies have reported the potential of water hyacinth extract in synthesizing metal nanoparticles due to its reducing and capping properties. For instance [3] demonstrated the successful synthesis of silver using water hyacinth extract, attributing its reducing capability to the presence of phenolic compounds.

Biosynthesis (or green synthesis) is more ecofriendly and cost effective with their chemical stability, excellent biocompatibility, easy fabrication process, convenient surface bio-conjugation with molecular probes, excellent optical properties related to surface plasmon resonance and low toxicity. The term pesticide means "pest-killer". It is a substance or mixture of substances used for controlling, preventing, destroying or reducing the number of pests. Pests are one of major problems that agriculture faces. They are defined as being any type of organism that is harmful, destructive, or annoying to humans and plants. They can destroy crops and cause diseases to humans, animals, or plants [1].

The most common pests are insects, unwanted plants, microorganisms such as fungi or bacteria, and rodents. Organic pesticides are major sources of soil pollution in agricultural lands and rivers. Most of these pesticides are persistent and tend to bio accumulate in humans upon consumption of contaminated plants.

Recently, nanotechnology has revolutionized several scientific areas in search of alternatives to improve the human living conditions such as health field, social environment, and environmental. Scientists have generated nanometer-scale materials with very specific properties for targeted applications. In the health field, silver nanomaterials have been studied and manufactured by different synthesis methods according to their specific application. Although pure and well-defined nanoparticles can be produced with chemical and physical methods, these methodologies are expensive and

potentially dangerous to the environment because of the chemical reagents used.

On the other hand, the use of biological systems such as microorganisms, plant extracts, or plant biomass could be an alternative to produce nanoparticles in an ecological way compared with chemical and physical methods. Based on different chemically synthesized, iron nanoparticles that have been obtained by the biogenic routes are free from toxic contamination with by-products. On the other hand, biological synthesis of gold nanoparticles has advantages such as simple, single-step, cost-effective, environmentally friendly, and biocompatible nature of synthesized iron nanoparticles. The biocompatibility is essential for biomedical and biosensing applications. A shorter time is required in order to achieve biosynthesis of gold nanoparticles than the chemical synthesis. Other than that, it reduces the number of steps with the attachment of functional groups to nanoparticles surface in order to make them biologically active [4].

Silver nanoparticles are intensively explored nanostructures ranging between 1 and 100nm, primarily used for unconventional and enhanced biomedical applications in such areas as drug delivery, wound dressings, tissue scaffolding and protective coating applications. Moreover, the impressive available surface of Nano silver allows the coordination of many ligands, thus enabling tremendous possibilities with respect to the surface functionalization of silver nanoparticles. Silver is routinely used in the form of silver nitrate (NO_3^-) for antimicrobial activity. In addition, silver nanoparticles are more beneficial as compared to free silver because their greater surface area increases the exposure of microbes. Furthermore, silver nanoparticles have emerged as a great field of interest for researchers because of their unique activity against a large range of microorganisms and due to resistance against commonly used antibiotics.

To date, several studies have reported applications in fields such as food processing, agriculture and agro-based industries, biomedical and medical remediation, healthcare products, consumer products, numerous industries, pharmaceuticals, in diagnostics, orthopedics, drug delivery, imaging, filters as antitumor agents and as

enhancer of tumour-killing effects of anticancer drugs. organophosphate pesticides are compounds that contain phosphorous atom doubly bonded to either oxygen or sulphur [5].

The environmental and health issues posed by organophosphate and organochlorine pesticides, and the need for an effective solution through the use of biosynthesized iron and silver nanoparticles. The accumulation of these pesticides in water bodies can lead to contamination of water resources and disruption of aquatic ecosystems. contamination can have far-reaching consequences on biodiversity and ecosystem balance.

Green synthesis is an excellent tool that can be utilized for synthesis of AgNP as it uses natural origin medicinal herbs and its extracts which contain wide range of metabolites specifically water-soluble flavones, quinones causes rapid and quick reduction of silver when compared to fungi and microbes. Green chemistry approach is safe, cost efficient, easily scalable to mass productions, easily availability of raw materials at cheaper cost. Phytochemicals directly take part in reduction process of the silver ions a during synthesis of AgNP [6].

Silver nano particles are one of the most attractive materials for commercialization applications. As antibacterial agents silver nanoparticles were used for wide range of applications from disinfecting medical devices and home appliances to water treatment. AgNPs promisingly used in drastic fields such as healthcare products, food storage, textile and medicinal devices. In antibacterial potential AgNPs free silver ions are released at slower rate along with higher surface area which produces noxious environment and this is the main reason for broad spectrum antibacterial potential of AgNPs [7].

The concentration of water hyacinth extract plays a crucial role in the synthesis of magnetite AgNPs and Fe_3O_4 nanoparticles. High concentrations of the extract can result in faster reduction kinetics and higher yield of nanoparticles (López-Millán *et al.*, 2019) . On the other hand, low concentrations may lead to slower reduction and smaller particle sizes. The synthesis of Fe_3O_4 and AgNPs nanoparticles involves the reduction of iron salts in the presence of water hyacinth extract. The choice of precursor and concentration of extract play a

crucial role in determining the size, shape, and magnetic and silver properties of the synthesized nanoparticles. For example [8] demonstrated that using ferric chloride as a precursor and optimizing the extract concentration resulted in the formation of uniform and well-dispersed nanoparticles.

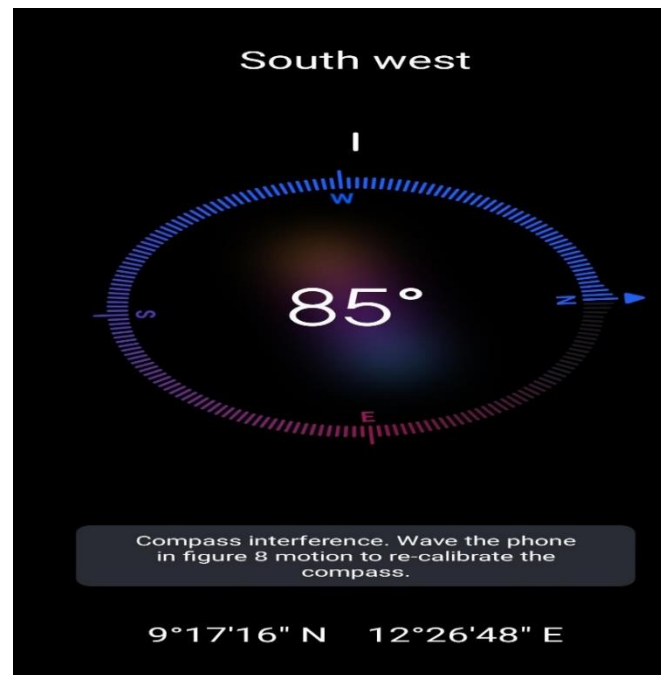
2.0 Materials and Methods

• 2.1 Sampling location

Water Hyacinth (*Eichhornia crassipes*) was obtained in Hayen Gada, Mubi road, Girei local government area of Adamawa state.

2.2 Sample techniques

Simple random sampling technique was employed to collect the plant samples within Adamawa metropolis. The samples were collected manually using gloved hands and placed in polythene bags, then transported to the laboratory for further preparation. The plant samples were identified by and application called (Picture this) and authenticated by a botanist from the botany section of the department of biological sciences, MAU Yola.



SKETCHING OF BIOSYNTHESIS OF SILVER AND IRON NANOPARTICLES USING EXTRACT OF WATER HYCINTH

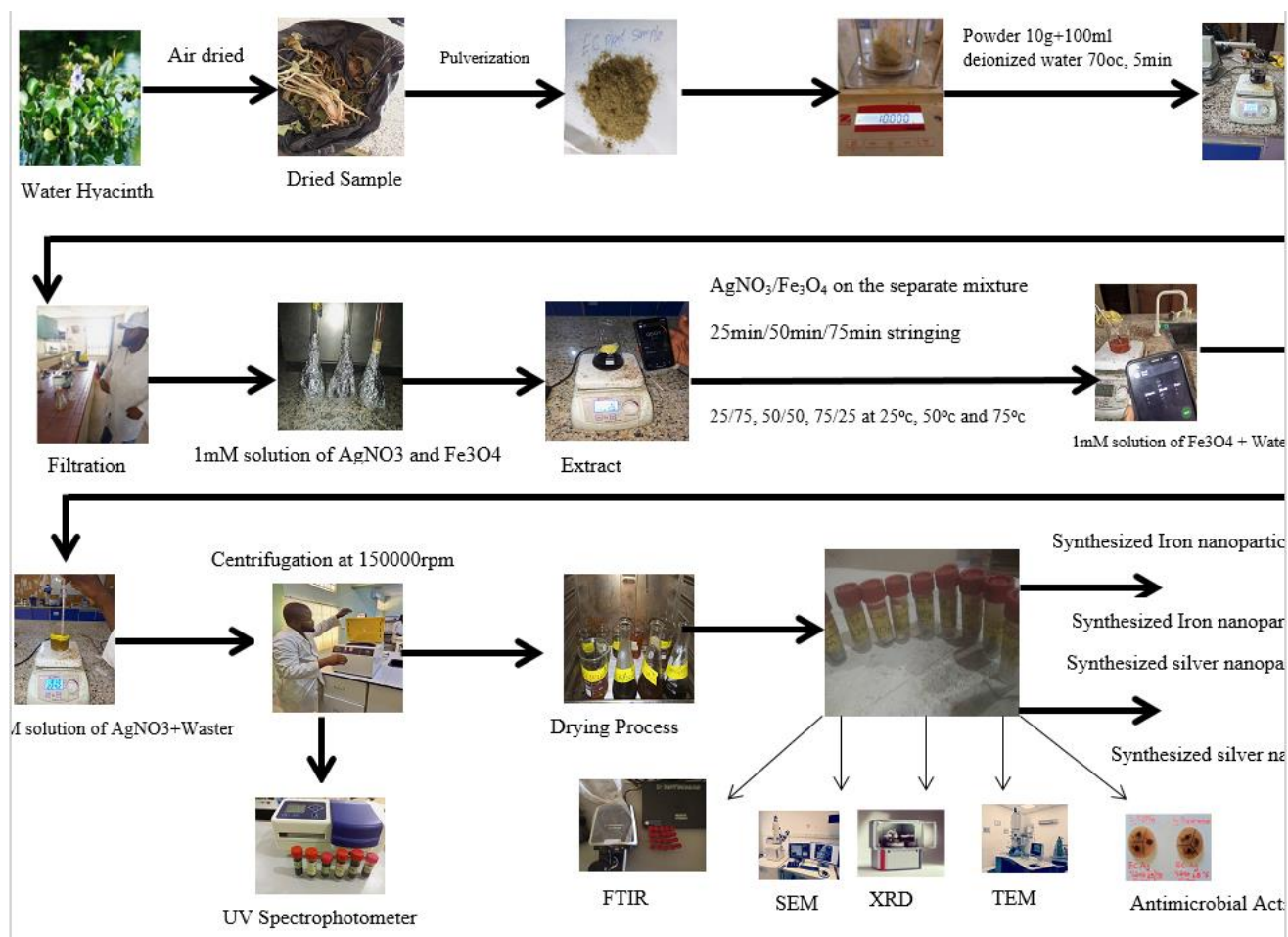


Figure 1

2.3 Sample preparation

The collected samples were washed with 5% (v/v) nitric acid (HNO₃) and then rinsed with deionized water to remove the possible contaminant. The samples were then cut into pieces with a stainless-steel knife and air-dried in the laboratory for 21 Days before oven-dried at 75 °C for 24hours. The samples were then crush into powder in a mortar with a pestle. Thereafter, the samples were sieved through a 2 mm nylon sieve to obtain a powder sample which was then be transferred into polyethylene bag for analysis.

2.4 Preparation of extract

A 10 g of the powdered sample was weighed by means of an analytical balance and transferred into a 250ml

beaker containing 100ml of deionized water then boiled for 10 minutes. The mixture will be cooled and then filtered through Whatman number 1 filter paper with the aid of suction pump. The extract obtained was stored in an oven 50°C for further analysis [9].

2.4.1 Biosynthesis of the *Eichhornia crassipes* silver nanoparticles (EC-AgNPs)

25mL of leaf extract was added to 75 mL of 1 mM silver nitrate (AgNO₃) solution as a silver ion reducing agent. The effect of the reaction was analyzed at different temperatures (25°C, 50°C and 75°C). The reduction of silver nitrate with respect to time was also evaluated at different time intervals (25 min, 50 min, and 75 min) for each temperature. The formation of silver nanoparticles was confirmed when the mixture turns yellowish brown in color.

2.4.2 Antioxidant activity of AgNO₃ NPs

For the evaluation of anti-oxidant activity, AgNO₃ NPs of *Eichhornia crassipes* (methanol and aqueous extracts) of 200, 400, 600, 800 and 1000µL were taken in a series of test tubes along 1ml of 100µM of 1,1-Diphenyl-2-picrylhydrazyl (DPPH) which is dissolved in methanol. The mixture is made up to 2ml using methanol. This was incubated in dark for the period of 30 minutes to react. After incubation time, the absorption value was measured using UV–vis spectrophotometer (UV-1800, Shimadzu) at nm. In this experiment, methanol served as blank, DPPH solution devoid of any test sample as positive control and Ascorbic acid as standard. The experiments were repeated thrice and their mean value for absorbance was recorded. The inhibition percentage was determined with the following equation,

$$\% \text{ Radical Scavenging activity} = \frac{\text{Absorbance of blank} - \text{Absorbance of sample}}{\text{Absorbance of control}}$$

2.4.3 Antioxidant activity of Fe₃O₄NPs

For the evaluation of anti-oxidant activity, AgNO₃NPs of *Eichhornia crassipes* (methanol and aqueous extracts) of 200, 400, 600, 800 and 1000µL were taken in a series of test tubes along 1ml of 100µM of 1,1-Diphenyl-2-picrylhydrazyl (DPPH) which is dissolved in methanol. The mixture is made up to 2ml using methanol. This was incubated in dark for the period of 30 minutes to react. After incubation time, the absorption value was measured using UV–vis spectrophotometer (UV-1800, Shimadzu) a nm. In this experiment, methanol served as blank, DPPH solution devoid of any test sample as positive control and Ascorbic acid as standard. The experiments were

repeated thrice and their mean value for absorbance was recorded. The inhibition percentage was determined with the following equation,

$$\% \text{ Radical Scavenging activity} = \frac{\text{Absorbance of blank} - \text{Absorbance of sample}}{\text{Absorbance of control}}$$

2.4.4 Anti-bacterial Screening of the Extracts

Anti-bacterial activities of all extracts of *Z. spina-christi* and *H. acida* were evaluated by the paper disc diffusion method. ESBL-producing *P. aeruginosa* was first reactivated by inoculating into a freshly prepared peptone water, and incubated for 48hrs at 37°C. Broth cultures of ESBL-producing *P. aeruginosa* in sterile solution of 0.9% normal saline already adjusted to 0.5% Mc Farland standard were inoculated onto cetrimide agar. Sterilized filter paper discs (Whatman No. 1; 6 mm in diameter) soaked in different test tubes containing the dissolved extracts of different concentrations were picked with sterilized forceps and air-dried and placed on plates with the organisms. The plates were incubated for 24hours at 37°C. The sensitivity or susceptibility of the bacteria to the standard drug was tested by placing discs seeded with amoxy-clav (20/10µg) on each plate of different concentrations; this was used as positive control while sterilized paper discs with DMSO were used as negative controls for all the plates. The experiment was performed in triplicates. Bactericidal activity were determined by measurement of zone of inhibition around each paper discs and results for that of the anti-pseudomonad agent was compared with those of the plant extract [10].

3.0 Result and Discussion

The results of the qualitative photochemical screening for *Eichhornia Crassipes* are shown in table 1 below.

Table 1: Qualitative Phytochemical Screening for *Eichhornia Crassipes*

Secondary metabolite	Result
Saponin	+
Flavonoids	-
Steriods	-
Tanins	-
Terpernoids	+
Phenolics	+
Terpenes	-
Alkaloids	-
Amino acid	-
Cabohydrates	+

Key: + Present, - Absent

The result in Table 1 shows the qualitative phytochemical studies which revealed that the leaf extract of *Eichhornia crassipes* contains carbohydrates, terpenoids, phenolics, saponins, while the amino acid, terpenes, Alkaloids, tannins, steroids, and flavonoids were observed absent.

3.1 Confirmation of Biosynthesized AgNPs

The color of the reaction changes as the temperature increases 25^o C to 75^o C from yellow to brown, when 75 cm³ of 1mM AgNO₃ solution extract was added to 25 cm³ of the plant extract on stirring. The yellow-brown color was obtained from the formation of AgNPs. This conforms to the results obtained [11].

Formation and stability of magnetite nanoparticles in aqueous colloidal solution is confirmed by using UV-vis spectral analysis.

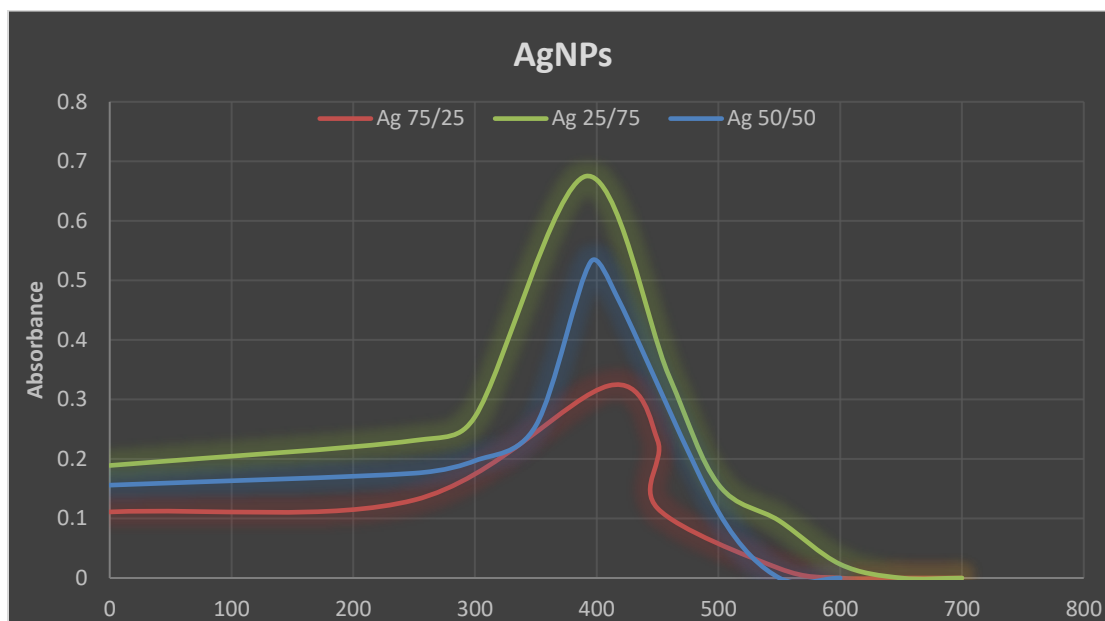


Figure 1 shows the behavior of the absorbance of silver nano particle solutions obtained at a temperature of 75^oC. A plasmon shift (391 to 410 nm) and an increase in absorbance as concentration increases (25/75, 50/50 and 75/25 Ag nanoparticles) were observed, reaching a maximum absorbance at 25/75 of reaction. The absorbance increase has been associated with a higher

concentration of metallic silver, while the displacement of the maximum peak towards longer wavelengths is mainly owing to the increase in particle size. The trends described at 75^oC were maintained at 50^oC and 25^oC, however, at these reaction temperatures the displacement towards higher wavelengths was less marked (391 to 410 nm).

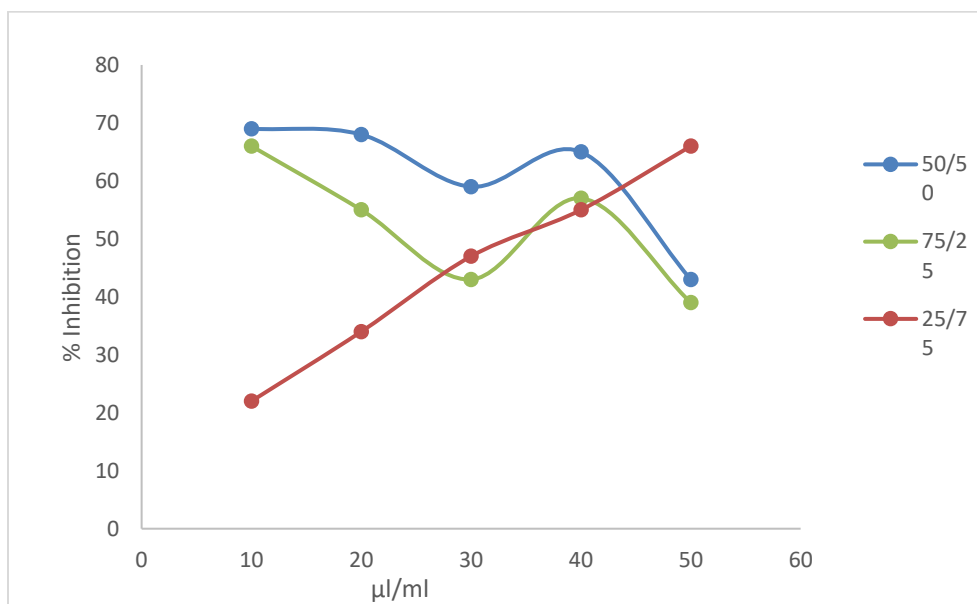


Figure 2: Spectrum of AgNPs

DPPH free-radical Scavenging Activity DPPH is stable free radical and is mostly used to estimate the ability of antioxidant activity of plant extract or natural compounds to work as scavenging of free radical or donating hydrogen atom [12] antioxidant activity of AgNPs extracts by DPPH method is shown in (Figure 2). AgNPs 25/75, AgNPs 50/50 and AgNPs 75/25 showed scavenging activity by inhibition of free radicals. The scavenging activity of AgNPs 25/75, 50/50 and AgNPs 75/25 extracts was very effectual and the power of extracts- significantly decreased with increasing of concentrations, while AgNPs 25/75 increases with increasing concentration. ApNPs 50/50 extract showed antiradical activity (69, 68, 59, 65 and 43% at 10, 20, 30, 40 and 50 µl/ml) respectively, and AgNPs 75/75 Showed the % Inhibition of (66, 55, 43, 57 and 39 % at 10, 20, 30, 40 and 50 µl/ml) respectively whereas, the AgNPs 25/75 (22, 34, 47, 55 and 66 % at 10, 20, 30, 40 and 50) The results showed a strong effect of AgNPs 25/75 extracts on scavenging activity of DPPH free radicals with non-significant difference between the effect of extracts. The results also indicate that the chelating effects of AgNPs 25/75 are higher than AgNPs 50/50 and AgNPs 75/25 over the increasing concentrations.

Furthermore, among the two extracts, the chelating activity of AgNPs extract is slightly higher than that of them. Most of the researches have mentioned that high phenolic content will lead to high radical scavenging activity. Nonetheless, the present study on AgNPs shows

a positive relationship between results of DPPH radical scavenging activity assay. AgNPs 25/75 is proven to be better radical scavenger as compared to 75/25 and 50/50 AgNPs even though it expresses phenolic content. Most of the previous findings have discovered that even though certain samples possess potent radical scavenging activity, the samples either possess moderate or weak ion chelating activity. For instance, a study conducted by [13] on guava has shown that the samples of guava possess potent radical scavenging activity but they have weak ion chelating effects.

3.2 Confirmation of Biosynthesized Iron Nanoparticles (Fe)

The color of the reaction changes as the temperature increases 25 °C to 75 °C from light red to brick red, when 75 cm³ of 1mM Fe₃O₄ solution extract was added to 75 cm³ of the plant extract on stirring. The fine brick red appears immediately after introducing the Fe₃O₄ Solution which shown the formation FeNPs [1]. In this research, different amount in percentages of EC plant extract were used, which were 75 %, 50 % and 25 % respectively. It was observed that increases in the EC plant volume, increases the absorbance values as well as decrease in the amount of EC plant extract decreases the absorbance value. Formation and stability of magnetite nanoparticles in aqueous colloidal solution is confirmed by using UV-vis spectral analysis.

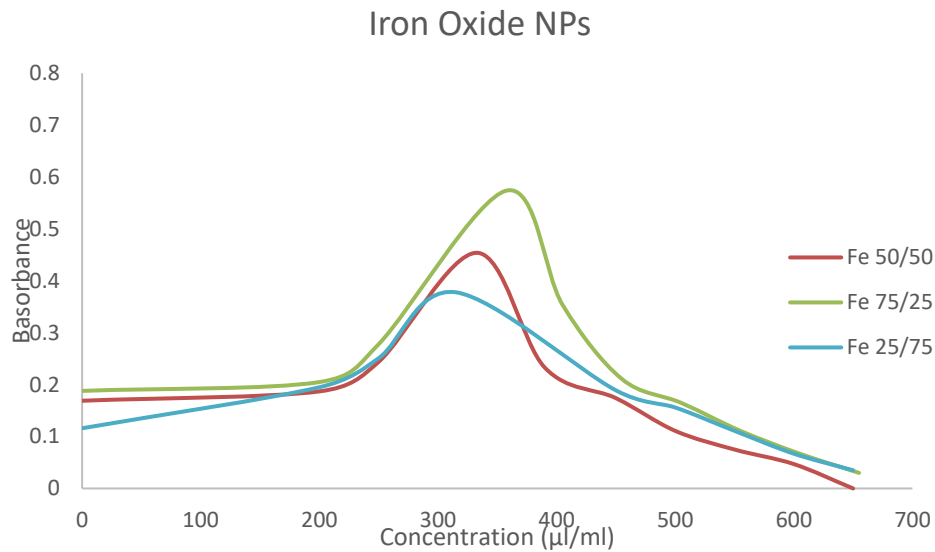


Figure 3: UV Spectrum of Fe₃O₄

Figure 3 shows the behavior of the absorbance of iron nano particle solutions obtained at a temperature of 75 °C. A plasmon shift (315 to 358 nm) and an increase in absorbance as concentration increases (25/75, 50/50 and 75/55 Fe Nano) were observed, reaching a

maximum absorbance at 25/75 of reaction. The absorbance increase has been associated with a higher concentration of metallic Fe, while the displacement of the maximum peak towards longer wavelengths is mainly owing to the increase in particle size.

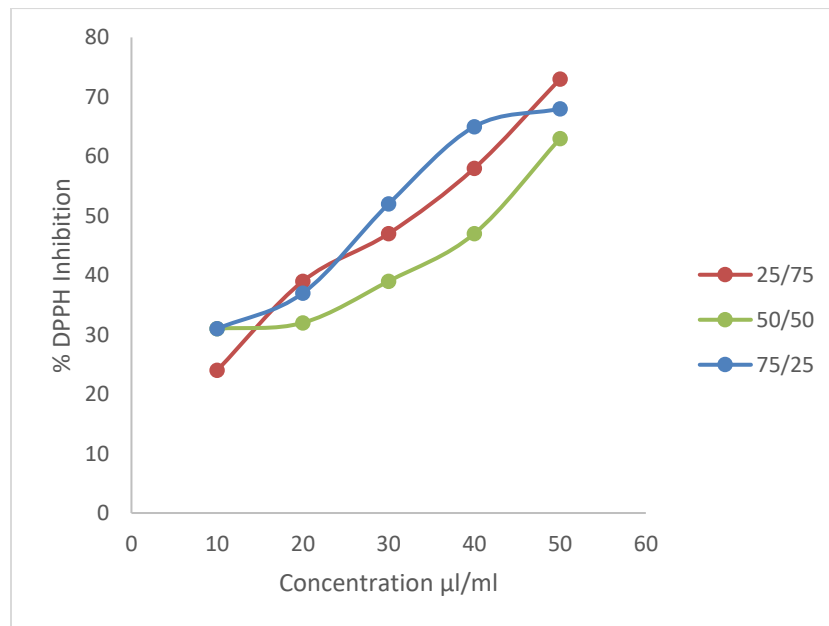


Figure 4: Antioxidant activity of Synthesized material

In this study, DPPH radical-scavenging activity were measured to identify antioxidant activity of EC Fe Nano 25/75, EC Fe Nano 50/50 and EC Fe Nano 75/25 extracts. DPPH radical-scavenging activity (%) of EC 25/75 Nano was increasing as the concentration increases 10, 20, 30, 40 and 50 $\mu\text{l/ml}$ with 24, 39, 47 55 and 73 % respectively. (EC 50/50 Nano), 31, 32, 39, 47 and 63%, Summing up with these data, this study concluded that water is the most suitable solvent to enhance radical-scavenging activity (Figure 4). the study results show that the EC Fe Nano 25/75 aqueous extract exhibited the highest DPPH (73 %), EC Fe 50/50 (63 %) and EC Fe 75/25 (68 %) radical-scavenging activity compared to the different extracts its antioxidant activities were also proportionally increased in the water extract. Comparing these results with the study developing turmeric extraction conditions [14] turmeric leaf ethanol extract (1 mg/mL concentration) showed higher DPPH radical-scavenging activity (80.11 %) than the water extract (74.65 %). where was also a significant difference in the radical-scavenging activity between ethanol extract and water extract in the same manner as the DPPH radical-scavenging activity. However, this indicates a slight difference, and we concluded that this distinction may come from the difference in the solvent concentration.

Bacteria/CONC.(mg/ml)		<i>Staph. aureus</i>				<i>Strep. Pneumoniae</i>				<i>E.coli</i>				<i>Salmonella typhi</i>				<i>Klebsiella pneumoniae</i>			
		R1	R2	R3	AVR	R1	R2	R3	AVR	R1	R2	R3	AVR	R1	R2	R3	AVR	R1	R2	R3	AVR
EC SILVER 25/75	400	22	24	21	22.3	20	20	19	20	21	22	21	21	18	19	18	18.3	16	15	17	16
	200	19	22	20	20.3	17	17	17	17	18	22	19	20	14	14	15	14.3	15	14	14	14.3
	100	16	14	14	15	11	9	8	9.3	16	13	15	15	9	10	10	9.6	9	9	9	9
	50	9	10	10	10	7	9	7	8	10	10	9	10	6	9	7	7.3	6	7	6	6.3
EC SILVER (50/50)	400	16	20	18	18	14	16	14	15	17	19	18	18	14	14	16	15	14	13	14	14
	200	14	16	13	14.3	10	10	10	10	13	15	14	14	10	11	12	11	11	12	13	12
	100	10	10	10	10	8	9	8	8.3	12	12	12	12	8	9	9	9	10	10	10	10
	50	6	6	6	6	5	3	4	9.3	7	7	7	7	5	7	6	6	5	5	5	5
EC SILVER (75/25)	400	11	12	11	11.3	11	10	9	10	11	12	11	11.3	10	9	10	10	10	10	10	10
	200	8	9	8	8.3	8	8	7	8	9	9	9	9	7	8	8	8	8	8	8	8
	100	5	7	7	6.3	5	5	5	5	6	6	6	6	5	4	5	5	5	5	5	5
	50	5	5	6	5.3	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
EC Fe (25/75) NANO	400	20	19	19	19.3	18	19	17	18	19	21	19	20	15	17	16	16	12	13	15	13.3
	200	16	16	16	16	14	18	16	16	16	18	17	17	11	13	12	12	11	11	10	11
	100	12	12	11	12	10	11	10	10.3	14	15	15	15	10	10	10	10	6	6	6	6
	50	7	7	7	7	5	7	5	6	10	10	10	10	6	6	6	6	5	5	5	5
EC Fe (50/50)	400	16	19	16	17	14	14	13	14	15	18	16	16.3	13	12	13	13	10	11	11	11
	200	14	16	15	15	12	12	12	12	12	12	12	12	9	8	8	8.3	9	7	9	8.3
	100	12	12	11	12	9	9	8	9	6	6	6	6	6	6	6	6	8	8	8	8
	50	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
EC Fe (75/25)	400	14	16	15	15	10	14	12	12	13	15	14	14	10	11	10	10.3	9	8	10	9
	200	11	13	13	12	9	8	6	8	9	7	9	8.3	8	9	8	8.3	8	7	8	8
	100	8	8	7	8	5	7	5	6	7	7	7		6	6	6	6	7	7	7	7
	50	4	4	4		0	0	0	0	6	6	6	6	0	0	0	0	5	5	5	5
EC	400	10	11	10	10.3	7	9	7	18	7	9	8	8	8	10	9	9	8	8	7	8
	200	6	8	6	7	6	6	6	6	6	6	6	6	6	6	6	6	8	6	8	7.3
	100	5	5	4	5	5	5	5	5	4	4	4	4	5	5	5	5	5	6	5	5.3
	50	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	00

Antimicrobial Activities of the EC extract and synthesized materials

3.3 Susceptibility test of the various

EC plant extract shows that it has activity against *S. aureus*, *Strep. pneumoniae*, *E. coli*, *Salmonella typhi* and *Klebsiella pneumoniae*. According to the world health organization report on infections disease, (2020) over coming antibiotic resistance is a major issue of the WHO for the next millennium. Hence the last decade witnesses an increase in the investigation of plants as a source of human disease management. This study

shown that increase in the investigation of plant as a source of antimicrobial activities of the extracts was enhanced by the increase in the concentration of the extract. This finding agrees with the report of [8] that higher concentrations of antimicrobial substances showed growth inhibition.

The activities of the plant extracts range from 25/75 Ec Silver Nano as first rank followed by Ec silver 50/50, Ec silver 75/25 as second, third and fourth respectively. The same pattern was observed in Ec Fe 25/75. Base on the organism staphylococcus aureus and Escherichia coli was shown to be more sensitive to the extracts.



Plate 1: Some of the plates showing zones of inhibitions

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Conflict of Interests

The authors declare no conflicting interest.

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