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RESEARCH PAPER

Studying the Effect of Silver Oxide Nanoparticles Using Laurus Nobilis Leaves in Inhibiting Bacterial Action

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ABSTRACT

In this work, the synthesis of silver oxide nano–particles (AgO-NPs) was carried out using Laurus nobilis leaves extract as a capping and reducing agent. Characteristics of the AgO-NP included; "XRD", "UV", "FT-IR", "SEM-EDX", and other systems. The UV-visible spectra of the aqueous medium peaked at 443.0 nm, which corresponds to the AgO-NPs' Plasmon absorbance. The XRD pattern shows the formation of AgO-NP with a face-centered cubic crystal structure. The SEM analysis showed that the particles had a spherical form. and roughly 16.9 nm in size. There were no harmful stabilizers or reducers added when the silver nanoparticles were being created.

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INTRODUCTION

In modern material science, nanotechnology is one of the most creative and productive areas of study. The life sciences, especially biotechnology and biomedical science, are greatly influenced by this ever-evolving scientific field [1,2]. Metal oxide nanoparticles, especially silver nano-particles (AgO-NPs), have recently attracted much attention due to their favorable properties related to the effect of quantum size and their wide range of possible uses in biomedical, nanostructure fabrication, optics, catalysis, and optoelectronics [3]. The green synthesis of silver oxide nano-particles has garnered global attention because of its low resource and energy consumption, as well as its environmental safety and biocompatibility [4]. AgO-NPs have a lot of potential for use in biology, including antibacterial qualities [5]. The reduction of metal ions in solution is the basic process behind the production of nanoparticles [6]. Since biomolecules released by the biomass can act as both stabilizing and reducing agents during the reaction, nanoparticles production is compatible with green chemistry techniques [7]. Three chemical ingredients are required for the preparation of nanoparticles: metal ions, reducing agents, and protecting agents [8]. Because plants contain a wide range of metabolites that can help stabilize, cap, and reduce silver ion, the amount and composition of metal nano-particles will vary depending on the plant type. [9]. Laurus nobilis leaves are widely used in cooking. It has shown antibacterial and antifungal properties in addition to hypoglycemic and antiulcerogenic qualities

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properties [10]. Plant extracts are easily converted into nanoparticles by simply agitating them at room temperature in an aqueous solution containing the metal salt; thus, this methodology can be regarded as an environmentally sustainable synthesis technique [11]. The preparation of nanoparticles in plants and microbes relies on the same basic mechanism. Metal salts consisting of metal ions are reduced by atoms reducing agents before particles are formed. After that, the resultant atoms group together and form tiny clusters [12]. The ability of NPs to exhibit antimicrobial activity is one of their most important applications in the medical field [13]. In comparison to the bulk form, the energy of the NPs increases due to increase the surface to- volume ratio, which reduce the stability of the NPs and increases their chemical activity, beside enhances both their chemical and physical characteristics of electron confinement and quantum production. The effects are easily observable and this makes them highly desirable materials for use in a variety of chemical applications [14,15]. Due to their unique biological properties, noble metal nanoparticles are the fastest growing class of nanoparticles and are used in a wide- range of studies. [16]. The exact mechanism by which noble metal nanoparticles works is still unknown, however, a number of theories have been proposed to explain their chemical effect [17]. Plant-based NP synthesis has certain advantages over additional biological procedures in that it can be suitably scaled up for large-scale NP generation and does not require laborious technology for maintaining cell cultures [18]. This work presents a green synthesis process that employs plant extract from Laurus nobilis leaves to produce silver nanoparticles in a more economical, ecologically benign, and effective manner.

MATERIALS AND METHODS

Method for Preparing Extracts from Plant Leaves

The plant (Laurus nobilis) were washed well with deionized water, then dried at 80°C in an air oven, then the leaves were pulverized into a powder. The second step involved combining 5 grams of the powder with 100 milliliters of deionized water at temperature 50 **°C** for 25 minutes. Finally, after removing plankton from the mixture by filtering it through filter paper, the mixture was centrifuged for 15 min at speed 400 rpm and kept inside a cold area until used.

Preparation of silver nitrate solution:

$AgNO_3$ is the chemical formula for nitrate of silver, an inorganic material. 0.2 M silver nitrate was produced and dissolved in 100 ml of deionized water without exposure to light. to form silver nitrate**.**

Preparation of the nanomaterial

At a temperature of 40°C, the Laurus nobilis plant extract was placed on a magnetic stirrer., and gradually distilling the AgNo3 solution to produce the extract by using a burette, and the temperature is slowly raised so that it does not exceed 60 degrees Celsius, and leave for 30 minute at least. This process is carried out until the color of the solution changes to give another color, i.e. a different substance is formed, which indicates that it obtained nanoscale silver. After completing the drip process, the resulting concentration was considered to be the original concentration, and then it will be diluted using distilled water in different proportions.

Preparation of concentration

The nanomaterial was diluted in different proportion using distilled water, The Concentrations (25,50,75%) were added to the ultrasonic device in order to disperse and separate the nanoparticles from each other for increase effectiveness.

Characterization Techniques

Nanoparticles can be characterized in a variety of ways. The first and simplest method is to alter the color of the solution. According to our research, the solution gradually changed color during the Ag-NP synthesis from light yellow to colloidal dim brown in a matter of thirty to forty-five minutes. Spectroscopy-based techniques such as “UV-vis”, “XRD”, “”SEM”, “EDX”, and “FT-IR”, are considered as indirect methods to determine details about the composition, structure, crystal phase, and characteristics of nanoparticles. A spectrophotometer “CARY, 100 CONC plus Uv - Vis - NIR, split beam optics, double detectors” equipped with a lamp of xenon and operating in the wave-length band of 300 - 1100 nm was used to assess the optical physical parameters. Potential bio-molecules responsible for the efficient stabilization and capping of silver nano-particles derived from Laurus nobilis plant extract were identified by the application of Fourier Transform-

Infrared (FTIR) spectroscopy. All thin layers were put to glass substrates prior to deposition. The nanoparticles' morphologies and diameters were determined using an X-ray technique (Shimadzu - XRD\6000; Shimadzu Company;"Japan"). The X-rays came from Cu-K_2 radiation, which has a wavelength of 0.15406 nanometers. With a 30 mA emission current, the system operates at 40 KV. A specimen of Ag nanoparticles was introduced into the sample container and examined under an electron microscope. The sample was next examined using a fully automatic scanning electron microscope"SEM" (JEOL; Model 6390).

RESULTS AND DISCUSSION

The Local Surface Plasmon Resonance (LSPR) has a significant impact on the optical characteristics of silver oxide nanoparticles. Due to the unusual optical features of nanoparticles, which depend on their aggregation state, surface, shape, and size as well as concentration, spectrophotometry is a useful method for the identifying and characterization of nanoparticles. The maximum absorbance ranges of AgO- NPs about "300– 550 nm", peak of Plasmon around "435 nm" is usual for AgO nanoparticles. The degree of colloid aggregation affects both the peak of plasmon and the half maximum of whole

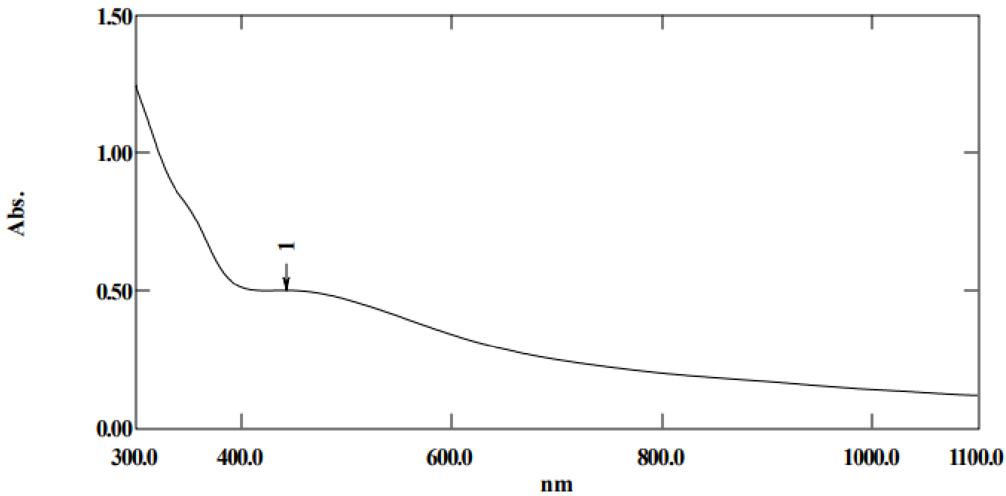

Fig. 1. Optical absorbance spectrum of AgO Nanoparticles

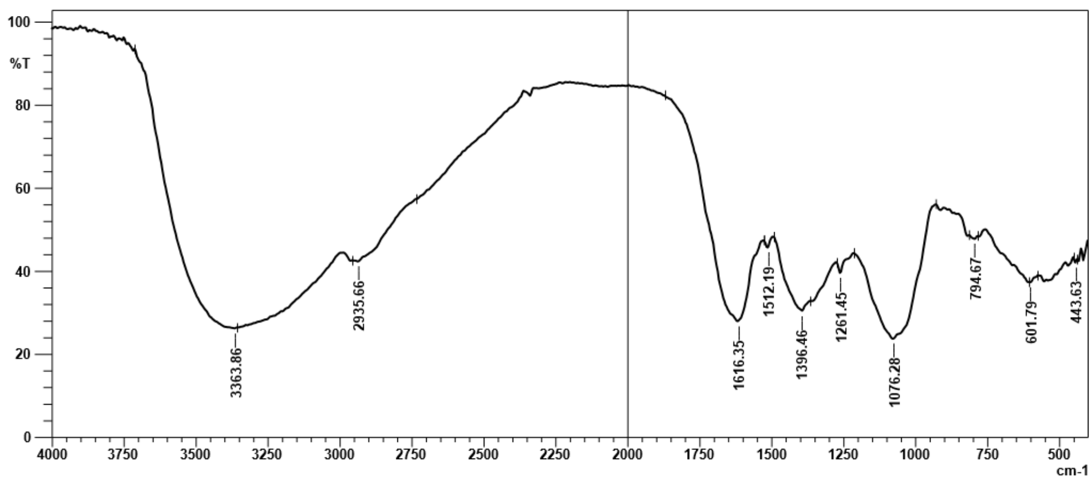

Fig. 2. FT-IR spectra of AgO Nanoparticles produced by Green Synthesis using plant extracts.

width [19]. Fig. 1. displays the optical absorbance curve of AgO nanoparticles. The wavelength of the spectrophotometer was set to between 200 and 1100 nm. the absorbance be effected by size of particle, medium of dielectric and biochemical environments. Since the minor spherical nanoparticles less than "20 nm" show weak surface of Plasmon range, the spectrum showed the presence of the Plasmon resonance peak at 443.0 nm, also, the UV-visible spectra shown confirm a wide peek at the surface plasmon resonance around 470 nm, this peak shows the reduction of silver nitrate into AgO-NPs. The absorbance curve decreases rapidly up to 550 nm before reaching to saturate. The quantum size effect has been shown to cause noticeable differences in absorption

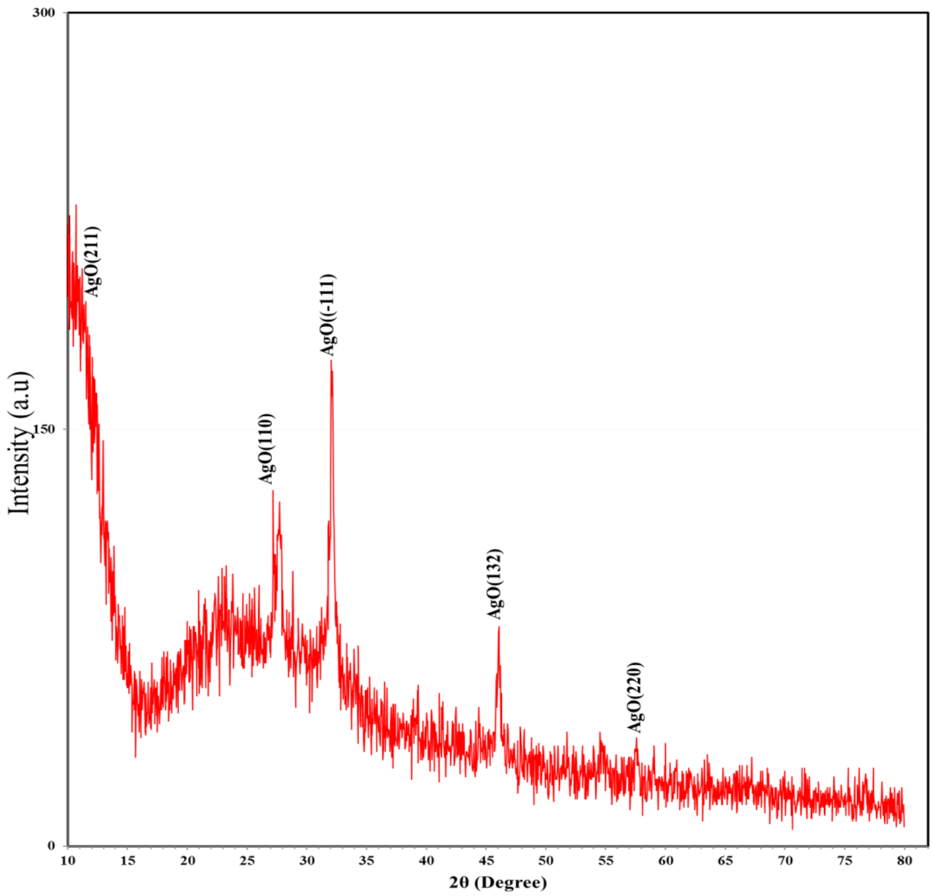

Fig. 3. XRD patterns of AgO nanoparticles prepared by Green Synthesis method using laurus nobilis leaves extract

Table 1. summery of X-ray characterization

2θ "Deg."	FWHM "Deg."	d_{hkl} Exp."Å"	G.S "nm"	d_{hkl} Std.	h k l
12.2570	0.1600	7.2153	49.9	7.2153	(211)
26.79	0.2390	3.4191	34.1	3.325	(110)
32.31	0.2390	2.7878	34.6	2.7614	(111)
46.46	0.2910	1.9530	29.7	1.953	(132)
57.63	0.7060	1.5982	12.8	1.6016	(220)

spectra.

FTIR spectroscopy is used to identify functional groups embedded in silver nanoparticles and to identify viable biomolecules responsible for the reduction of “Ag^+ “ions and the coverage of reduced AgNPs mixed with laurus nobilis extract. Functional groups have been seen in the 500–4000 range. As shown in Fig. 2, 3363.86, 2935, 1616.35, 1512.19, 1396.48, 1261.45, 1076.28, 800, 794.67, and 601.79. are notable peaks for the spectrum. The absorbance peak located at 3363.86 can be attributed to either hydroxyl (–OH) or bound amine groups (–NH). This hydrogen bonding might be responsible for the stability of the AgO nanoparticles. On other hand, the molecules that extend the vibrations of aliphatic group of the C-H bonds are at 2935Cm^{-1}. In the spectra of amides stretching vibrations of silver nano-particle, the C=O stretching peak performed at 1616.35cm^{-1}, and the carbonyl group also became noticeable in this range. Moreover, it is possible that -C C-of aromatic stretching and bending vibrations is the reason of the peak at 1512.19 cm^{-1}. The peak associated with the C-H bond lies in the range of 1396.48 cm^{-1}, while the C–N stretch of alcohol and carboxylic acid corresponds to 1261.45 cm^{-1}. The peak at 443.63 cm^{-1} can be assigned to C-O stretching from ester. Aromatic groups are associated with the peak at 800 cm^{-1}. Aromatic groups are responsible for the peak at 794.67 cm^{-1}. The peak at 601.79 cm^{-1} is related to the bonding of oxygen from the hydroxyl groups. In contrast, the bonding of AgNPs to oxygen was identified as the cause of the absorbance band at about 472 cm^{-1}. This result is in close agreement with reference [20].

The crystallinity of nanoparticles can be found through an “XRD” analysis. The pattern of XRD for AgO-NPs biosynthesized from extract bay leaf extract shows intense and powerful diffraction peaks with low “full width half maximum” (FWHM) of AgONP at 12.257º, 26.79º, 32.31º, 46.46º, and 57.63º (Fig. 3). These results indicate that the particles have crystallized. Furthermore, the Face-Centered Cubic crystal structure (FCC) of silver oxide atoms may be responsible for Miller indices (h k l) corresponding to the (211), (110), (-111), (132), and (220) planes. level (-111) was the dominating one, as evidenced by the intensity of its peak compared to other peaks. Structural Metal Deflections (FCC) (JCPDS Card No. 04-0783). Table 1: Summary of X-ray characterization.

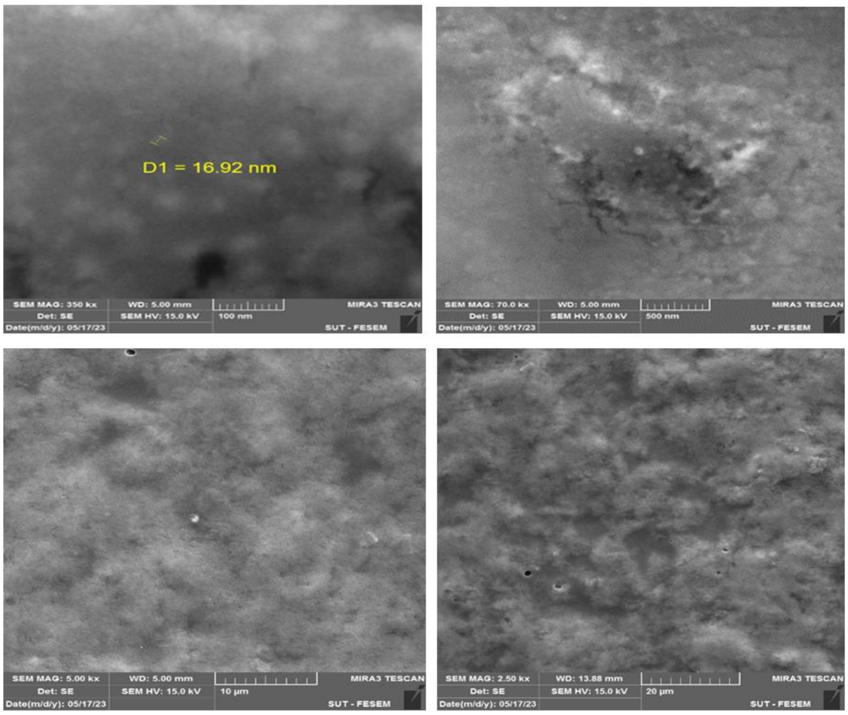

Fig. 4. SEM images of AgO biosynthesis NPs.

To learn more about the dimensions and form of the produced AgO nanoparticles, scanning electron microscopy, or SEM, was employed. The (SEM) revealed that form of nanoparticles is almost

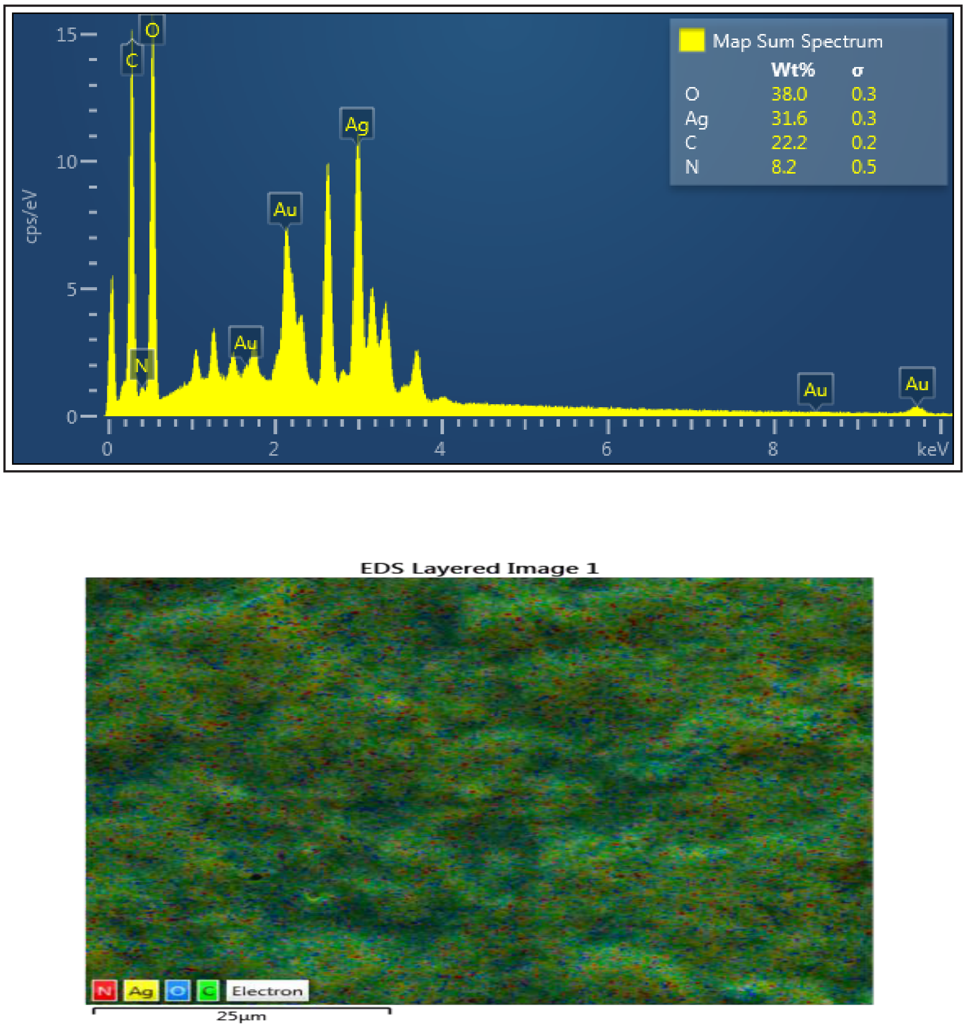

Fig. 5. (A) EDX spectrum of synthesized AgONPs , (B)distribution of Ag,O in elemental mapping

Table 2. summery of EXD characterization

Elements	Line Type	Apparent Concentration	k Ratio	Wt%	Wt% Sigma	Atomic %
C	K series	2.02	0.02024	22.16	0.25	36.17
N	K series	1.46	0.00260	8.20	0.53	11.48
O	K series	4.32	0.01455	38.03	0.34	46.60
Ag	L series	4.73	0.04734	31.61	0.31	5.75
Total:				100.00		100.00

spherical, average particle size of 16.92 nm. The high intensity distribution is highly asymmetric in nature of the nanoparticles in the sample (Fig. 4). large and small sized nanoparticles coexisted because of a time variation in their formation during synthesis. The SEM also revealed almost spherical shapes of nanoparticles found mostly in aggregated form.

The composition and purity of green synthesized AgO NPs were analyzed using an Energy Dispersive X-ray Spectroscopy "EDX"or "EDS". images from "EDX" taken after a 24-hour reaction time. The "EXD" analysis shown Fig. 5 indicates the presence of distinct elements in the sample by showing signals of varying intensities along the X-axis for the various binding energies (keV). The spectrum

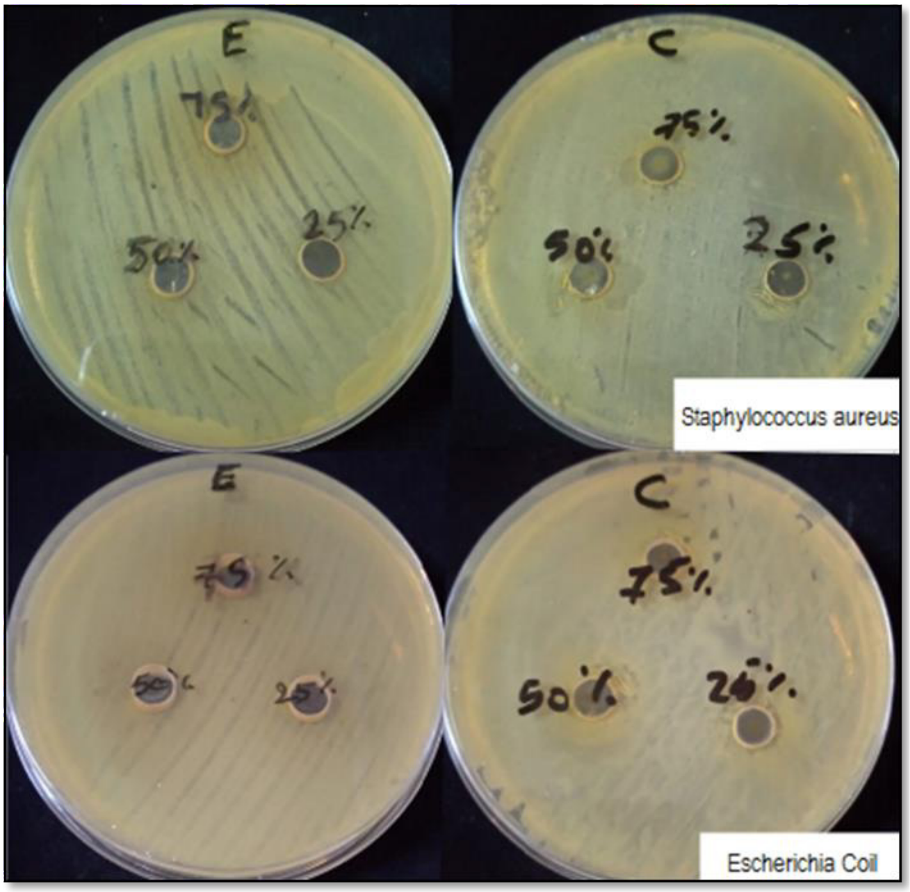

Fig. 6. The concentration of Ag NPs and aqueous extract of laurus nobilis leaves and effect on bacteria

Table 3. Biosynthesized Ag NPs (25%, 50%, 75%) inhibition zones (mm) against a subset of microorganisms of interest.

Test pathogens	Molar Concentration (µg/ml)	Staphylococcus aureus	Escherichia Coil
E (extract)	75%	13	11
	50%	11	9
	25%	-ev	-ev
C(compound nano)	75%	18	18
	50%	17	16
	25%	14	13

indicated that the most major components were showed intense peaks from O (38.03%) and Ag (31.6%) at 3.0 keV as Table 2 illustrates. A strong Ag, O signals with a distinct optical absorbance peak indicates that the extract was successfully used in the biosynthesis of AgONP. According the result of EDX, Moreover, EDX analysis revealed that there are dominant peaks corresponded to Nitrogen N and Carbon (C), Which indicates the presence of organic compounds (phytochemicals) on the surface of biosynthesized nano- particle. These components likely play a role in stabilizing plant extracts and coating biomolecules; In addition, they may be related to components found in plant proteins [21].

Staphylococcus aurous and Escherichia coli, two human harmful bacteria, were employed in this study, and the impact of the nanomaterial on the inhibition process was examined. After applying the previously created concentrations to the two types of bacteria, the outcomes indicated in the table below were attained, with a concentration of 75% showing the highest level of bacterial inhibition (Fig. 6). Additionally, as seen in Table 3, the results indicated that utilizing solely the plant extract had a minor beneficial effect on bacterial inhibition at a concentration of 75%.

CONCLUSION

Silver oxide nanoparticles can be manufactured using aqueous extract of the Laurus nobilis plant according to the results of this study. Reduction of silver nitrate using Laurus nobilis plant extract is a simple, efficient and clean method to fabricate silver nanostructures. Typical nanoscale properties of silver nanoparticles have been discovered through microscopic and spectroscopic investigations. From the FT-IR Study the stretching vibration of Ag-O group was found to be in range of 443.63 cm^{-1} which confirms the formation of AgO particles. According the result of EDX, the stoichiometric compound of AgO (31.61:38.03) is the most probable to form. The results of XRD shows that the diffraction angles 12.257º, 26.79º, 32.31º, 46.46º ,57.63º were attributed for (211), (110), (-111), (132) and (220) AgO samples.

CONFLICT OF INTEREST

The authors declare that there is no conflict of intere sts regarding the publication of this manuscript.

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