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*Department of Laser Physics and Its Applications*



***The Effect of Laser with Gold and Silver Nanoparticles on  
the Biological Activity of MDR Bacteria***

A Thesis

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Laser Physics and Its Applications

**By**

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1445 A.H

بِسْمِ اللَّهِ الرَّحْمَنِ الرَّحِيمِ

رَبِّ قَدْ آتَيْتَنِي مِنَ الْمُلْكِ وَعَلَّمْتَنِي مِنْ تَأْوِيلِ  
الْأَحَادِيثِ فَاطِرَ السَّمَوَاتِ وَالْأَرْضِ أَنْتَ وَلِيِّ فِي  
الدُّنْيَا وَالْآخِرَةِ تَوَفَّنِي مُسْلِمًا وَالْحَقِيقِي  
بِالصَّالِحِينَ ﴿١٠١﴾

صدق الله العظيم

سورة يوسف: 101

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**Zahraa**

## Dedication

To the dear daughter of the Prophet Muhammad, may God bless him and his family, and his darling.

To the pure woman.

To the woman who has suffered from wrongs and been robbed of her rights.

To the light that took me to a place of guidance.

My lady and majesty, **Fatima Al-Zahra (peace be upon her)**.

And to her grandson, whose we hope his appearance we, and will reveal the darkness, **Al-Mahdi Al-Hujjah bin Al-Hassan** (may God hasten his honorable appearance), the twelfth Imam who will fill the earth with justice and mercy.

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To the one whom the Lord - Glory be to Him - placed Paradise under her feet, and honored her in His Mighty Book ... (**My beloved mother**).

To the one who helped me achieve higher education (**my beloved father**).

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To those with whom I pledged that our hands would never stray from all goodness, my little sister and brothers.

Hoping for acceptance

## **Abstract**

Laser radiation and nanobiotechnology represent a point of contact between the fields of biology and nanotechnology, and it is a modern method used as a means of combating bacteria instead of traditional biomedicines. In the current study, the effect of nanoparticles (gold and silver) was studied, as well as the effect of nanoparticles and laser together to determine their effect on Gram-negative *Escherichia coli* and Gram-positive *Streptococcus pneumoniae* bacteria. The method used to prepare nanoparticles is pulsed Nd:YAG laser ablation with distilled water to prepare spherical-shaped gold and silver particles. Likewise, the laser ablation method was used by applying an electric potential difference to form rod-shaped gold and silver particles. In addition, the compositional and structural properties are studied through examinations such as transmission electron microscopy and ultraviolet-visible region spectroscopy.

There are three methods that have been used to determine the effect of nanoparticles on bacteria: the method of counting bacterial colonies, the second method is measuring the area of killing and inhibition, and the third method is spectroscopic analysis.

In all methods, it was found that silver nanoparticles were the best compared to gold nanoparticles. Spherical-shaped nanoparticles were more effective than rod-shaped nanoparticles. Also, using the property of these nanoparticles to absorb laser rays, it was shown that the highest decrease in bacterial cells occurred when irradiated with a 405 nm laser and in the presence of nanosilver. Both *E. coli* and *Streptococcus* bacteria were clearly affected by silver nanoparticles, with particles with a pulse number of 750 being the most effective. *E. coli* bacteria have been shown to show a greater response compared to *Streptococcus* bacteria in this context.

When using nanoparticles in conjunction with a laser, we had the best effect when exposing the bacteria for a duration of 5 minutes. This indicates that the combination of nanoparticles and laser has a positive effect on eliminating bacteria.

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## List of symbols

Symbol	Meaning	Units
CW	Continuous wave	
PT	Photothermal	
PDT	Photodynamic therapy	
NIR	Near infrared	
Q (X)	Absorbed power density	Wm <sup>-3</sup>
NPs	Nanoparticles	
UV	Ultraviolet	
$\mu_a$	Absorption coefficient	cm <sup>-1</sup>
Z <sub>thermal</sub>	Thermal penetration depth	
k	Temperature conductivity of the tissue	
PDT	Photodynamic therapy	
ROS	Reactive oxygen species	
APDT	Antibacterial Photodynamic Therapy	
SPR	Surface Plasmon resonance	
<sup>1</sup> O <sub>2</sub>	Singlet Oxygen	

# **Chapter One**

## **Introduction**

**1-1: Introduction**

Microorganisms like bacteria, molds, yeasts, and viruses frequently infect humans. As antibiotic-resistant microorganism's increase and healthcare costs rise, scientists are researching cost-effective antimicrobial agents. Silver based antiseptics have gained popularity due to their broad-spectrum activity and lower resistance to microbial resistance compared to antibiotics [1]. In fact, the potent antibacterial and broad-spectrum activity against morphologically and metabolically different microorganisms seems to be correlated with a multifaceted mechanism by which nanoparticles interact with microbes [2].

Many optical medical technologies employ laser radiation and fiber optics; therefore, coherence of light is very important for the analysis of light interaction with tissues and cell ensembles [3]. Nanoparticles absorb energy when irradiated, causing irreparable damage. Gold nanoparticles, in various modifications (spherical, rods, shells, etc), are promising candidates for photo thermal (PT) sensitizers due to their strength as strong nano absorbers and their absorption coefficient due to profound Plasmon resonance. These nanostructures are three orders-of-magnitude greater than other organic photosensitizers are for photodynamic therapy (PDT) [4]. The ultraviolet lasers generally produce both the photothermal and photomechanical effects, while the infrared and visible laser can produce thermal effects only [5].

Inorganic nanoparticles with strong near infrared absorbance bands, like gold, carbon, copper, and palladium, have been used in photothermal therapy for cancer treatment. By combining near infrared (NIR) lasers with these light-absorbing nanomaterials, local high temperatures are achieved, causing cancer cell death and converting light energy into thermal energy [6].

The unique properties of laser light, including monochromaticity, coherence, collimation, and high power, are the basis for the therapeutic

applications of laser energy. The energy, power, fluence, and irradiance are specific parameters of laser light that can be adjusted for specific clinical uses [7].

Parameters affecting tissue-light reaction in laser therapy have been classified: Laser parameters: laser power; energy density; wavelength; Operating mode (continuous or pulse); laser spot area. Optical properties of the tissue: tissue refractive index; surface circumference; target tissue depth; tissue temperature; thermal tissue properties. The monochromaticity property of laser light, related to its singularity of wavelength, is a determinant factor for the interaction with biological tissue, since it needs to be absorbed in order to interact with any tissue or matter [8].

Generation of heat is due to deposition of photons and by excitation and de-excitation of molecules increasing the internal energy . During the continuous illumination, the amount of optical energy absorbed per unit volume per unit time is called the heat absorbed power density,  $Q(x)$  in  $Wm^{-3}$  [9]. Lasers irradiation is described by several parameters. The wavelength is the most important. It determines the depth of the penetration by the light the higher the wavelength, the greater the laser penetration through the tissues, this is also associated with the thermal effect caused by the light which increases with the increasing wavelength [10].

Nanocomposite materials are gaining attention due to their unique optical, electrical, magnetic, biological, and mechanical properties and their ability to understand fundamental phenomena at the nanoscale [11]. Their bactericidal activity depends on size, concentration, morphology, and surface properties [12].

Nanoparticles (NPs) are regarded as nanoentities whose size ranges from 10 to 100 nm. The properties of the NPs influence their behavior in vivo. Particularly, morphological properties like shape and size, can influence NPs

circulation and targeting within the body, the shape and size of NPs are also responsible for specific cell signaling [13]

## 1-2: Literature review

In (2012) Babak Sadeghi *et.al.* Silver nanoforms, Ag-nanoplates (Ag-NPLs), Ag-nanorods (Ag-NRds), and Ag-nanoparticles (Ag-NPs) were evaluated for their antimicrobial properties. The results demonstrated that Ag-NPLs require a lower concentration than Ag-NRds and Ag-NPs to inhibit the development of *S. aureus*. The inhibition halo in Ag-NPLs increased with increasing surface area and was discovered to be dependent on the nanosilver morphologies [14].

In (2015) Juan, Carlos, *et.al.* They studied the bactericidal and bioprotective activities of gold nanofilms (AuNRs) against microorganisms using plasmonic photothermal therapy (PPTT). After experimental analyses, AuNRs PPTT shows better results in Minimum Inhibitory Concentrations (MICs) and Minimum Bactericidal Concentrations (MBCs), when it was compared with AuNRs alone. The laser employed to activate the AuNRs had no antibacterial effect against oral microbes. The MICs and MBCs values were higher for *S. aureus* and *E. coli* and lower against *S. oralis* [15].

In (2016) Lefta, Sadiq H *et.al.* Evaluated the antibacterial activity of silver nanoparticles. The AgNPs at 1  $\mu\text{gml}^{-1}$  and above showed noticeable antibacterial action for mainly following long incubation periods. The irradiation of bacterial suspension containing AgNPs with 405 nm laser light maximize the long term cytotoxicity of AgNPs detected after 24 hours following the treatment [16].

In (2017) Iftekhar Hossain *et.al.* Silver nanorods may have anti-microbial properties due to their sharp tips, which can puncture bacterial cells and kill them via impalement. A study comparing silver and CdS nanorods found no significant difference in anti-microbial properties, suggesting that any material may possess anti-microbial properties. The study suggests that nanorods made of any material could be a universal anti-microbial agent [17].

(2018) Lihui Yuwen *et.al.* Modified MoS<sub>2</sub> nanosheets (MoS NSs) with polydopamine (PDA) and then grew silver nanoparticles (AgNPs) on their surface to form MoS<sub>2</sub>-PDA-Ag nanosheets (MPA NSs) as antibacterial nanomaterials for the treatment of *Staphylococcus aureus* (S. aureus) membranes. Vitality and a *Staphylococcus aureus*-infected lesion. The results demonstrate that after treatment with MPA NSs under near infrared (NIR), laser irradiation can effectively eliminate S. aureus biofilms by destroying 99.99% of bacteria within the biofilms, demonstrating significantly enhanced therapeutic efficacy in comparison to the MPA group. Just. Or only NIR laser range [18].

In (2017) Marwah, Shereen A. Fouad *et.al.* Gold nanoparticles 25 nm were synthesized by co-precipitation method and characterized by different techniques including; Transmission Electron Microscope (TEM), X-ray Diffraction and Dynamic Light Scattering. Three concentrations of AuNPs (50, 100 and 200 lg/mL) were utilized for estimating the bacterial growth rate and the Minimum Inhibitory Concentration (MIC). The mechanism of interaction between AuNPs and bacteria was evaluated by transmission electron microscopic image analysis. Results revealed that MIC of AuNPs and AuNPs – laser combined therapy were 200 lg/mL and 100 lg/mL respectively [19].

In (2018) Acharya, Debashish *et.al.* Evaluated the effect of AgNP-sp and Ag-NRs for their antibacterial activity against Gram-positive and Gram-negative bacteria. Disc diffusion studies revealed greater efficacy of AgNPs against *Klebsiella pneumoniae* AWD5 at doses of 249 and 392 g. FESEM of the bacterial culture treated with AgNPs confirmed the antibacterial activity, and the killing kinetics confirmed the higher mortality rate against AgNP-sp compared to Ag-NR [20].

In (2019) Al-Sharqi, Anes *et al.* Studied the use of visible laser light to increase the antibacterial efficacy of Ag-NPs against *Escherichia coli* and *Staphylococcus aureus*, four concentrations of Ag-NPs (12.5, 25, 50, and 100

g/ml) were used. Results showed that the laser-activated Ag-NP treatment reduced the surviving population to 14% of the control in the *E. coli* population, while the survival in the *S. aureus* population was reduced to 28% of the control upon 10 min exposure time at the concentration of 50 µg/ml. These results indicate that irradiated Ag-NPs are more effective than conventional antibacterial agents [21].

In (2019) Hashimoto, Masanori, *et.al.* In this study they investigated the effect of AuNP size on *Streptococcus mutans* (*S. mutans*) cell attachment and initial biofilm formation. Three diameters of AuNPs used: 5, 10 and 20 nm. 5 nm AuNPs induced modest inhibition of *S. mutans* biofilm growth, whereas adhesion of 10 nm and 20 nm AuNPs to bacterial cells was negligible. It is assumed that the binding of AuNPs to *S. mutans* disrupts the formation of primary biofilms [22].

In (2019) Annesi, Ferdinanda *et.al.* Demonstrated that SEM analysis of the dried culture of *E. coli*/GNRs clearly interpreted the morphology of the bacteria, whereas backscattered electron microscopy (BSE) imaging demonstrated that the GNRs were not in contact with the bacteria. Photothermal experiments using laser light (=810 nm) demonstrated that by irradiating GNRs in *Escherichia coli*, the temperature could be increased to 50 °C in about 5 minutes, and cell viability experiments demonstrated that under this laser photoirradiation ( $I = 6.3 \text{ W/cm}$ ), the presence of GNR in the *E. coli* culture was able to induce a 2 log CFU reduction in live bacteria [23].

In (2020) Krce, Lucija, *et.al.* Created silver nanoparticles in deionized water by laser ablation and tested them against *Escherichia coli* DH5 cells. The MIC/MBC values for AgNPs were  $73 \pm 11 \text{ g/mL}$ . Using OD measurements, growth curves of bacteria treated with sub-MIC and MIC concentrations of AgNP were obtained. The primary effects of treatment included an increase in

baseline, an increase in apparent latency time, and a decrease in maximal Optical density [24].

(2020) Linklater, Denver P *et.al.* Studied uptake of NPs increases membrane tension, which leads to cell rupture and death. To study the antibacterial mechanism of *Pseudomonas aeruginosa* and *Staphylococcus aureus*, semi-spherical and golden-shaped NPs with increased affinity are synthesized, which leads to increased elongation and membrane rupture [25].

(2021) Ahmed O. El-Gendy *et.al.* Femtosecond laser to blue light, they demonstrated the possibility of enhancing the bactericidal effect of modest concentrations of silver nanoparticles. Using 50 mW laser radiation and 400 nm femtoseconds, the growth dynamics of *Pseudomonas aeruginosa*, *Listeria monocytogenes*, and methicillin-resistant *Staphylococcus aureus* were measured. The combined therapy was more antimicrobial than either silver nanoparticles or photoirradiation alone. MRSA was more resistant to the bactericidal effects of silver nanoparticles than *P. aeruginosa* and *L. monocytogenes* [26].

(2021) Jimmy Gouyau *et.al.* Synthesized gold and silver nanoparticles with a diameter of 12 nm and evaluated their antibacterial activity against *Staphylococcus aureus* and *Escherichia coli* using a chemical method. The results demonstrated that AuNPs possessed weak antibacterial activity, whereas silver nanoparticles exhibited no activity against *S. aureus* but a minimum inhibitory concentration of 128 mol/L against *Escherichia coli* [27].

(2021) Sivaji Sathiyaraj *et.al.* Demonstrated a color change synthesis of AuNPs, which was further confirmed by UV-vis spectra against the highest absorption peak at 527 nm. The primary structure and spherical shape of the PG-AuNPs were confirmed by HRTEM and EDS analysis. Zones of maximum and minimum inhibition for the antibacterial activity of PG-AuNPs were observed

against *K. pneumoniae* ( $17.12 \pm 0.14$  mm) and *B. subtilis* ( $11.42 \pm 0.58$  mm) [28].

In (2022) Aadim Ph D *et.al.* They used pulsed-laser ablation by liquid technology (PLAL) to synthesize silver nanoparticles (AgPNs) using Nd:YAG lasers with (355 nm) and (532 nm) wavelengths at 500 mJ and 600 mJ, respectively. Its antibacterial activity was evaluated against two strains of Gram-positive bacteria and two strains of Gram-negative bacteria. The results indicate that the AgNPs produced by the PLAL method have antibacterial properties and can be used to eliminate harmful and pathogenic bacteria [29].

In (2022) Faizah A. AlMalki *et.al.* Synthesized quasi-spherical carbon nanomaterials of varying diameters using the liquid method and pulsed laser ablation. The 220 mJ laser energy produced CNMs with the highest activity against both strains, with an inhibition zone (IZ) of approximately 34 0.1 mm in *S. aureus* and 31 0.5 mm in *E. coli*. The synergistic effect of carbon NPs resulted in an IZ of 14 mM for *S. aureus* and 12 mM for *E. coli* when compared to CNMs prepared at 160 mJ [30].

(2023) Angela Candreva *et.al.* Created 50-nm spherical gold particles (AuNP) coated with polyethylene glycol and administered them to an *Escherichia coli* culture. Experiments were conducted with various concentrations of gold nanoparticles in the absence and presence of light. At low concentrations, AuNs-PEG-SH aqueous solutions do not inhibit bacterial growth, but at 1.56 g/ml and 3.54 g/ml, we observe a bacteriostatic effect (46% growth inhibition at 3.54 g/ml) and a bactericidal effect (99% growth inhibition) respectively [31].

### **1-3: Aim of the study**

This research aimed to evaluate the antibacteria potential of nanoparticles (NPs) as stand-alone agents and their combined efficacy with laser therapy against multi drug resistant bacteria. Nanoparticles, due to their unique physicochemical properties, have been recognized as promising materials in various fields, particularly in the domain of antimicrobial treatments. Laser, in combination with NPs, can potentiate antimicrobial effects by causing localized hyperthermia and triggering photocatalytic processes.

Studying the effect of the shape of gold and silver nanoparticles on some different types of bacteria.

Studying the effect of gold and silver nanoparticles with different laser wavelengths on some different types of bacteria.

# **Chapter Two**

## **Theoretical part**

## **2-1: Introduction**

Laser technology offers unique advantages in manipulating microorganisms due to its precision, versatility, and ability to generate intense and focused light beams. Different types of lasers, such as ultraviolet (UV), visible, and infrared lasers. These lasers can interact with microorganisms at the cellular or molecular level. Nanoparticles, on the other hand, have emerged as powerful tools for targeted therapies and biomedical applications. These microscopic particles, typically ranging from 1 to 100 nm in size, possess distinct physicochemical properties that enable them to interact with microorganisms in unique ways. The combination of lasers and nanoparticles has shown great promise in enhancing the biological activity of microorganisms. This study aim to explore the effect of laser irradiation and nanoparticles on the biological activity of two type bacteria.

## **2-2: Molecular absorption of laser light**

Light interacts with tissues through transmission, reflection, scattering, and absorption. Transmission passes light through tissue without affecting its properties. Reflection repels light off the tissue's surface, with 4% to 7% reflecting off skin. Scattering spreads light within the tissue, resulting in larger radiation areas. The amount of scattering is inversely proportional to laser wavelength [32]. Water and proteins absorb light in two regions: ultraviolet and infrared. The absorption peak for aromatic rings of proteins and nucleic acids is in the UV region, leading to poor light penetration into tissues. Blood absorbs light in a broad wavelength range up to red light (630 nm), and above 600nm, absorption is poor. Melanin absorbs light from ultraviolet to near infrared. The absorption coefficient of tissue particles is small between 600 nm and 1.3  $\mu\text{m}$ , allowing laser light penetration into tissues. Laser-excited particles emit fluorescence or phosphorescence light,

depending on the excited electron lifetime [33]. The monochromaticity property of laser light, related to its singularity of wavelength, is a determinant factor for the interaction with biological tissue, since it needs to be absorbed in order to interact with any tissue or matter. Biological tissues have light receptors (chromophores) that are highly selective to the wavelength it absorbs. In the case of biological tissues, some common chromophores include hemoglobin, oxyhemoglobin, melanin and water [34]. Laser can increase the temperature of cells and it results in denaturation of proteins and collagen that leads to coagulation of tissue and it can necrotize cells [35].

Biological tissues have an average refractive index higher than that of air. When light interacts with the tissue surface, part of the light is reflected at the air/tissue interface; while the remaining light interacts with the tissue and penetrates into. Figure (2-1) shows the behavior of a laser beam interacting with the structure of absorbers and scatters on a tissue slab. The arrows indicate photon propagation: reflection at the air/tissue interface; backscattered photons, also named diffuse reflection; and absorbed and transmitted photons, also named diffuse transmission [36].

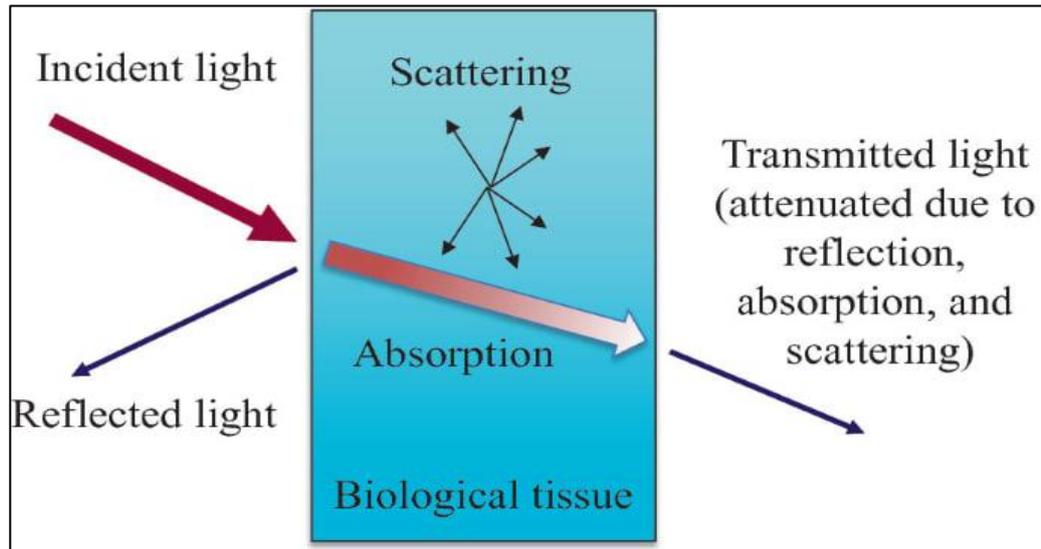


Figure (2-1): Basic effects of light-tissue interactions [37].

### 2-3: Laser-Tissue Interaction

Three types of reactions occur when laser light interacts with tissue:

- **Photochemical Interactions:** When a photon is absorbed by a molecular chromophore, it converts it into an excited state, storing laser energy as chemical energy. This excited state can participate in a photochemical reaction, producing various reactions like bond breaking, crosslinking, radical formation, oxidative injury, or photodestruction. The molecular chromophore can be endogenous or an exogenous molecule introduced into the body [38].

- **Thermal Reactions:** The energy of laser irradiation is transferred into heat due to the absorption of the photons by tissue components, DNA/RNA, chromophores, proteins, enzymes, and water. According to the degree of heating, stepwise and selective thermal damage can be achieved: 42–45°C: beginning of hyperthermia, conformational changes, and shrinkage of collagen; 50°C: reduction of enzymatic activity; 60°C: denaturation of proteins, coagulation of the collagens, membrane permeabilisation [39].

- **In plasma-induced photoablation** a free electron is accelerated by the intense electric field in the vicinity of the laser beam. By colliding with a

molecule and freeing another electron, it initiates a chain reaction of similar collisions, resulting in Plasma is a mixture of ions and free electrons [40].

A light-absorbing medium will absorb a fraction of incident light per incremental path length of travel within the medium. The absorption coefficient  $\mu_a$  ( $\text{cm}^{-1}$ ) is defined as [41].

$$\mu_a = -\frac{1}{T} \frac{\partial T}{\partial L} \dots \dots \dots (2 - 1)$$

Where T (dimensionless) is the transmitted or surviving fraction of the incident light after an incremental path length  $\partial L$  (cm). This fractional change  $\partial T/T$  per  $\partial L$  yields an exponential decrease in the intensity of the light as a function of increasing path length L .

In addition, the photons absorbed in the second layer will heat this layer and dissipate to surrounding regions where the temperature is lower. Thermal dissipation will also occur into the tissue. The depth where the temperature decreases to 1/e of its peak value will be the thermal penetration depth ( $z_{\text{thermal}}$ ), described as [42]

$$z(t)_{\text{thermal}} = (4kt)^{-1} \dots \dots \dots (2 - 2)$$

Where k is the temperature conductivity of the tissue and t is the time of laser action on the tissue (laser pulse duration). The temperature conductivity is related with the heat conductivity (k) expressed in W/mK, the tissue density ( $\rho$ ) expressed in  $\text{kg/m}^3$ , and specific the heat capacity (c) expressed in kJ/kg K.

The intensity I of the electromagnetic wave propagating into the material (z-direction) decreases exponentially following the Beer-Lambert expression, according to [43]:

$$I(z) = I_0 e^{-\alpha z} \dots \dots \dots (2 - 3)$$

With

$$\alpha = \frac{4\pi \cdot k}{\lambda} \dots \dots \dots (2 - 4)$$

The inverse of the absorption coefficient  $\alpha$  is the optical penetration depth, i.e. the depth in which the major fraction of the energy is deposited (1/e of the total amount absorbed).  $k$  Is the extinction coefficient.

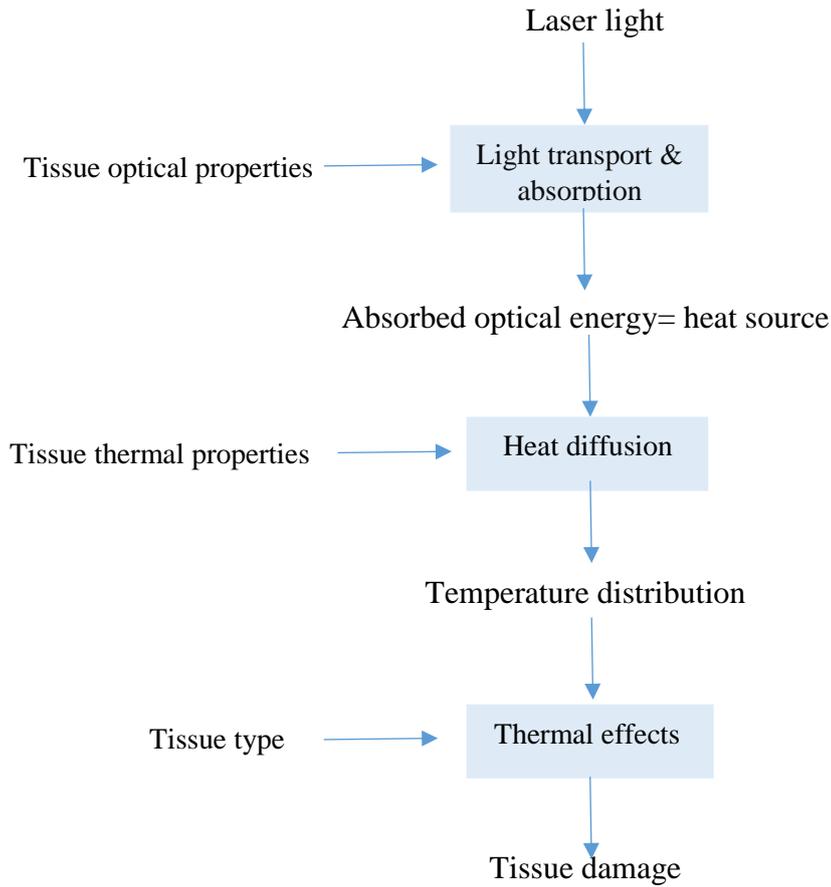


Figure (2-2): The various aspects involved in thermal interactions of light with tissue [44].

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## 2-4: Photosensitization

Photodynamic therapy (PDT) is a process which involves bacterial cell death by the use of light energy of appropriate wavelength and photosensitive drugs or dyes. During PDT, a photosensitizer is added to a bacterial sample, which absorbs light energy and causes production of reactive oxygen species (ROS) within or outside microbial cell, depending upon the distribution of PS and penetration of light energy [45]. The main advantage of antibacterial photodynamic therapy (aPDT) is that it can be used against both Gram-positive and Gram-negative bacteria [46]. Photodynamic therapy requires basically three elements: photosensitizer, light and oxygen [47]. The initial physical event in a photosensitization reaction is the absorption of a UV/visible photon by the photosensitizer, the initial process is an electron transfer from the biological target to Sens\* giving rise to the corresponding pair of radical ions (Sens•– and S•+), which in turn, can be in equilibrium with their corresponding neutral radicals [48]. The triple-excited PS molecule can also transfer energy with molecular oxygen to form  $^1\text{O}_2$ . As the most threatening species of ROS,  $^1\text{O}_2$  can directly cause oxidative damages to biological molecules, such as unsaturated lipids, peptides, enzymes, and other cellular components, and thus effectively kill bacteria (type II mechanism) [49].

Visible light has a suitable wavelength that is compatible with the absorption of the thermal photosensitizer material. Here, gold and silver nanoparticles were used. The key to this technique is a photothermal transducer able to absorb near infrared (NIR) light and convert it into heat effectively. Gold nanoparticles (Au NPs) are capable of absorbing and scattering a wide range of visible and NIR light, depending on their particle size and shape. NIR laser-induced electronic oscillations at Au NPs surface transfer into heat, bringing about elevated temperature of Au NPs as well as the surrounding environment [50]

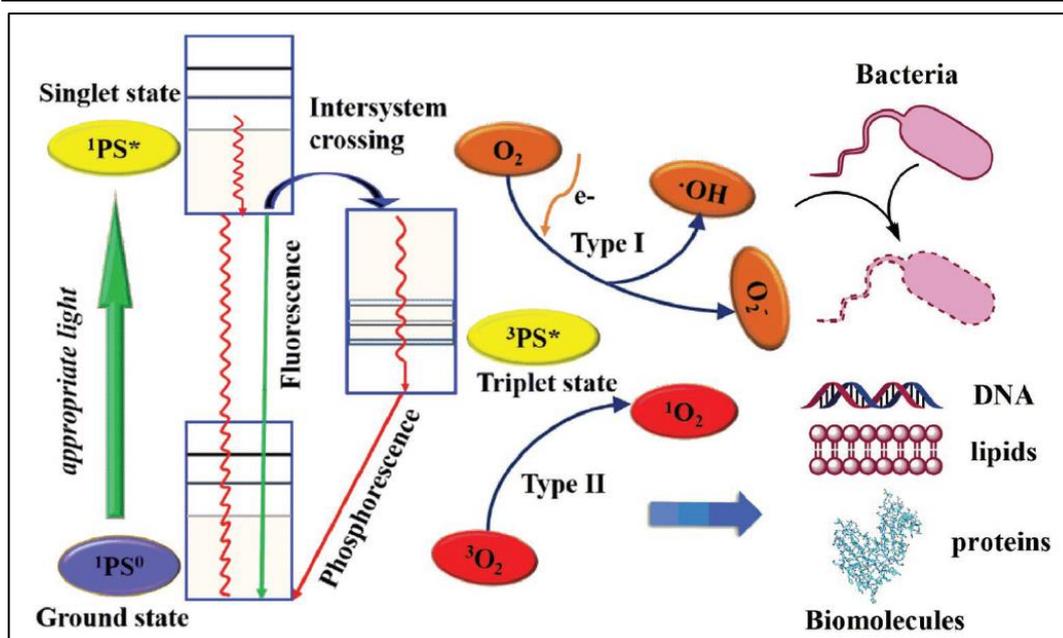


Figure (2-3) PDAT mechanism of PSs under appropriate light emission [51].

## 2-5: Surface Plasmon resonance (SPR)

It is observed that the percentage of killing and inhibition in the presence of nanomaterials and lasers is higher than that in the presence of nanomaterials alone, and this is due to the surface plasmon resonance, and its effect on silver and the 405 nm laser is greater than that of nanoscale gold and the 532 nm laser, because the Plasmon effect on silver is greater than Gold because it depends mainly on the intensity of the fall and the width of the beam. SPR is deemed the most remarkable optical property of metals and is briefly defined as the collective oscillations of the delocalized electrons in a metallic surface stimulated by the incident light under resonance conditions. When the frequency of the incident light matches the oscillation frequency of the surface Plasmon's, the resonance condition is achieved and the energy transfer occurs between the light's electric field and the surface plasmons, leading to a dramatic decrease in the reflected light intensity [52]. Typical metals that support surface Plasmon's are silver and gold, but others such as copper, titanium, and chromium have been used [53]. It is possible that laser radiation in the surface

Plasmon resonance mode changes the electronic state of Ag-NPs, which increases the antibacterial activity of the system as a whole [54]. There is a strong interest in the use of plasmonic metal nanoparticles for bacterial infection treatments through photothermal therapy. As NIR light is reported to penetrate the subcutaneous cell layer up to a depth of 10 mm to a few centimeters, the structural dimensions of metal nanostructures are specifically designed to ensure that they maximally absorb NIR light for subsequent effective conversion into heat [55].

## **2-6: Nanomaterials**

Nanoscale materials are defined as a set of substances where at least one dimension is less than approximately 100 nanometers [56]. Optical, mechanical, electrical and color properties of the same material in macro/micro and nano size may be different or even the opposite of other scales. Some properties that do not occur in macro size may appear in nano size. The main reason for this is the increased surface area/volume ratio with decreased material size and the non-continuous dimensions in nano-scale compared to macro dimensions [57]. Nanoparticles (NPs) are heavily used in biomedical, industrial, and commercial applications due to the benefits associated with the specific physical and chemical properties of both the bulk and the nanoscale material [58]. The two main approaches to explaining nanotechnology to the general public have been oversimplified and have become known as the ‘top-down’ approach and the ‘bottom-up’ approach [59]. Classification of nanomaterials on the basis of dimensions to One-dimensional nanomaterials, two-dimensional nanomaterials, and three-dimensional nanomaterials are the three types of nanomaterials [60]. There are several ways to prepare nanoparticles, including physical, chemical, and biological methods [61].

## **2-7: Nanoparticles used for killing and inhibition**

Metallic nanoparticles have unique properties relative to their bulk counterpart, and have been found to be of great value in antimicrobial applications, biosensor materials, conductive materials, and electronic components [62]. The metal nanoparticles such as silver, gold are becoming indispensable in various fields because of their vast applications against various diseases and environmental applications [63]. Smaller silver nanoparticles seem to have a higher toxicity compared to larger ones [64]. It is hypothesized that the NP mediated inhibition of the initial formation of biofilms occurs through one or more of the following mechanisms: 1) Destruction of bacteria, 2) Inhibition of glucosyltransferases (GTFs) production in bacteria due to disruption of cellular functions, 3) Inactivation of GTFs by the direct binding of the NPs to the enzyme. The most common mechanism of antibacterial activity of NPs is the over-production of reactive oxygen species (ROS) [65]. Studies have revealed that AgNPs show toxic behavior against mitochondria and generate reactive oxygen species (ROS) [66].

### **2-7-1: Silver Nanoparticles**

Silver is a chemical element, its color is silver, its atomic number is 47, it is a good conductor of heat and electricity, its atomic weight is 107.868, and its specific weight is 10.5. Silver melts at 962 degrees Celsius. Silver nanoparticles attracted tremendous interest in the biomedical field, thanks to their attractive and unique nano-related properties, including their high intrinsic antimicrobial efficiency [67]. The possible antimicrobial mechanism of silver ions, silver nanoparticles, and silver-based nanocomposites on the microbes has been suggested according to the morphological and structural changes found in the bacterial cells [68]. The antimicrobial activity of silver nanoparticles (AgNPs) can be explained through various mechanisms. AgNPs

have a high affinity to sulfur and phosphorus, which are key elements in its antibacterial action. AgNPs can interact with sulfur-containing proteins, which are highly abundant on bacterial cell membranes. This interaction increases the membrane's permeability, which compromises bacterial cell viability [69]. Another mechanism is believed to be that silver ions interact with three main components of the bacterial cell to produce the bactericidal effect: (i) the peptidoglycan cell wall and the plasma membrane, causing cell lysis; (ii) the bacterial (cytoplasmic) DNA, preventing DNA replication; and (iii) the bacterial proteins, disrupting protein synthesis [70]. The small particle size of AgNPs allows them to penetrate cell membranes, causing DNA damage and cell death, even in biofilm organisms that are typically resistant to antibacterial agents [71]. Ag-NPs are also known to generate ROS after they enter a cell. An increase in ROS levels results in a significant drop in GSH level and a concomitant increase in LDH in the medium, ultimately inducing apoptosis [72]. Previous studies have shown that the antibacterial activity of AgNPs is proportional to their specific surface area and size, with smaller particles displaying greater antibacterial activity. This size-dependent efficacy is due to their high surface area to volume ratio, which allows efficient bonding with the bacterial surface [73]. They hypothesized that the effect of morphology on the antibacterial activity of AgNPs was due to the specific surface areas and the reactivity of the facets; AgNPs with larger effective contact areas and more reactive facets exhibit greater antibacterial activity (X Hong et.al 2016) [74].

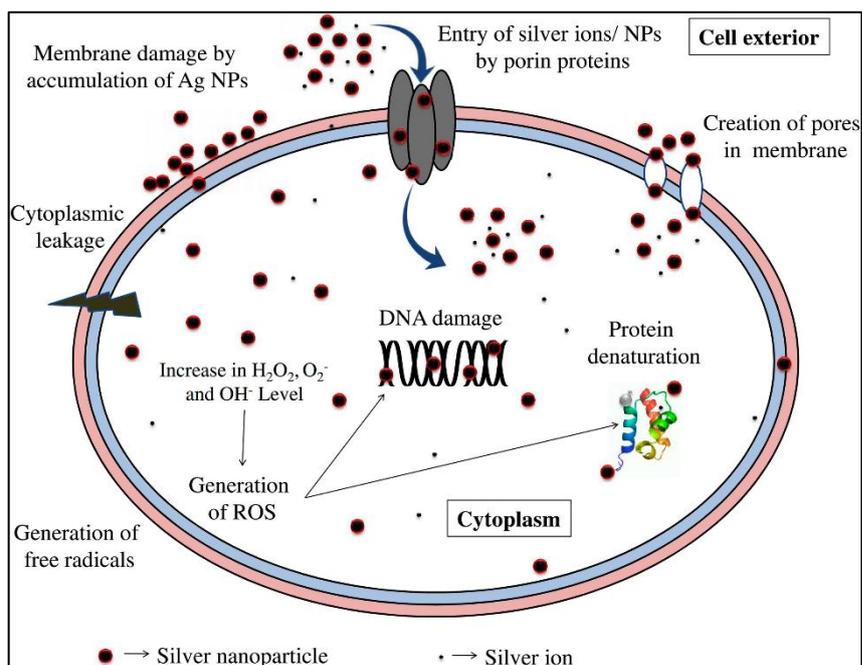


Figure (2-4): The effect of silver nanoparticles and silver ions on bacteria  
[75]

### 2-7-2: Gold nanoparticles

Gold is a bright yellow metallic chemical element with an atomic number of 79 and an atomic weight of 196.967. It melts at a temperature of 1064 degrees Celsius and boils at 2856 degrees Celsius. Typically defined as particles of 1–100 nm in size, which is in the sub-wavelength regime of visible light. It is a good conductor of heat and electricity. Nanoparticles (NPs), particularly gold nanoparticles (AuNP), have been widely used as nanocarriers and diagnostic agents due to their optical properties and ease of synthesis of a wide range of sizes and shapes. The physical and chemical properties of AuNP, such as the size, surface charge, and hydrophobicity of the particle coating, can influence the nanoparticle compatibility with cellular immune responses, such as cytokine productions, reactive oxygen species (ROS) production, transport processes, cellular damage, and cellular functions [76]. The size of gold nanoparticles remains a key parameter that dominates their properties. Since particle size controls endocytosis effectiveness, cellular

localization and accumulation sites *in vivo*, it is natural to understand that the cytotoxicity of gold materials depends on particle size [77]. The strong absorption, efficient light/heat conversion, and high photostability, contribute to arousing increasing interest in the photothermal applications of gold nanoparticles that permit a directional control of the incident radiation on the administration region of these phototransducers, resulting in localized heat transfer to the surrounding environment [78]. It is logical to state that the binding of gold nanoparticles to the bacteria depends on the surface area available for the interaction. Nanoparticles have large surface area available for interactions which enhances bactericidal effect than the large sized particles; hence they impart cytotoxicity to the microorganism [79].

## **2-8: Bacteria**

Bacteria are distinct living entities whose structural organization differs from that of all other microorganisms. Bacteria have unique structural organization, with the prokaryotic cell being the organizational unit. They lack nuclear membranes and nuclear material distribution, and lack mitochondria. The prokaryotic cell has minimal internal detail and minimal change in appearance [80]. In fact, however, bacteria come in a wide variety of shapes and sizes, called the morphology of the organism. The most common shapes are rod-like, called the bacillus (plural, bacilli) form, or spherical, called coccus (plural, cocci) form [81]. Gram's stain is a staining technique used to classify bacteria based on the different characteristic of their cell walls [82]. The cell membrane of bacteria is the part in contact with the environment and is therefore susceptible to play a significant role on bacteria adsorption capacity. Gram-positive and -negative bacteria have been shown to differ considerably in the structure of their cell membrane [83]. Gram-negative bacteria are surrounded by a thin peptidoglycan cell wall, which itself is surrounded by an outer membrane

containing lipopolysaccharide. Gram-positive bacteria lack an outer membrane but are surrounded by layers of peptidoglycan many times thicker than is found in the Gram-negatives [84].

Multidrug resistance MDR is a condition in which microbes are resistant to more than one antibiotic. The reason behind this is sequential use of the same antibiotics against diseases for years. Antibiotic resistance occurs when bacteria, fungi, viruses, or protozoa cannot be fully inhibited or killed by antibiotics [85]. Many strategies have been introduced to counter the menace of MDR bacteria in recent times and one such approach is the use of nanotechnology-developed novel nanomaterials that have different shapes and sizes and possess broad-spectrum antimicrobial action [86].

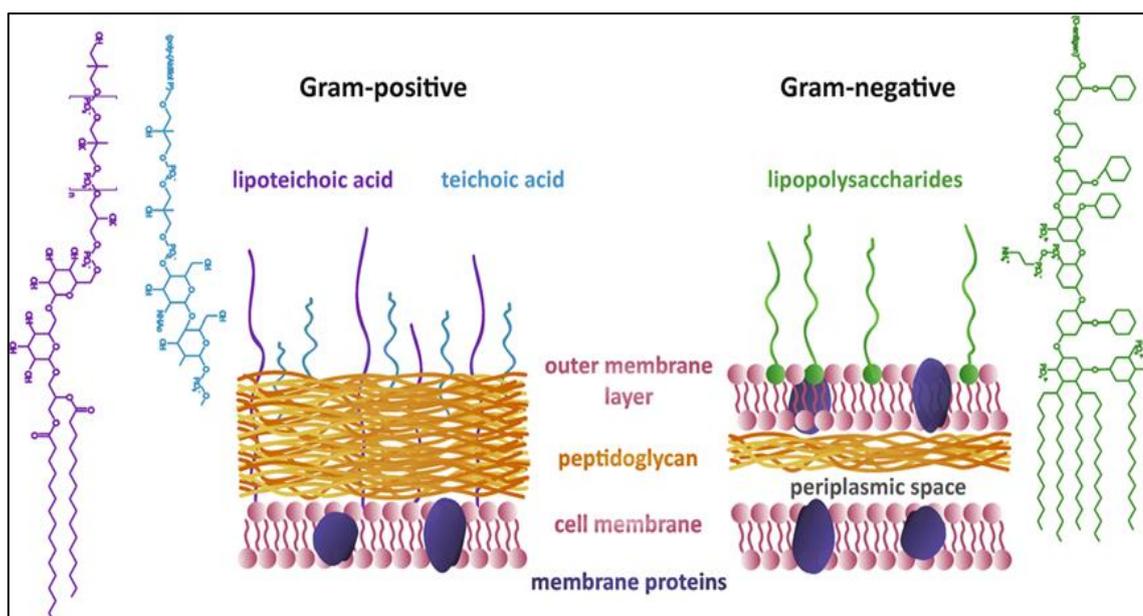


Figure (2-5): Differences between Gram-positive and Gram negative bacterial cell walls [83].

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**2-8-1: *Escherichia coli* bacteria**

*E. coli* is a gram-negative, non-sporulating, rod-shaped, facultative anaerobic, and coliform bacterium pertaining to the genus *Escherichia* that commonly inhabits the environment, foods, and warm-blooded animals' lower gut. Cells are typically rod-shaped, with  $1\text{--}3\ \mu\text{m} \times 0.4\text{--}0.7\ \mu\text{m}$  (micrometer) in size around  $1\ \mu\text{m}$  long,  $0.35\ \mu\text{m}$  wide, and  $0.6\text{--}0.7\ \mu\text{m}$  in volume. The optimal growth of *E. coli* occurs at  $37^\circ\text{C}$  ( $98^\circ\text{F}$ ) but some laboratory strains can multiply at temperatures of up to  $49^\circ\text{C}$  ( $120.2^\circ\text{F}$ ). It takes as little as 20 min to reproduce in favorable conditions [88]. *E. coli* is a facultative aerobe that can activate or repress metabolic enzymes based on oxygen levels. It can colonize the human infant intestine due to its ability to metabolize oxygen or ferment. *E. coli* is closely related to pathogens like *Salmonella*, *Klebsiella*, *Serratia*, and *Yersinia pestis*. Although mostly harmless, pathogenicity islands have been identified in *E. coli*, resulting in strains colonizing different tissues. Its building blocks include 55% protein, 25% nucleic acids, 9% lipids, 6% cell wall, 2.5% glycogen, and 3% other metabolites [89]. Generate a wide range of infections (mainly urinary and gastrointestinal, but also systemic, meningeal or lung infections) [90]. The *E. coli* cell envelope comprises a cytoplasmic membrane, peptidoglycan layer, outer membrane, capsular layer, slime-layer, extracellular polymeric substances, pilli, fimbriae, and flagella [91].

**2-8-2: *Streptococcus pneumonia* bacteria**

*Streptococcus* is a genus of spheroidal bacteria belonging to the family Streptococcaceae. Streptococci are microbiologically characterized as Gram positive and nonmotile. *Streptococcus* contains a variety of species, some of which cause disease in humans and animals, while others are important in the manufacture of certain fermented products [92]. The pneumococcal cell wall has peptidoglycan and teichoic acid, similar to other streptococci. The

capsular polysaccharide is covalently bound to the peptidoglycan and to the cell wall polysaccharide. *S pneumoniae* (pneumococci) is a member of the *S mitis* group. Pneumococci are readily lysed by surface-active agents, which probably remove or inactivate the inhibitors of cell wall autolysins. Pneumococci are normal inhabitants of the upper respiratory tract of 5–40% of humans and can cause pneumonia [93]. *Streptococcus pneumoniae* is a major causative agent of human diseases, which include chronic otitis media, sinusitis, pneumonia, septicemia, and meningitis [94].

### **2-9: The Physiological State of the Bacteria.**

The normal bacterial growth curve has four stages: lag phase, log (exponential) Phase, stationary phase, and death phase [95]. The killing mechanism is affected by the physiological state of the bacteria, such as the growth phase. The first phase observed under batch conditions is the lag phase in which the growth rate is essentially zero. When an inoculum is placed into fresh medium, growth begins after a period called the lag phase, the second phase of growth observed in a batch system, is the exponential phase. The exponential phase is characterized by a period of exponential growth—the most rapid growth possible under the conditions present. During exponential growth, the rate of increase of cells in the culture is proportional to the number of cells present at any particular time; the third phase of growth is the stationary phase. The stationary phase in a batch culture can be defined, as a state of no net growth, the final phase of the growth curve is the death phase, which is characterized by a net loss of cultural cells. During death, cells metabolize and divide, but more viable cells are lost than gained, resulting in a net loss of viable cells [96].

# **Chapter Three**

## **Materials and**

## **Methods**

### 3-1: Introduction

This chapter includes a description of the stages of the practical part of the research, in terms of describing the materials and tools used to prepare the samples, the devices used to prepare the nanoparticles, measurement methods, and the effect of the preparation conditions (laser energy, number of pulses) on the structural and optical properties of the nanoparticles, and then conducting tests on those samples.

### 3-2: Devices, materials and tools used in research

#### 3-2-1: Devices

Table (3-1) shows the devices used

The device name	Country of origin
Pulsed Nd-YAG laser	Germany
Diode laser (405 nm)	Germany
Nd:YVO4 laser (532nm)	USA
autoclave	UK
incubator	Spain
fridge	Japan
Elisa reader	Germany
Spectrophotometer (UV-Visible)	England
Transmission electron microscope (TEM)	Germany

**3-2-2: Materials**

Table (3-2) shows the materials used

<b>Materials</b>	<b>Name of origin</b>
Silver nanoparticles	It was prepared at the College of University of Babylon
Gold nanoparticles	It was prepared at the College of University of Babylon
Mueller Hinton Agar	India
Distilled water	College of Science for Women
Barium Chloride	England

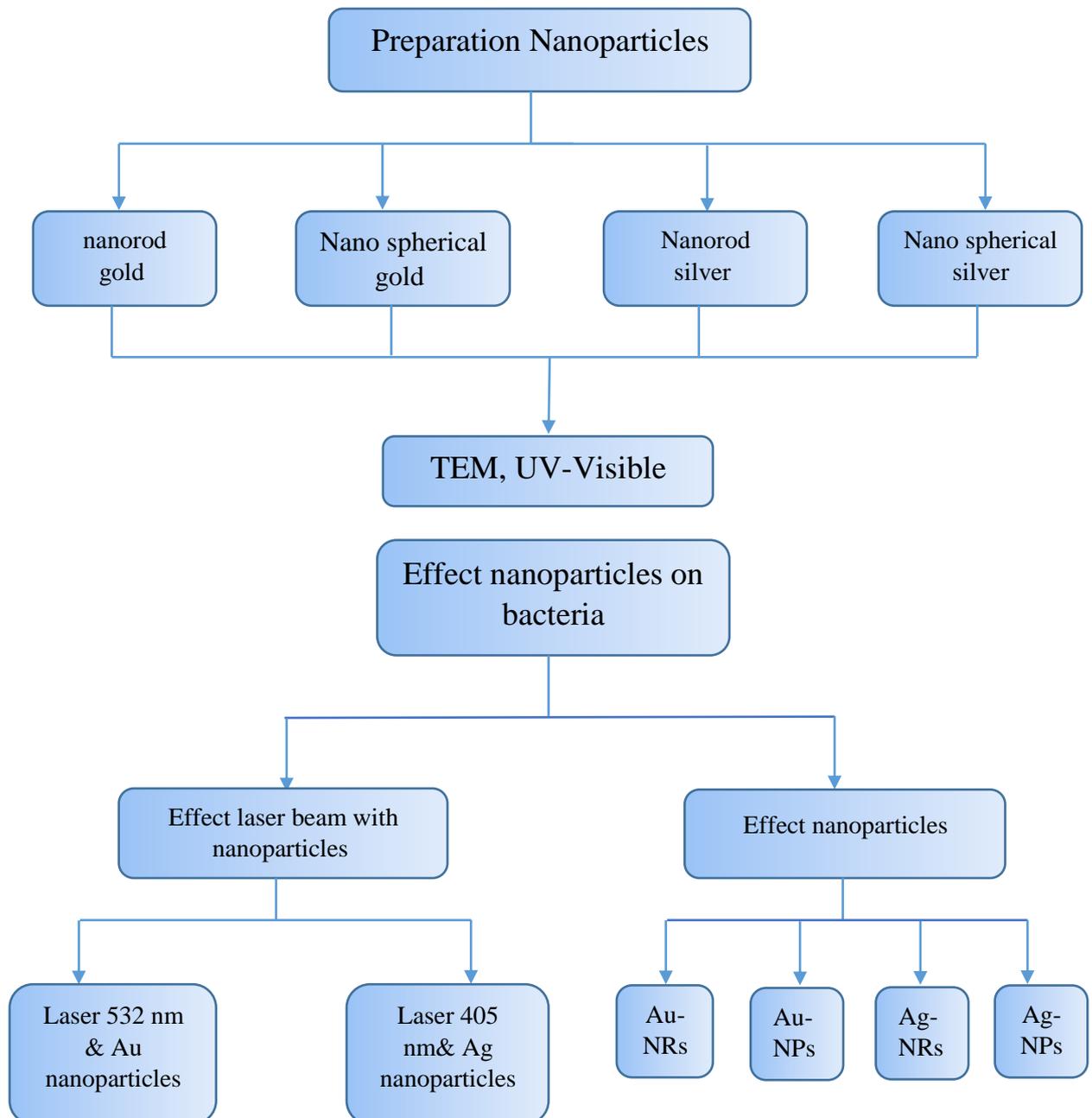
**3-2-3: Tools**

Table (3-3) shows the tools used

<b>Tools</b>
1. Plate
2. Glass baker
3. Glass flasks
4. Glass pipettes
5. Plastic test tubes
6. Cotton
7. Cotton swabs
8. Burner
9. Supports and stands
10. petri dishes
11. Micro Pipet

### 3-3: Methods

The design of study was illustrated in figure (3-1).



### 3.4. Laser Nd:YAG

The laser used is a Q-switch Nd-YAG laser, which produces pulses with a wavelength of 1064 nm, with a maximum energy of 1000 mJ per pulse. The pulse duration is (7) nanoseconds, the repetition rate is (6) Hz, and the effective beam diameter is (5) mm. It is used for laser ablation, and the lens used is with a focal length of (15.3 cm), to achieve a high laser.



Figure (3-1): Shows an image of a Q-Switch Nd-YAG laser

### 3.5. Preparation Nanoparticles

#### 3-5-1: Spherical Gold Nanoparticles and Silver Nanoparticles

Two pieces of gold and silver of high purity were used as the metal target. Then the two pieces are placed at the bottom of the quartz cell (both separately) containing (15) mL of the double ionized water solution. The Q-switch Nd: YAG laser with 80 mJ energy was ablated on the pure gold and silver plate to produce NPs of them. The laser wavelength, repetition rate, and ablation time were

adjusted in 1064 nm wavelength, 10 Hz, and 4, 3 and 2 mins. The laser beam was focused on the gold target after reflecting from a mirror and passing through a convex lens ( $f=100\text{mm}$ ). In addition, it is stirred during the ablation of the targets at room temperature. Three different concentrations, 50, 25 and 12.5%, were obtained because of irradiation times of 4, 3 and 2 min, respectively, It was prepared in Iran.

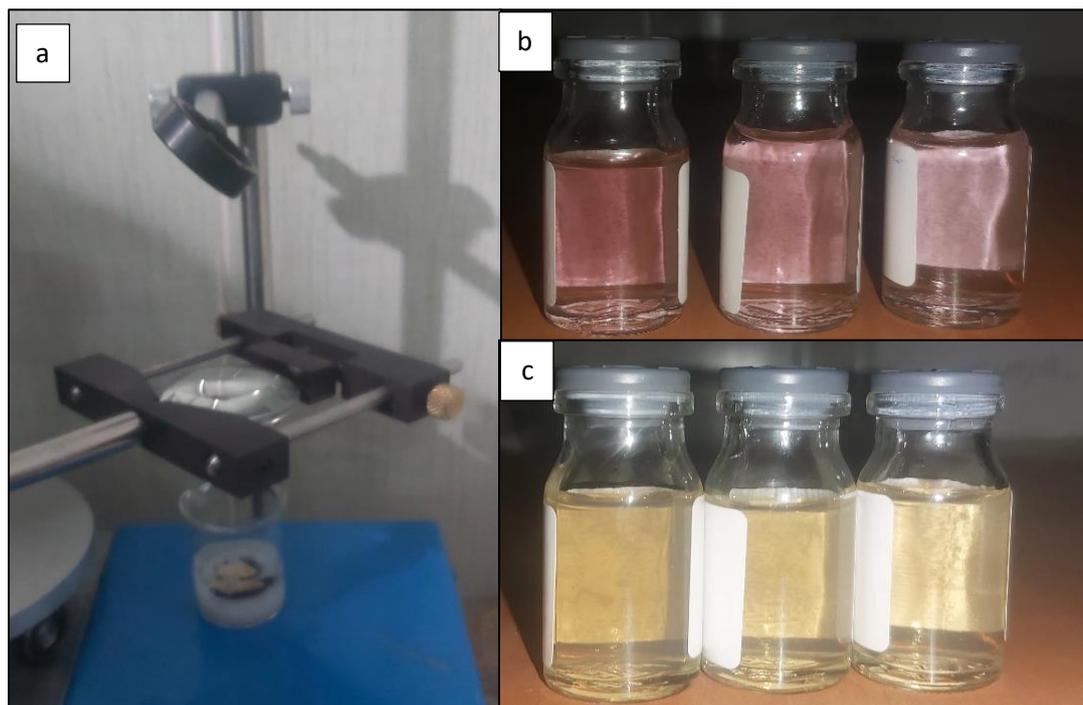


Figure (3-2): (a) Photograph of nanoparticle preparation. (b) Gold NPs. (c) Silver NPs

### 3-5-2: Gold nanorods and silver nanorods

The nanorods were manufactured using laser ablation. The difference between the production of nanorods and nanoparticles is that during production, an electrical voltage was applied to the sample that was hit by the laser. The voltage applied by two electrodes to the target sample was about 10 volts and the power of the first harmonic of the laser was 1064 nm, 100 mJ. The sample production process is shown schematically below, it was prepared in Iran.

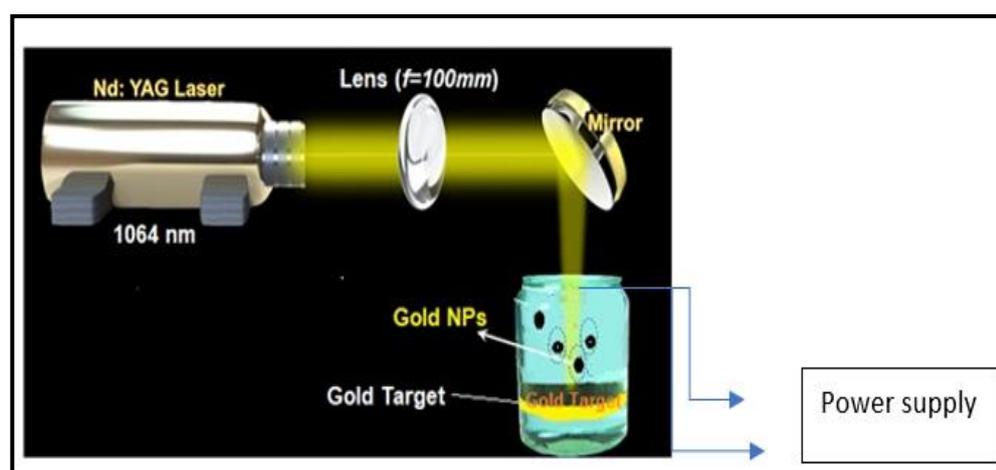


Figure (3-3): Liquid Ablation Laser experimental step to produce nanorods.

### 3-5-3: Preparation of gold and silver nanoparticles.

Two pieces of gold and silver with a high purity of 99.9% were used, used as a metal target. The two metal pieces were polished using paper to remove impurities, after which they were washed with distilled water. Then the two pieces are placed using tweezers at the bottom of the quartz cell (both separately) containing (4) mL of distilled water. The laser ablation process was carried out in aqueous media using a laser (Q-switched Nd:YAG) (Sigma Aldrich, Germany) of wavelength 1064 nm, at a repetition rate of (4) Hz, with a pulse width of (10) nanoseconds, and a pulse energy of (100 mJ), for a number of pulses (250, 500, and 750) pulses. It was prepared at the College of Science for women, University of Babylon

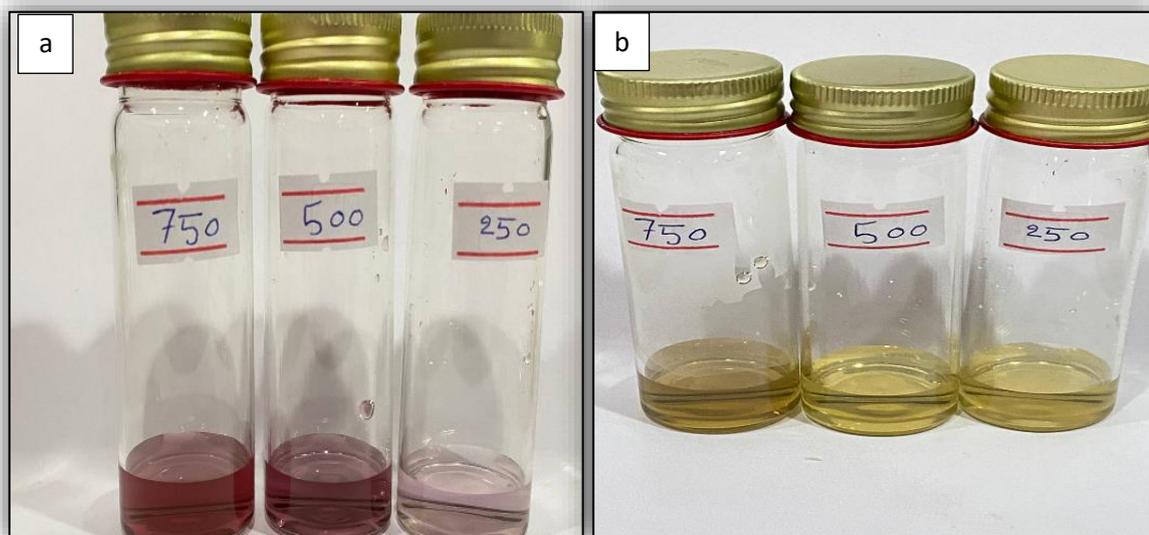


Figure (3-4): Illustration of the prepared nanoparticles (a) gold nanoparticles, (b) silver nanoparticles

### 3-6: Measurement Devices

#### 3-6-1: Absorption Spectrometry

This spectrometer covers a wide range of the electromagnetic spectrum from the ultraviolet region to the near infrared region. The device includes two sources for consultation as:

1. Deuterium lamp (190-360 nm) Deuterium lamp
2. Tungsten lamp (360-1100 nm) Tungsten lamp

Of CECIL CE 7200 origin, made in England. All resistive spectra were measured at room temperature in a quartz cell in the Advanced Laser Laboratory at the College of Science for Girls, University of Babylon, as stated in Figure (3-5).



Figure (3- 5): Absorption testing device

### 3-6-2: Transmission Electron Microscope

It uses a beam of electrons that passes through an ultra-thin sample, where the process of interaction occurs between them during their passage, which leads to the appearance of an image that undergoes a process of amplification and focus on imaging devices such as fluorescent screens or on photographic film. Samples of NPs were characterized using a transmission electron microscope (Model: LEO 912 AB (Made in Germany) the examination process takes two hours. A drop of the NPs solution is placed on a copper grid covered with an amorphous carbon film. The drop is dried with an infrared lamp (Philips-100w) until all the solvent evaporates. The carbon grid is then carried for examination. It was prepared in (Magneto Plasmonic Lab, Shahid Beheshti University, Iran.) Figure (3-4) shows the transmission electron microscope.

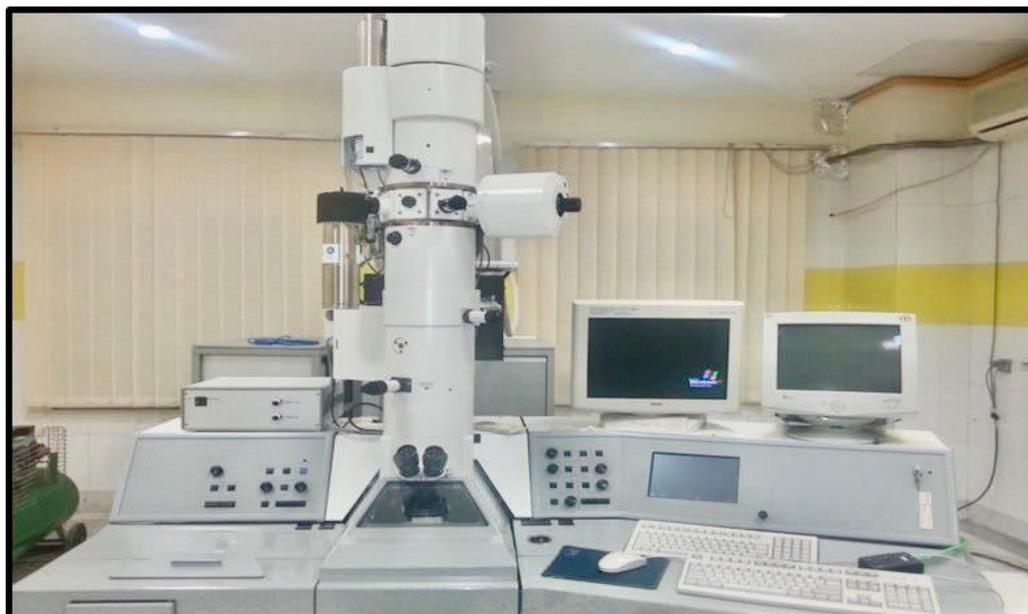


Figure (3-6) a photograph of a transmission electron microscope (TEM).

### 3-7: Irradiation by Laser

A laser of with a wavelength of (532 nm) (SHG) and a power of (8 mw) was used. This laser is being tested in the field of bacterial killing in the presence of nanoparticles.

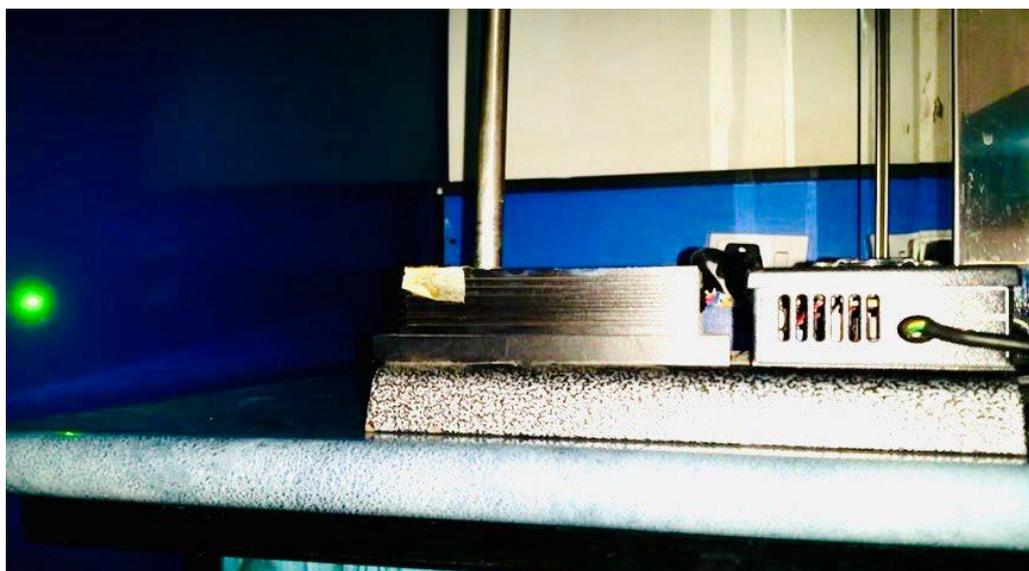


Figure (3-7) Nd:YVO4 laser device

A blue diode laser with a wavelength of 405 nm and a power of (10 milliwatts) was used. This laser is being tested in the field of killing bacteria alone and in the presence of dispersed silver particles.

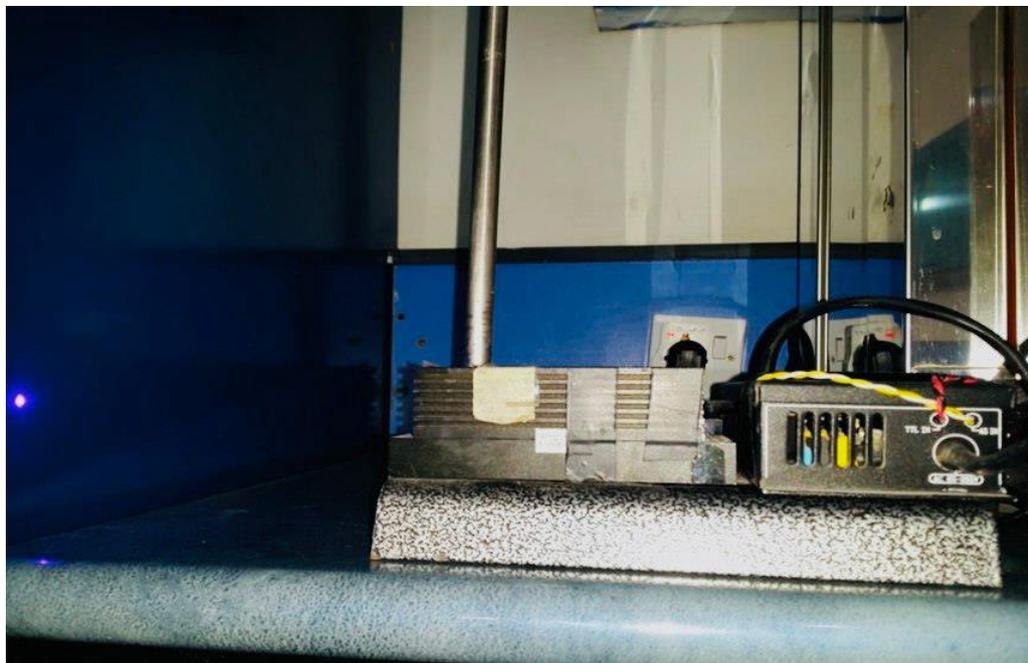


Figure (3-8) Diode laser with a wavelength of (405 nm)

### **3-8: Effect gold nanoparticles or Silver Nanoparticles on bacterial growth**

The broth bacteria are instilled to a volume of 0.1 mL in five wells. Gold nanoparticles or silver nanoparticles are added in cells in a amount 0, 0.05, 0.1, 0.5, 1 mL. Then incubation these solutions at 37°C for 24 hours. After incubation at 37°C for 24 hours, the bacteria were counted.

### **3-9: Effect gold nanorods or Silver nanorod on bacterial growth**

The broth bacteria are instilled to a volume of 0.1 mL in five wells. Gold nanorod or silver nanorod are added in wells in amount of 0, 0.1, 1 mL. Then incubation these solutions at 37°C for 24 hours. After incubation of 37°C for 24 hours counting the bacteria.

**3-10: Effect of gold nanoparticles and laser on bacterial growth**

Use a laser based on the absorption peak of the nanostructures to obtain the killing efficiency. due to the absorption peak of gold nanoparticles at 532 nm, use green laser. Bacteria are added to a volume of 0.1 mL in 5 wells. Gold nanoparticles or silver nanoparticles are added in wells in concentrations of 0, 0.05, 0.1, 0.5, 1 mL. Then each cell is hit by a laser for 4 minutes. Then incubation these solutions at 37°C for 24 hours, Then counting the bacteria.

**3-11: Effect of gold nanorods & Silver nanorod and laser on bacterial growth**

Use a laser based on the absorption peak of the nanostructures to obtain the killing efficiency. Due t the absorption peak of gold nanoparticles at 532 nm, use green laser. Bacteria are added to a volume of 0.1 mL in 3 wells. Gold nanoparticles or silver nanoparticles are added in wells in concentrations of 0, 0.1, 1 mL. Then each wells is hit by a laser for 4 minutes. Then incubation these solutions at 37°C for 24 hours, Then counting the bacteria.

**3-12: Counting method**

The first stage of serial dilution preparation, a liquid sample was diluted with a dilution factor of 0.1 (1 ml of the first tube was added to 9 ml of the solution in the second tube). 0.1 ml was taken from each prepared tube (serial dilution) and transferred to the Mueller Hinton Agar solid culture center. Next to the flame, using a sterile swab, the whole sample was spread on the culture medium.

The cultures were placed in an incubator at 37 degrees Celsius for 24 hours. After 24 hours, the number of colonies in the samples was counted. Then the number of colonies in each plate was counted and by multiplying by the dilution factor, the number of colonies was obtained in CFU units.

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**3-13: Effect of silver nanoparticles & Gold Nanoparticles on bacteria (*Streptococcus, Escherichia coli*) in solid media (disk method)**

Dissolve (3.8 g) of Mueller Hinton agar powder in (100) mL of distilled water in a glass beaker and shaken well so that all the powder dissolved and became completely liquid, then placed in the autoclave at a temperature of (121C°) for (15) minutes. Then it is poured into the dish and left for 5 minutes to solidify, after which the active bacteria are placed in a horizontal and vertical schematic form to cover all parts of the dish. After that, we take sterile filter paper with a diameter of (5 mm), dipped in the nanomaterial and added to the dish, then kept in the incubator at 37 degrees for a period of 24 hours. We will notice the results of the effect of the silver nanoparticle in killing and inhibiting bacteria.

**3-14: Effect of gold nanoparticles & Silver Nanoparticles on the growth of *Escherichia coli* and *Streptococcus pneumonia* bacteria.**

Transfer 100 microliters of a bacterial suspension of approximately 0.5 McFarland to each hole. After that, the nanomaterial is added. Read by elisa device and record the spectral absorbance when zero minutes . The plate was placed incubation at 37 °C and the spectral absorbance was read after 24 hours. The readings are sequenced, bacterial growth is extracted, and inhibition determined.

**3-15: Effect of laser with Silver & gold nanoparticles on the growth of *Escherichia coli* and *Streptococcus pneumonia* bacteria.**

Transfer 100 µl of approximately 0.5 McFarland bacterial suspension to each well After that, the nanomaterial is added, and then the irradiation process takes place at different times (1, 3, 5) minutes as shown in Figure (3-9).

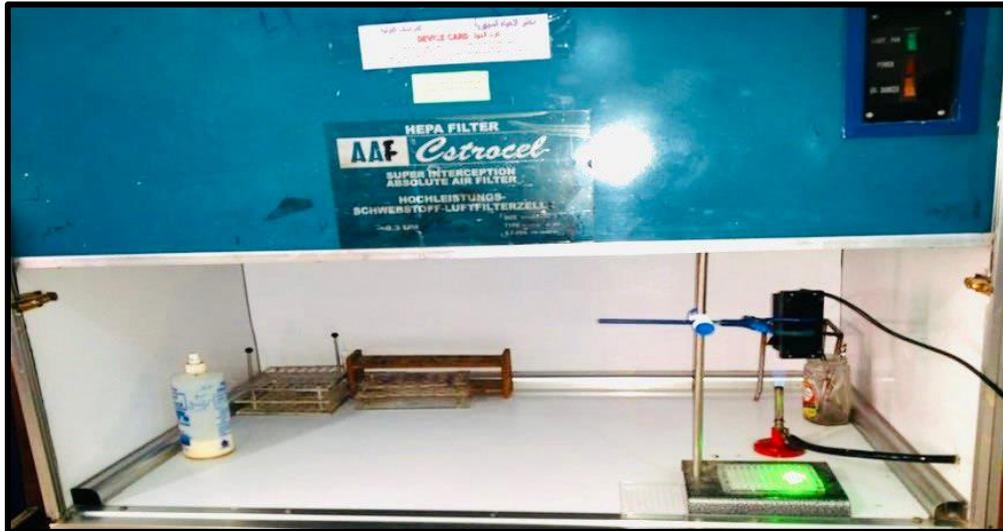


Figure (3-9):Irradiation of bacteria using laser.

The reading is then done with an ELISA device and the spectral absorbance is recorded at zero. The plate was placed in incubation at 37°C and the spectral absorbance was read after 24 h. The reads are sequenced, bacterial growth is extracted, and inhibition is determined.

# **Chapter Four**

## **Results and Discussion**

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## 4-1: Introduction

In this chapter, the practical results presented and discussed through knowing the effect of some physical parameters on the vital activities of bacteria (*Escherichia coli*, *Streptococcus*), whether with silver and gold nanoparticles alone, or the presence of a laser beam with them.

## 4-2: Optical Properties

### 4-2-1: Spectral Absorbance

The absorption spectrum for gold nanoparticles represent in figure (4-1) peak maximum in water, using the wavelength (1064 nm). When the laser pulses fall on the surface of the metal immersed in (15 ml) of water, it was observed that the color of the solution changes with the increase time of irradiation, this lead to increase the concentration of nanoparticles. To have three concentrations of nanoparticles, 50, 25, and 12.5%, were obtained using 4, 3 and 2 minutes for laser irradiation time.

Figure (4-2) Nanorods have two exact resonant absorption peaks, the first for the transverse mode and secondy for the longitudinal mode. The transverse mode remains at a fixed wavelength, while the longitudinal mode can be tuned across a wide range of spectrum. This results in a broader absorption spectrum result from spherical nanoparticles. Gold and silver nanorods can exhibit two Plasmon modes, longitudinal mode (LM) and transverse mode (TM), in the visible and near-infrared regions due to the oscillations of the free electrons along and perpendicular to the major axis of the nanorods [97].

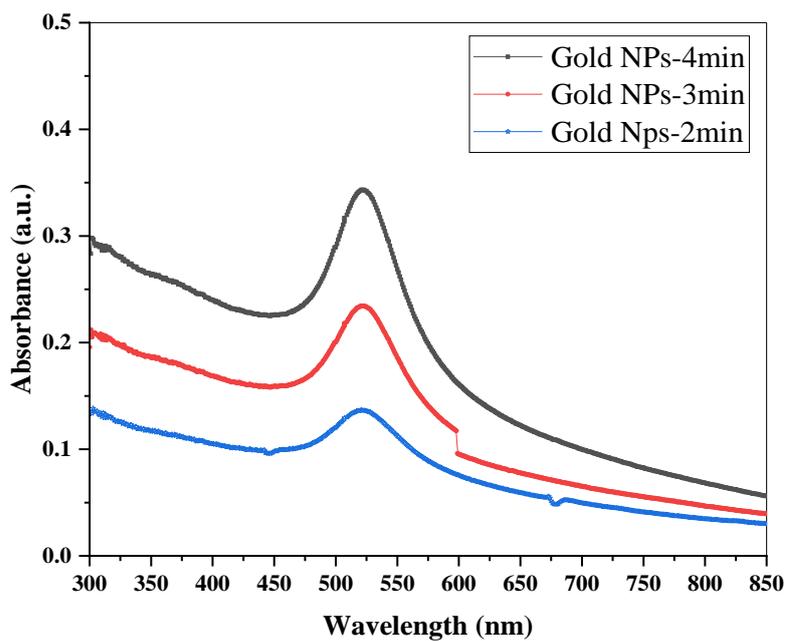


Figure (4-1): Absorption spectrum of spherical gold nanoparticles with different irradiation times.

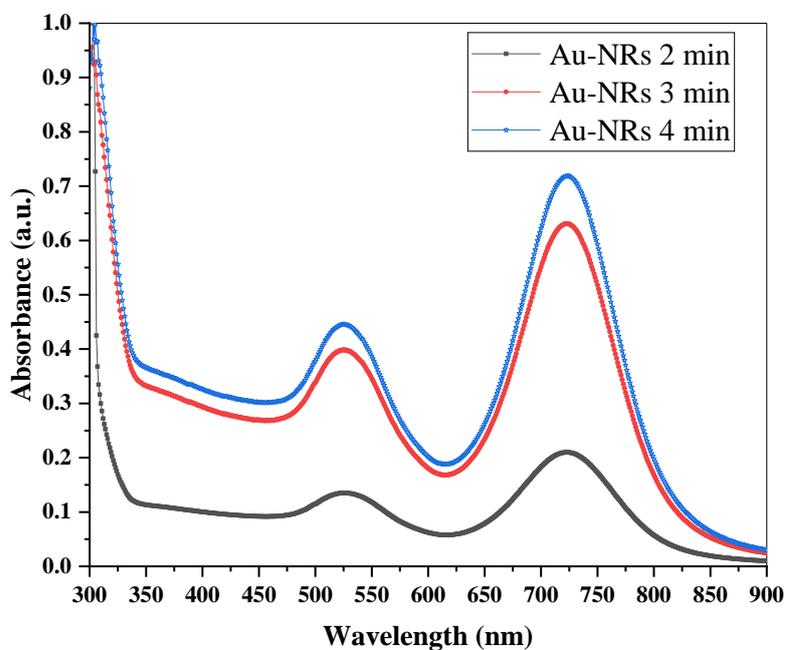


Figure (4-2): Absorption spectrum of gold nanorods with different irradiation times.

Figure (4-3) it is due to the reason that the spectrum of nanorods is broader than that of nanoparticles is due to their different shapes and sizes, which leads to a difference in the characteristic of plasmon the absorption spectrum of nanorods is broader than spherical nanoparticles. The main reason for this difference lies in the shape and aspect ratio of these nanostructures.

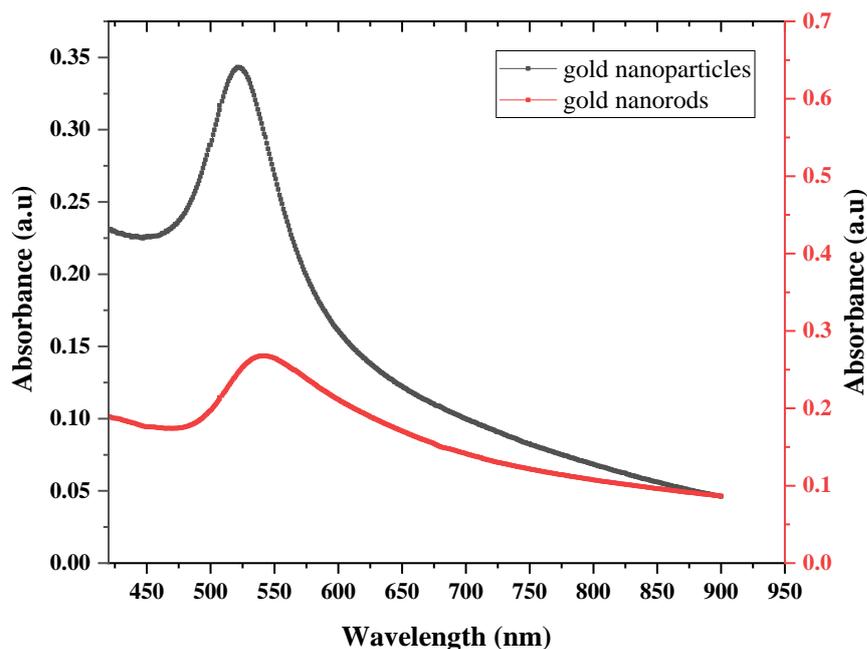


Figure (4-3) comparison of the absorption spectrum of spherical gold nanoparticles and gold nanorods at 50% concentration.

The figure (4-4) shows the change of the absorbance spectra of Ag-NPs aqueous solution as a function of wavelength, and where the absorption peak of Ag-NPs was at (403, 408, 413 nm) nm corresponding to the laser irradiation times (2, 3, 4 minutes), three concentrations were obtained, which are (12.5, 25 and 50%), respectively. The reason for the appearance of these peaks is due to increase in the production rate of nanoparticles with increasing irradiation time, which leads to an increase in the concentration of nanoparticles and thus changing the color of the liquid to darker.

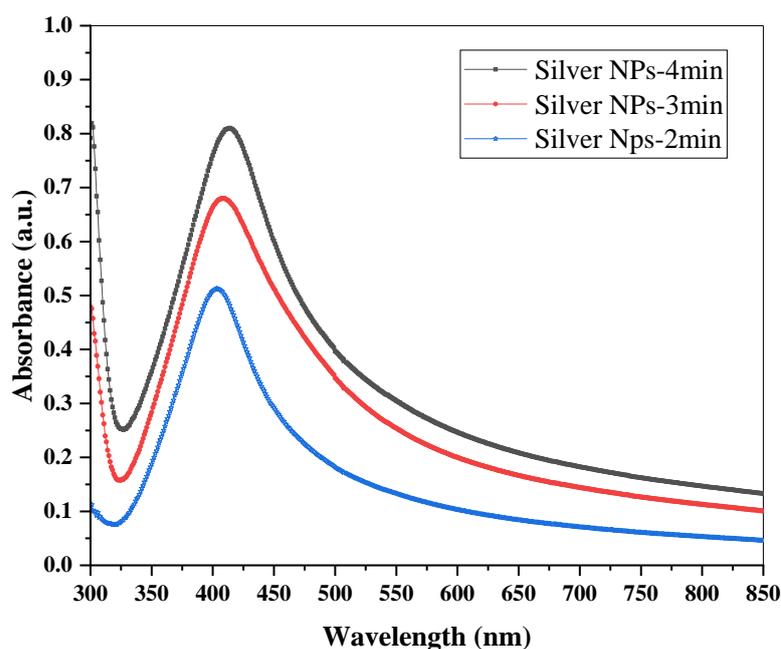


Figure (4-4): Absorption spectrum of spherical silver nanoparticles with different irradiation times.

Figure (4-5) show the reason that the spectrum of nanorods is broader than that of nanoparticles due to their different shapes and sizes, which affect their plasmonic properties. Silver nanoparticles have a spherical shape and smaller size, which products a narrow plasmon resonance peak in the spectrum. The reason belong to that, the electrons in the nanoparticles are confined to a smaller volume. In contrast, silver nanorods have a more elongated shape and larger size, which leads to a broader Plasmon resonance peak in the spectrum. From a spectroscopic view, this case may be happen that the electron has large area to move inside the nano.

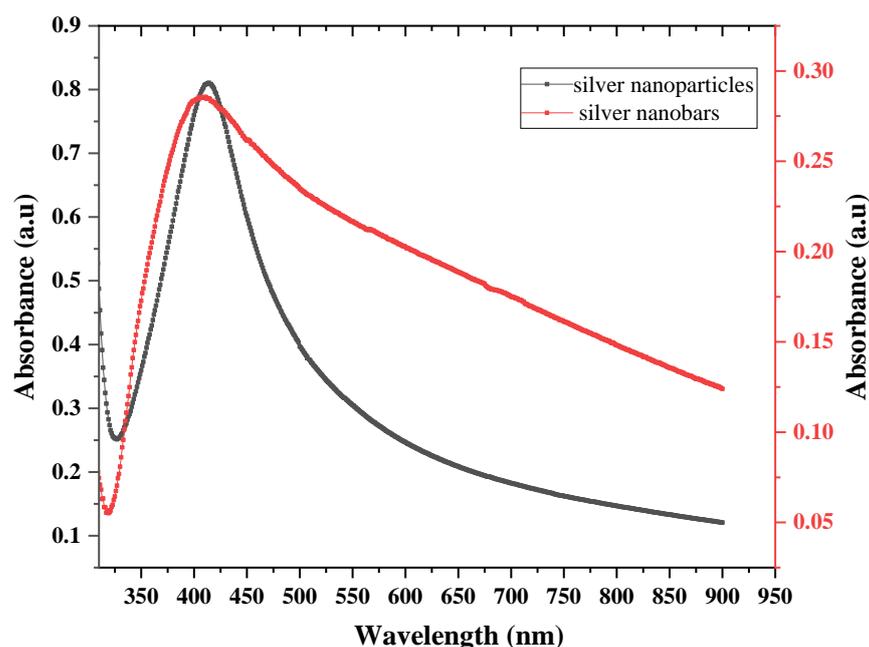


Figure (4-5) comparison of the absorption spectrum of silver nanoparticles and nanorods

Figure (4-6) shows the absorption spectrum of a solution of silver nanoparticles in water using the wavelength (1064 nm). When laser pulses fall on the surface of a metal immersed in 4 ml of water. The spectral absorbance increases with the number of pulses, which leads to an increase in the concentration of nanoparticles and thus an increase in the color intensity of the solution. As for the color of gold nanoparticles, it is purple, as shown in Figure (4-7). When the number of laser pulses increases, the absorbance increases, and thus the concentration of nanoparticles increases. Variation in Pulse Counts (250, 500, 750): Changing the number of laser pulses can impact the size, distribution, and characteristics of the synthesized nanoparticles. Typically, a higher number of pulses may result in more material being ablated, leading to a larger quantity of nanoparticles.

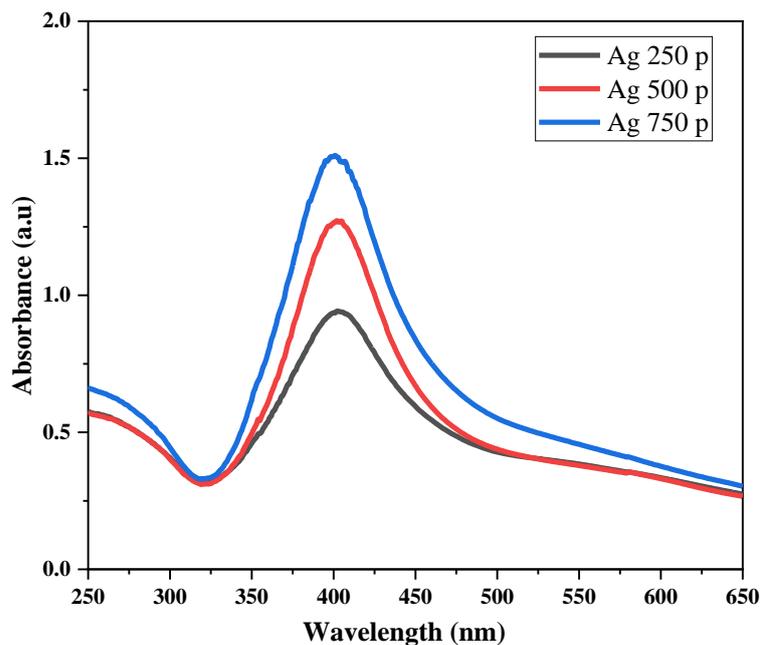


Figure (4-6) shows the absorbance spectrum of Ag-NPs when using different number of pulses.

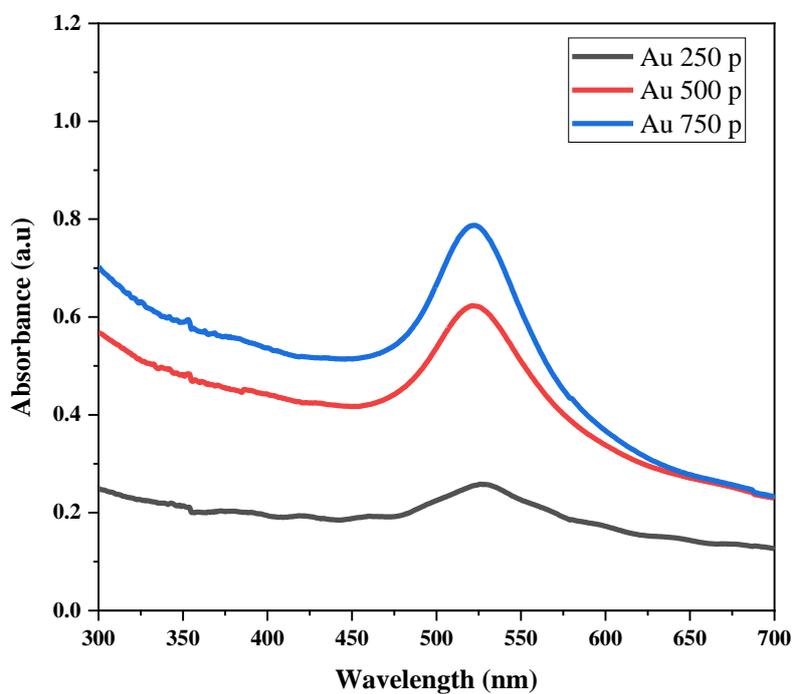


Figure (4-7) shows the absorbance spectrum of Au-NPs when using different number of pulses.

#### 4-2-2: Transmission Electronic Microscopy

Figure (4-8.a) depicts TEM image of Ag NPs prepared at 80 mJ laser energy for ablation time of 4 min. Shows the spherical shape of the nano-silver material which has apparent granular size of different dimensions, small and spherical. Such results indicate the absence of coalescence of the nano atoms; therefore, they appeared in a very clear spherical shape, as their dimensions are (19 nanometers). Figure (4-8.b) shows the size and shape of the nano-rods obtained by applying a voltage of about 10 V to the sample, using a laser energy of 100 mJ. Indicates the clear coalescence of the nano atoms, as the image appeared in the form of a rod and very small dimensions, showing the state of plasmon resonance in a form. It is better, as the nanometer dimension is estimated according to the apparent measurement below (15 nm).

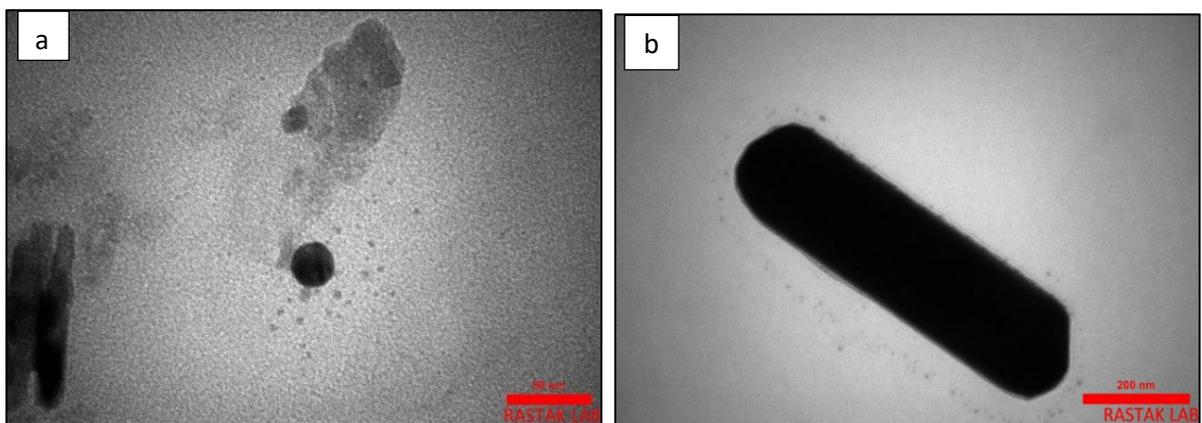


Figure (4-8) The TEM image (a) Ag Nanoparticles (b) Ag Nanorods

Figure (4-9.a) shows the TEM analysis of the shape of the obtained particles at 80 mJ laser energy with an ablation time of 4 min. It was found that the gold nanoparticles produced by laser ablation were spherical in shape. Figure (4-9.b) shows the shape of the nanorods obtained by applying a voltage of about 10 V to the sample, using a laser energy of 100 mJ.

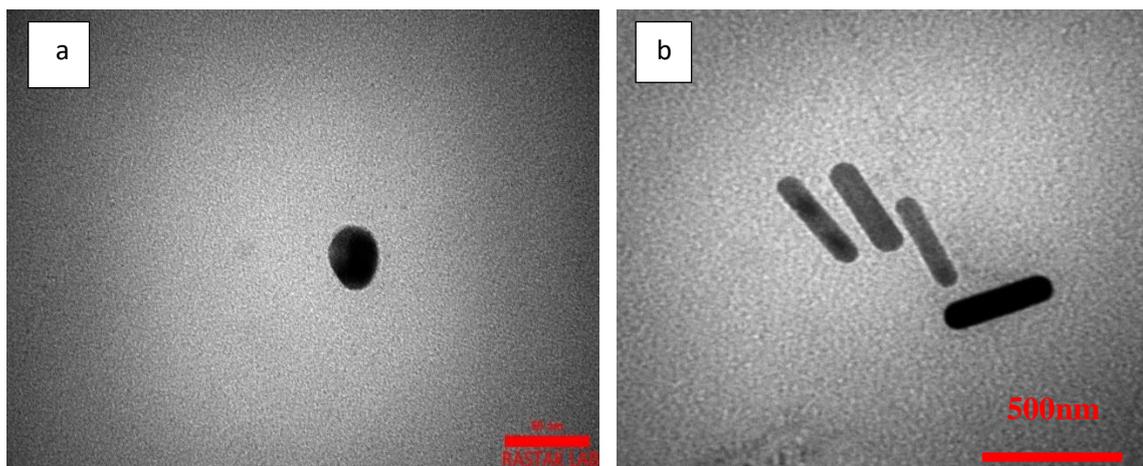


Figure (4-9) The TEM image (a) Au Nanoparticles (b) Au Nanorods

### **4-3: Effect of Silver Spherical Nanoparticles with and without Irradiation Laser on bacteria *Escherichia coli***

It is clearly from table (4-1) that increasing the amount of non-irradiated spherical Ag-NPs to decrease of bacterial viability where the number of bacteria was decreased from  $2.74 \times 10^8$  CFU/mL to  $1.8 \times 10^8$  CFU/mL at the larger amount (1 mL) as a compared with control group. Furthermore, in all amounts of nanoparticles, there was a decrease in the number of bacterial cells. The highest decrease in the number of bacterial cells was when treated with 1 mL of irradiated Ag-NPs and the number of viability bacteria was  $1.7 \times 10^8$  CFU/mL. The lowest decrease in the number of bacterial cells was when they were exposed to silver nanoparticles at 0.05 mL and laser light. The results indicated that the photoactivated silver nanoparticles' antibacterial activity increased with the amount of Ag-NPs.

From the figures (4-9) (4-10), at an amount of 0.1 mL, the impact of silver nanorods is more effective, and at an amount of 1 mL, nanoparticles are more effective on bacteria. It has been found that rod-shaped silver nanoparticles have stronger antibacterial activity than spherical silver nanoparticles and can be used in smaller quantities. When Ag NPs are distinguished from other trace

metals, hydroxyl radicals are produced. These radicals connect to DNA molecules, which damages vital proteins and leading disordering of the DNA structure. When comparing the results with previous studies. It has been observed that the shape of AgNPs also causes a critical impact on their antimicrobial activity. Plate- and rod-shaped AgNPs showed higher antibacterial activity as compared to spherical AgNPs and thus they could be used in lower concentrations. In fact, it was observed that the bactericidal activity of plate- and rod-shaped AgNPs was favored by the presence of high atom density facets {111}, whereas, due to predominance of {100} facets on spherical AgNPs, the latter showed relatively lesser bactericidal activity [98].

When the bacterial cells are in contact with silver, they incorporate silver ions that can inhibit many cell functions. Silver is a weak acid with a natural behavior to react with a base. The majority of protein cells are composed of sulfur and phosphor, which are weak bases. The action of these NPs over the cells might cause a destruction of the cells. The interaction between DNA containing sulfur and phosphor as majority components and silver NPs interferes with DNA's replication, so in this way total elimination of the bacteria was realized [99]. Previous research has shown that Ag NPs can cause membrane damage in Gram-negative Escherichia coli; it is possible that Ag-NP treatment damaged cellular membranes to increase permeability, or penetrated bacterial cell membranes to cause protein dysfunction [100].

Table (4-1) the effect of Ag-NPs on the survival of E.coli at various concentrations with and without irradiation.

Without laser		With laser
Ag-NPs amount (ml)	Survival viability (CFU/ml)	Survival viability (CFU/ml)
Control	$2.74 \times 10^8$	$2.5 \times 10^8$
0.05	$2.65 \times 10^8$	$2.45 \times 10^8$
0.1	$2.42 \times 10^8$	$2.1 \times 10^8$
0.5	$2.1 \times 10^8$	$1.8 \times 10^8$
1	$1.8 \times 10^8$	$1.7 \times 10^8$

#### 4-4: Effect of Silver Nanorods with and without Irradiation Laser on bacteria *Escherichia coli*

Table (4-2) shows the results of the effect of silver nano-rods alone and the effect of silver nanorods irradiated with a 405 nm laser for 4 minutes of irradiation. We noticed that when adding silver nano-rods alone, the number of live bacteria decreased due to the increase in silver nano-rods, when 1 mL of nano-rods, the number of bacteria was  $1.95 \times 10^8$  CFU/mL. As for the combined treatment with silver nano-rods and laser, we noticed a significant decrease in bacteria compared to the treatment with AgNRs alone. The highest reduction was at 1 mL of the AgNRs and upon laser irradiation, where the number of viable bacteria became  $1.8 \times 10^8$  CFU/mL. Compared to the control group.

The shapes of nano-silver (Ag-NPs, Ag-NRds and Ag-NPls) show different anti-bacterial activity. It was found that surface area of nano-silver shapes are key factor for controlling antimicrobial activity inside of the *S. aureus* and *E. coli* bacteria. Anti-bacterial activity dependent on the shape and size of nanoparticles silver. showed that Ag-NPls, as compared with those of Ag-NRds and Ag-NPs, require a lower concentration to inhibit development of

the *S. mutans* and *E. coli* strains, and this is probably due to the increasing surface area in Ag-NPIs compared with Ag-NRds and Ag-NPs [101].

Table (4-2) Effect of Ag-NRs on the survival of *E.coli* for different amount of Ag-NRs with and without irradiation.

Without laser		With laser
Ag-NRs amount (ml)	Survival viability (CFU/ml)	Survival viability (CFU/ml)
Control	$2.5 \times 10^8$	$2.4 \times 10^8$
0.1	$2.3 \times 10^8$	$1.95 \times 10^8$
1	$2.15 \times 10^8$	$1.8 \times 10^8$

#### **4-5: Effect of gold Spherical Nanoparticles with and without Irradiation Laser on bacteria *Escherichia coli***

Table (4-3) shows the results of adding different quantities of manufactured gold nanoparticles at a concentration of 50% with an irradiation period of 4 minutes, as well as the effect of gold nanoparticles exposed to 532 nm laser light. Where we notice a slight decrease in the number of viable bacteria when increasing the amount of nanoparticles at concentrations of 0.5 and 1 mL of, the number of live bacteria was  $2.3 * 10^8$  CFU/ mL, indicating that there was no noticeable difference in the bactericidal effect. Au-NPs can inhibit bacterial growth and division, and this effect tends to increase with increasing amount of Au-NPs. When the cultures were treated with Au-NPs at concentrations of (0.5, 1) mL followed by photoirradiation for 4 min compared to the control group, decrease in the number of bacterial cells was observed.

The results show that the antibacterial activity of the photoactivated Au-NPs increases with increasing NP amount in the presence of laser light because the correlation between the amount of gold nanoparticles and the rate of bacteria destruction is dose dependent, which leads to an increase in the

concentration of the nanoparticles. The highest decrease in bacterial cell survival was observed upon treatment with 1 mL of photoactivated Au-NPs. More germicidal. The combination of gold nanoparticles and a 532 nm laser can significantly cause bacterial cell death.

When nanoparticles are irradiated, they absorb energy, which is quickly transferred through nonradioactive relaxation into heat and accompanied effects, and eventually leads to irreparable damage. Among the different nanostructures, gold nanoparticles, in different modifications (spherical, rods, shells, etc.), are the most promising candidates for such photothermal (PT) sensitizers because they are the strongest nano absorbers; their absorption coefficient due to profound Plasmon resonance is at least if not more than three orders-of-magnitude greater when compared to other organic photosensitizers for PDT. Gold nanoparticles are photostable, nontoxic, and easily conjugated to antibodies (Abs) or proteins [102]. Local temperature increase is extensively exploited in photothermal therapy, where light is used to induce cellular damage. at different gold nanoparticle concentrations, in the dark and under irradiation. In both cases, the nanoparticles penetrated the bacterial wall, but a different toxic effect was observed; while in the dark we observed an inhibition of bacterial growth of 46%, at the same concentration, under irradiation, we observed a bactericidal effect (99% growth inhibition) [103].

Table (4-3) Effect of Au-NPs on the survival of E.coli for different amount of Au-NPs with and without irradiation.

Without laser		With laser
Au-NPs amount (ml)	E.coli (CFU/mL)	E.coli (CFU/mL)
0 mL	$2.9 \times 10^8$	$2.55 \times 10^8$
0.05 mL	$2.8 \times 10^8$	$2.35 \times 10^8$
0.1 mL	$2.75 \times 10^8$	$2.3 \times 10^8$
0.5 mL	$2.3 \times 10^8$	$2.2 \times 10^8$
1 mL	$2.3 \times 10^8$	$1.8 \times 10^8$

#### 4-6: Effect of gold Nanorods with and without Irradiation Laser on bacteria *Escherichia coli*.

Table (4-4) shows the addition of gold nanorods in different quantities, as well as the effect of laser beam (532 nm) in the presence of gold nanorods on the antibacterial efficiency of *Escherichia coli*. Only the effect of Au-NRs with different amounts (0.1, 1 ml) was investigated. After 24 h treatment with 0.1 mL Au-NRs, the number of live bacteria was lower compared to treatment with 1 mL Au-NRs. The cause of this aggregation that may be belong to, at higher concentrations, gold nanorods may aggregate, thereby reducing their effective surface area for interacting with bacteria. On the other hand, the large amount of gold nanorods may not be evenly distributed throughout the medium. While the results showed that Au-NRs that were exposed to laser light had a stronger antibacterial effect than Au-NRs that were not exposed to laser light, the antibacterial effect of Au-NRs that were not exposed to laser light was comparable. Compared to the control group, exposure to green laser light significantly reduced the survival of *Escherichia coli*. With 1 mL of

photoactivated Au-NRs, the greatest reduction in bacterial cell survival was observed.

From the table (4-3) (4-4), it had been notice that there is a greater effect of spherical nanoparticles of gold than rods nanoparticles of gold in the same quantities. The nanoparticles used in this research for gold were in two spherical and rods forms as well as silver. Furthermore, rod-shaped gold particles showed a lower uptake in comparison to spherical shaped nanoparticles. The rates of uptake were lower with an increasing aspect ratio. The fraction of rod-shaped NPs exocytosed was higher than spherical-shaped nanostructures [104].

Table (4-4) Effect of Au-NRs on the survival of E.coli for different amount of Au-NRs with and without irradiation

Without laser		With laser
Au-NRs amount (ml)	Survival viability (CFU/ml)	Survival viability (CFU/ml)
Control	$2.51 \times 10^8$	$2.55 \times 10^8$
0.1	$2.45 \times 10^8$	$2.35 \times 10^8$
1	$2.5 \times 10^8$	$2.3 \times 10^8$

**4-7: Effect of Gold Nanoparticles and Silver Nanoparticles on *Escherichia coli*, *Streptococcus Pneumonia* Bacteria in solid media using the method of disk.**

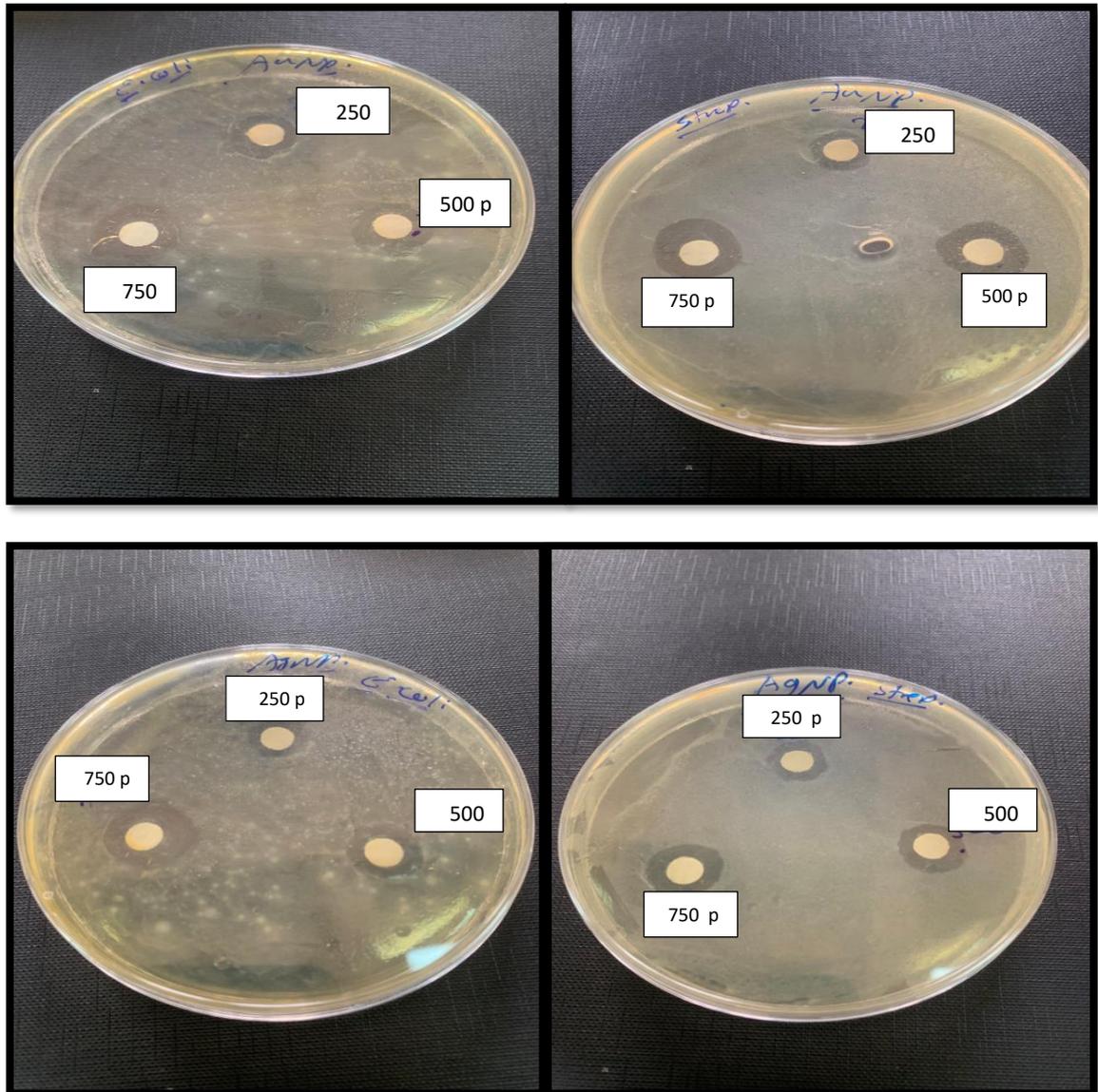


Figure (4-10) Killing and inhibition of bacteria *E.coli*, *Streptococcus pneumoniae* using silver and gold nanoparticles

Table (4-5) Average killing and inhibition diameters of *Escherichia coli*, *Streptococcus* bacteria for each concentration used.

Gold nanoparticles concentration	Diameter of killing and inhibition (mm) <i>E.coli</i>	Diameter of killing and inhibition (mm) <i>Streptococcus</i>
250 P	10	8
500 P	12	10
750 P	14	12

Silver nanoparticles concentration	Diameter of killing & inhibition (mm) <i>E.coli</i>	Diameter of killing & inhibition (mm) <i>Streptococcus</i>
250 P	11	9
500 P	12	10
750 P	15	12

It is observed from Table (4-5) that the greater the number of pulses, the higher the concentration of nanoparticles, leading to increased diameters of killing and inhibition for both types of bacteria. However, the killing rate of *Escherichia coli* bacteria is greater than the killing rate of *Streptococcus* bacteria. Gram-negative bacteria are more susceptible to silver nanoparticles. The cellular wall of gram-negative bacteria is narrower than that of gram-positive strains. The thick cellular wall may reduce the penetration of nanoparticles into cells. The different antibacterial effects of silver nanoparticles on gram-negative and gram-positive bacteria suggest that uptake of silver nanoparticles is important to the antibacterial effect [105].

It is noted from Table (4-2) that the nanomaterial with a pulse count of 750 is considered the best, as the zones of killing and inhibition increased for both types of bacteria. conclude that negative bacteria are affected more than positive bacteria. Experiment with *E. coli* show that AgNPs can cause cell lysis and cell shrinking, with a significant inhibition of the cell growth. A recent hypothesis on the action of silver NPs is that their degree of toxicity is proportional to the release of Ag<sup>+</sup>. AgNPs can also produce free radicals [106].

Silver nanoparticles (AgNPs) have been extensively studied, and they have been reported to have an effect against both Gram-negative and Gram-positive bacteria, although Gram-positive bacteria are less sensitive to the action of AgNPs than Gram-negative ones. This is attributed to the difference in the Gram-negative and Gram-positive bacteria surfaces: Gram-negative bacteria have a thin cell membrane (8–12 nm) with negatively charged lipopolysaccharides, promoting nanoparticles adhesion, while Gram-positive bacteria, on the other hand, have a thicker membrane (20–80 nm) and negatively charged peptidoglycans that can be an obstacle for Ag-NPs penetration [107].

#### **4-8: Effect Laser Irradiation in the Presence of Gold Nanoparticles for Both Types of Bacteria (*E. coli*, *Streptococcus*)**

When wells containing bacteria added to (100  $\mu$ L) of gold nanoparticles were irradiated (500, 750 pulses) with a 532 nm laser at a certain distance and at different times, it was observed that the spectral absorbance, after an incubation period of 24 h, began to decrease for all time periods. (1, 3, 5) minutes, meaning that there is a killing of bacteria that is directly proportional to all time periods. From the figure (4-11)(4-12), we note that the killing rate when the concentration of 750 pulse was greater than 500 pulse, as the killing

rate for *Escherichia coli* bacteria was higher than that of *Streptococcus* bacteria.

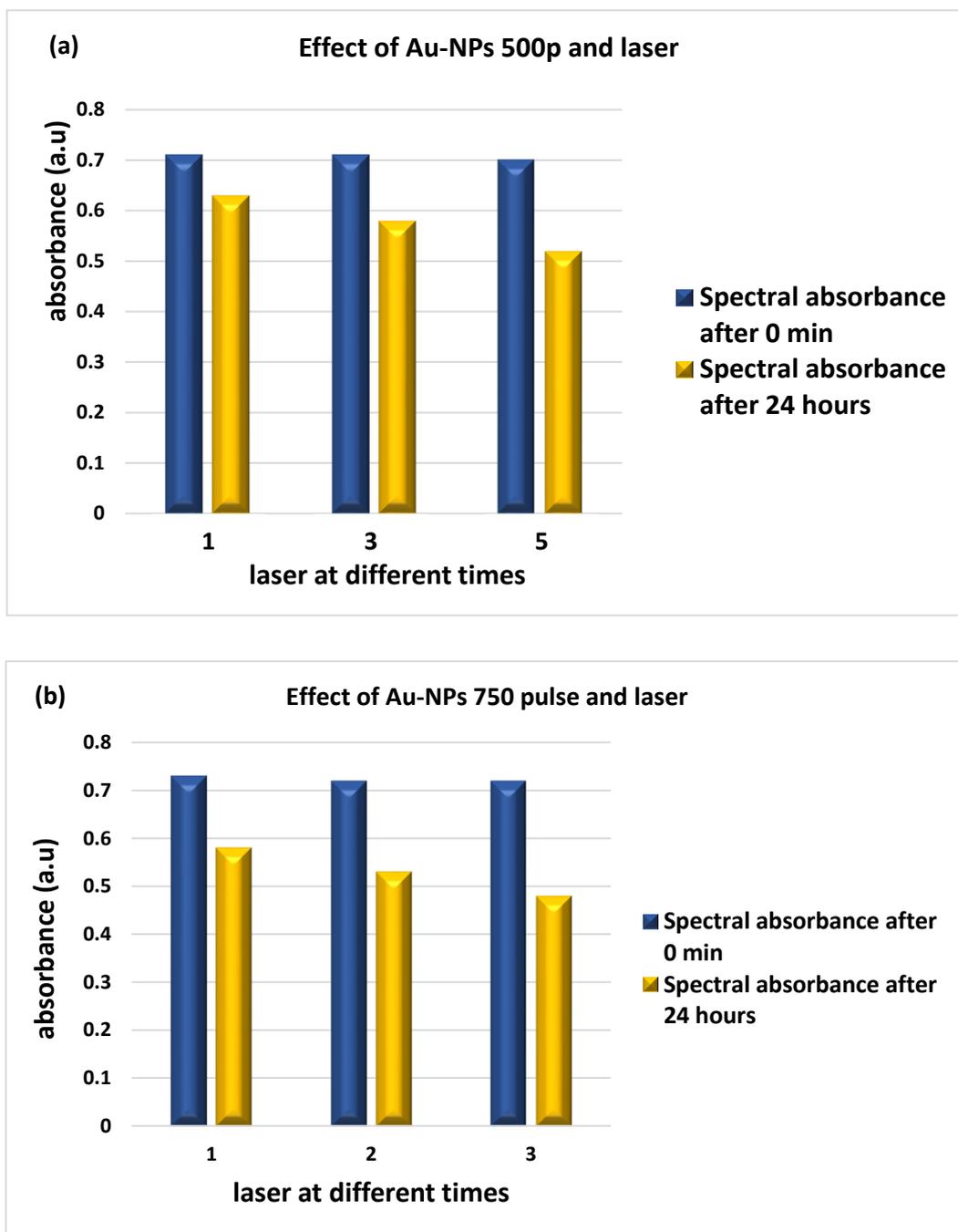


Figure (4-11): inhibition of *Streptococcus* as a function to laser exposure time (1, 3 and 5) with different concentration (a) gold nanoparticles 500 pulse (b) gold nanoparticles 750 pulse.

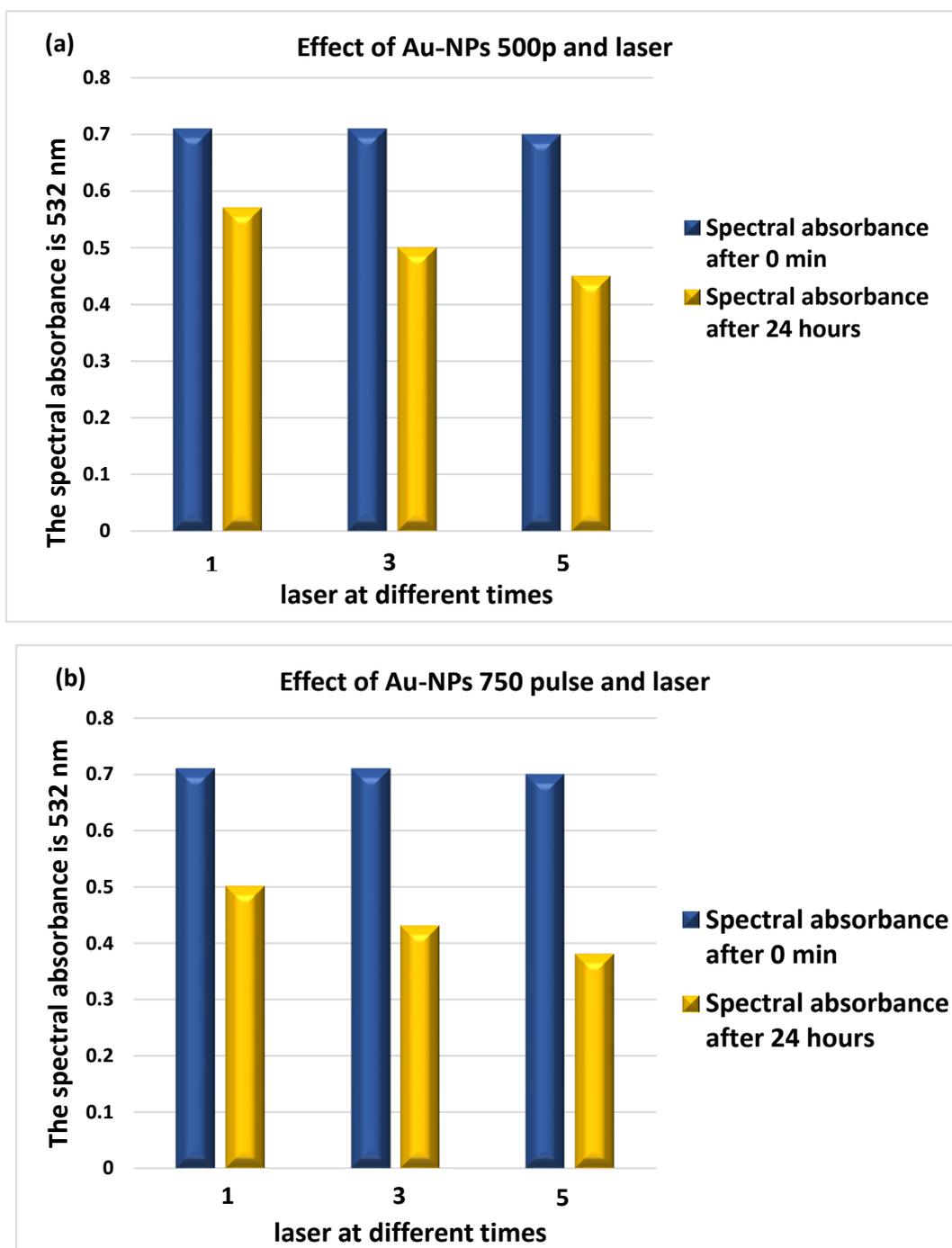
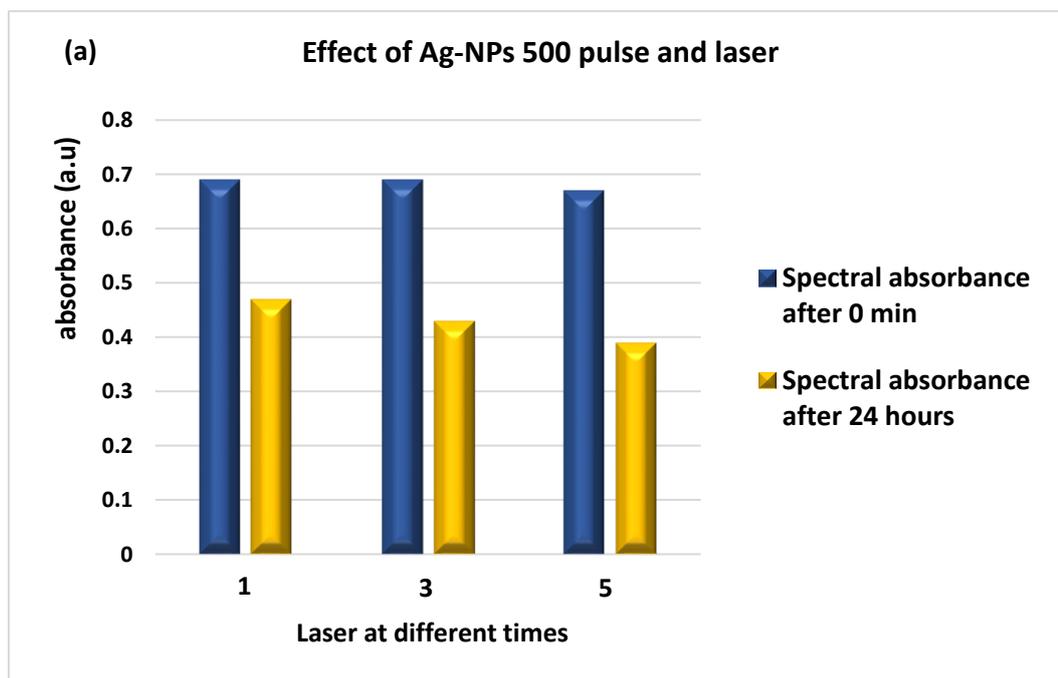


Figure (4-12): inhibition of *E.coli* as a function to laser exposure time (1, 3 and 5) with different concentration (a) gold nanoparticles 500 pulse (b) gold nanoparticles 750 pulse.

#### 4-9: laser irradiation in the presence of silver nanoparticles for both types of bacteria (*E. coli*, *Streptococcus*)

When samples of both types of bacteria containing (100  $\mu$ l) of silver nanoparticles, i.e. at a concentration of (500, 750 pulses), were irradiated with a semiconductor laser at a certain distance, for a period of (1, 3, 5) minutes, it was observed that, with an incubation period of 24 hours, the spectral absorbance decreased. That is, there is killing and inhibition for all periods during which the bacteria are exposed to the laser. The killing rate is directly proportional to the increase in time used. From the figures (4-13) (4-14), we note that the killing rate when the concentration of 750 pulse was greater than 500 pulse, as the killing rate for *Escherichia coli* bacteria was higher than that of *Streptococcus* bacteria.



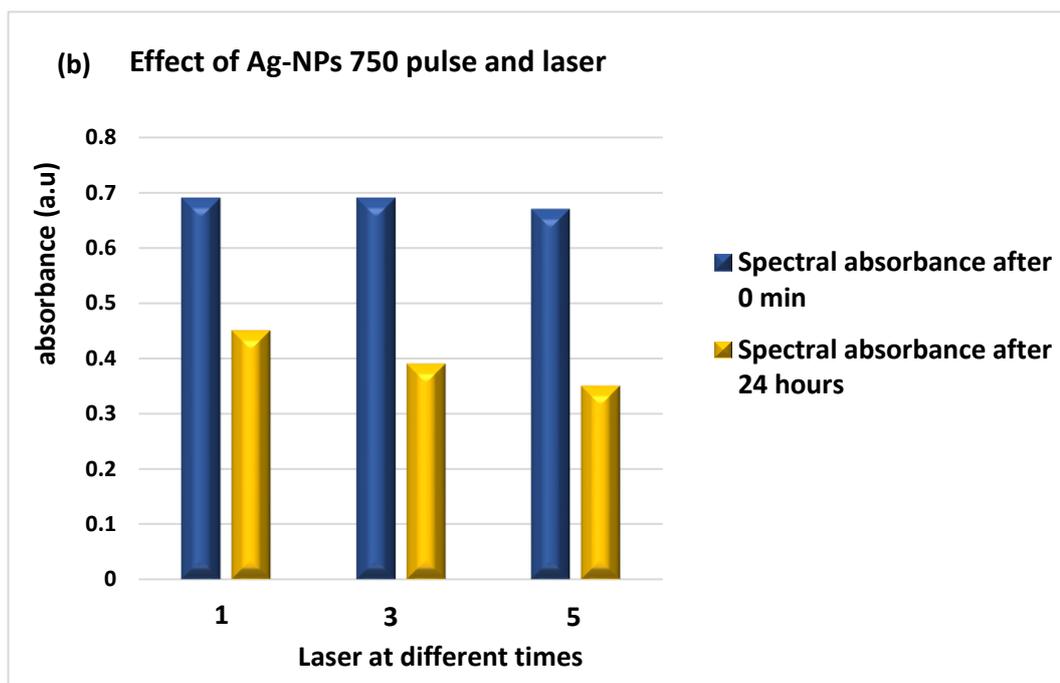
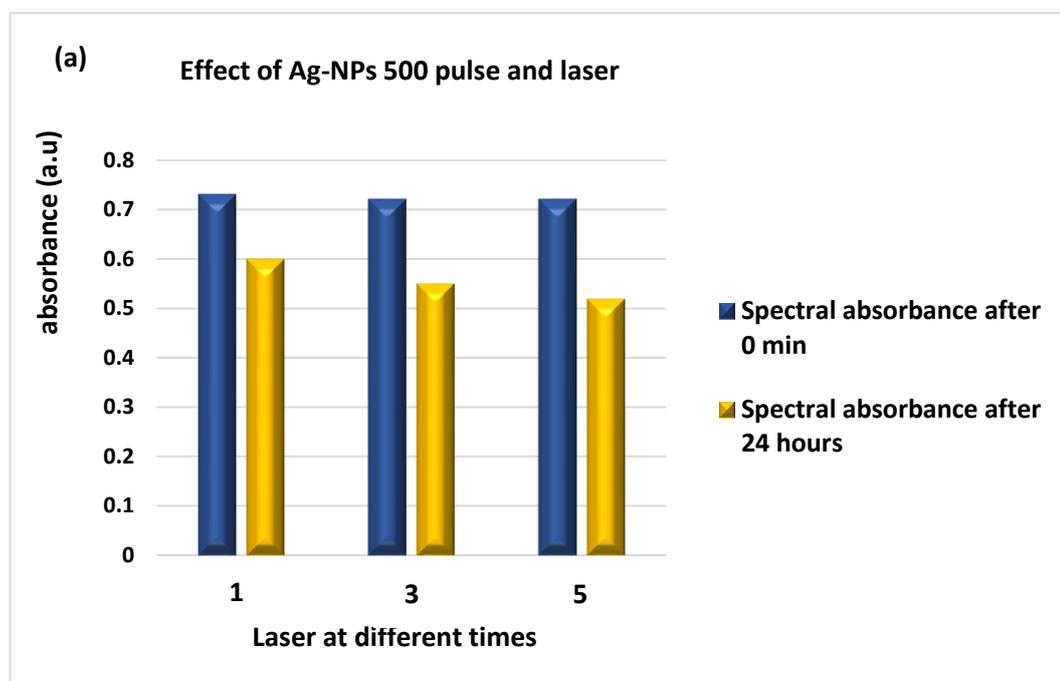


Figure (4-13): inhibition of *E.coli* as a function to laser exposure time (1, 3 and 5) with different concentration (a) silver nanoparticles 500 pulse (b) silver nanoparticles 750 pulse.



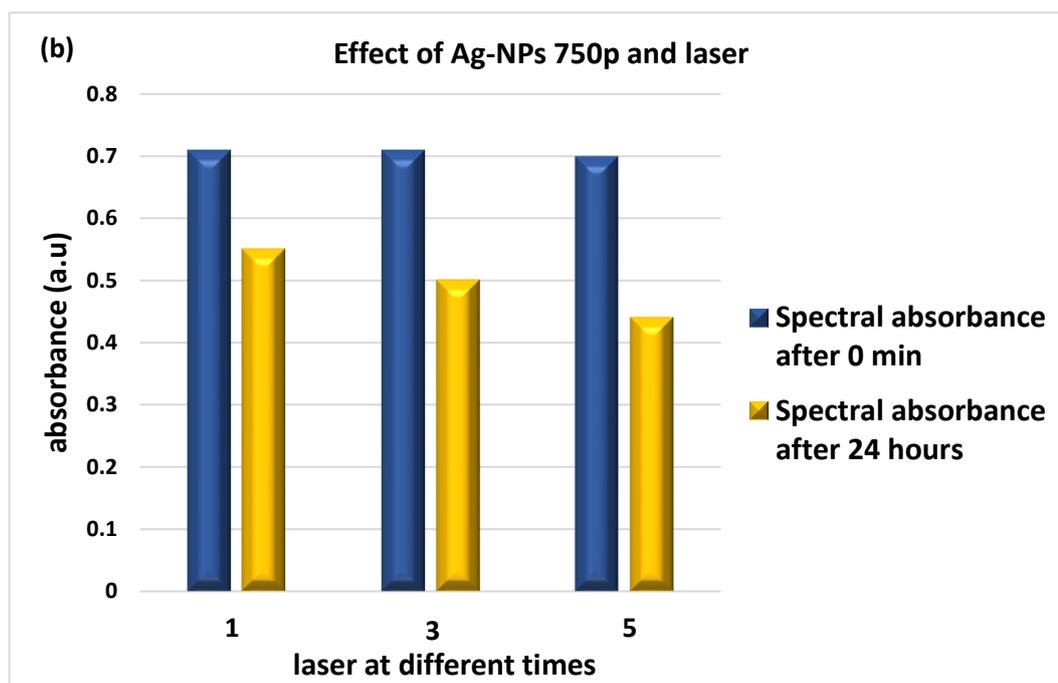


Figure (4-14): inhibition of *Streptococcus* as a function to laser exposure time (1, 3 and 5) with different concentration (a) silver nanoparticles 500 pulse (b) silver nanoparticles 750 pulse.

#### 4-10: Effect of gold nanoparticles on *Escherichia coli* and *Streptococcus pneumonia* bacteria.

Figures (4-15) (4-16) show the process of inhibition and killing of bacteria as a function of the concentration of gold nanoparticles, as the spectral absorbance decreases by a certain amount, and it is noted that there is a clear difference. Nano-gold with a number of pulses of 750 gives a killing and inhibition rate higher than 500 pulses for both types of bacteria. *Escherichia coli* and *Streptococcus*. Of the two forms, the best killing and inhibition is when the concentration is 750 pulses and with an incubation period of 24 hours. The killing and inhibition rate for *Escherichia coli* bacteria is greater than the killing and inhibition rate for *Streptococcus* bacteria. Figure (4-22) Killing and inhibiting *Streptococcus* bacteria with gold nanoparticles concentrations (500, 750 pulse) and an incubation period of 0 minutes and 24 hours.

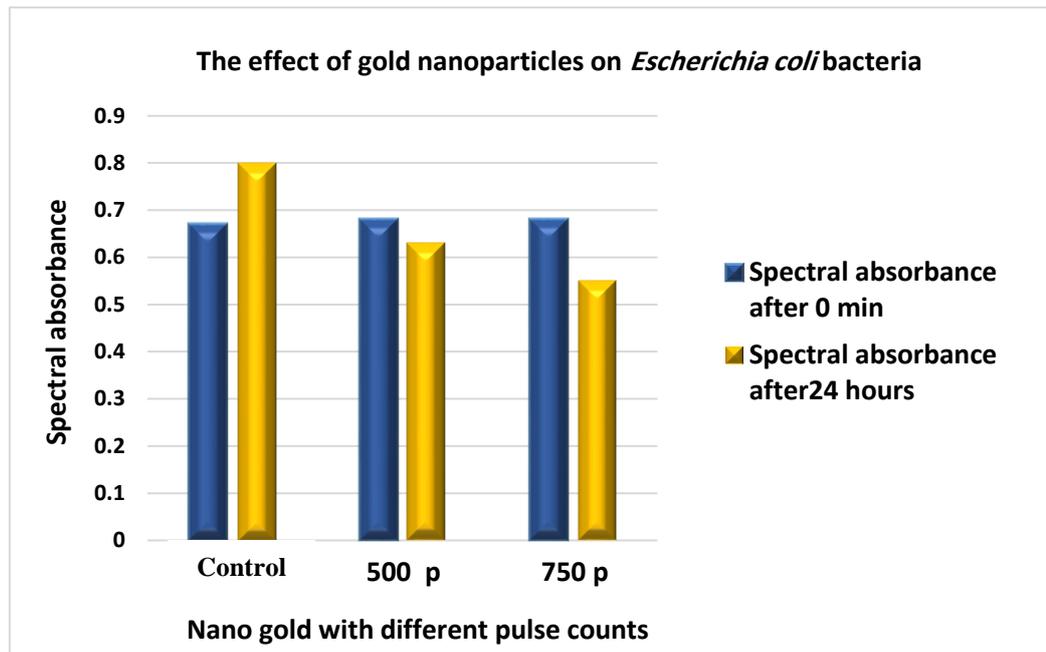


Figure (4-15) Killing and inhibiting *Escherichia coli* bacteria with gold nanoparticles concentrations (500, 750 pulse)

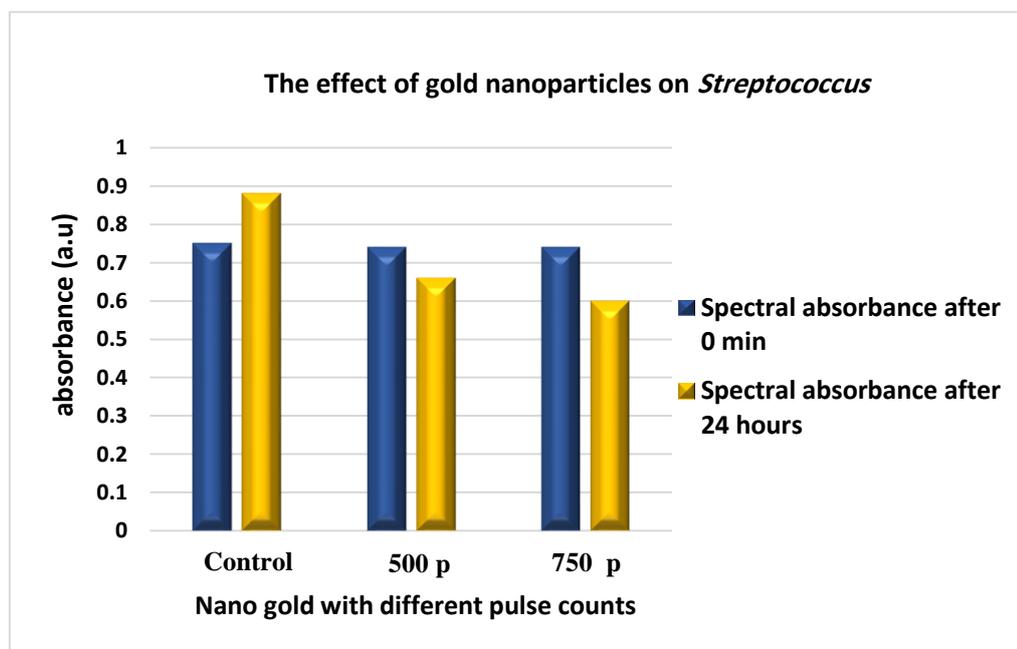


Figure (4-16) Killing and inhibiting *Streptococcus* bacteria with nano-gold concentrations (500, 750 pulse).

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**4-11: Effect of silver nanoparticles on both types of bacteria *Escherichia coli*, *Streptococcus Pneumonia*.**

Figures (4-17) and (4-18) show the results of adding nanoscale silver concentrations (500, 750 pulses) to both types of *Escherichia coli* and *Streptococcus* bacteria. It is noted that there is a difference in the spectral absorbance when incubated for 24 hours, as it decreases with silver nanoparticles with a pulse number of 750 pulses. From the two figures, we conclude that the killing and inhibition in *Escherichia coli* bacteria is higher than in *Streptococcus*. When the bacterial cells are in contact with silver, they incorporate silver ions that can inhibit many cell functions. Silver is a weak acid with a natural behavior to react with a base.

The majority of protein cells are composed of sulfur and phosphor, which are weak bases. The action of these NPs over the cells might cause a destruction of the cells. The interaction between DNA containing sulfur and phosphor as majority components and silver NPs interferes with DNA's replication, so in this way total elimination of the bacteria was realized [108]. Reactive oxygen species (ROS) generation and oxidative stress appear to be two possible mechanisms of silver nanoparticles toxicity [109].

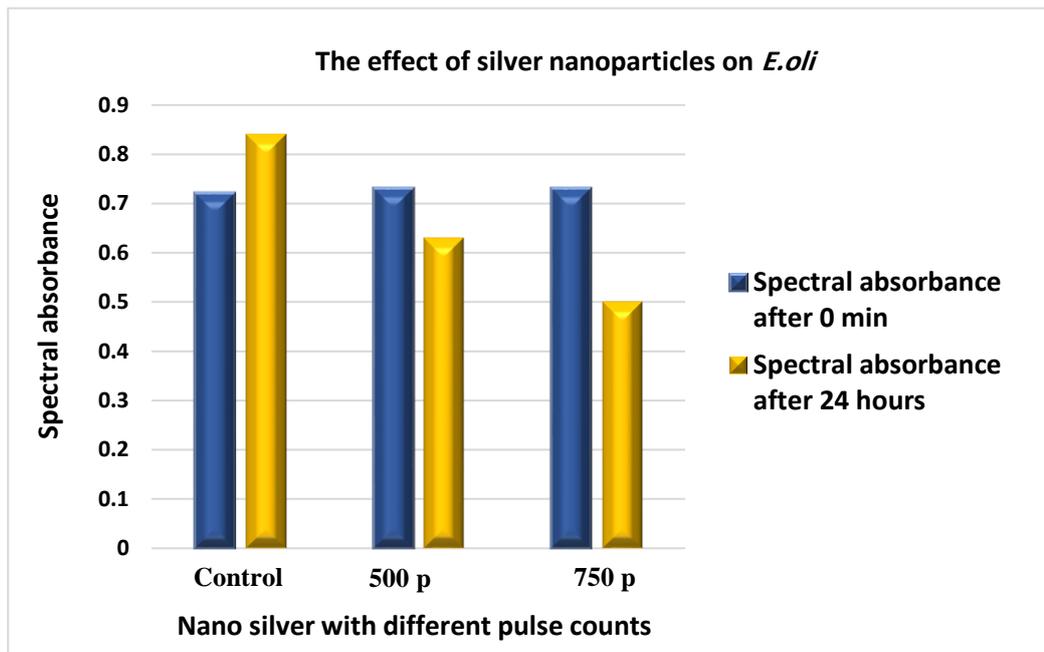


Figure (4-17) Killing and inhibiting *Escherichia coli* bacteria with silver nanoparticles concentrations (500, 750 pulse)

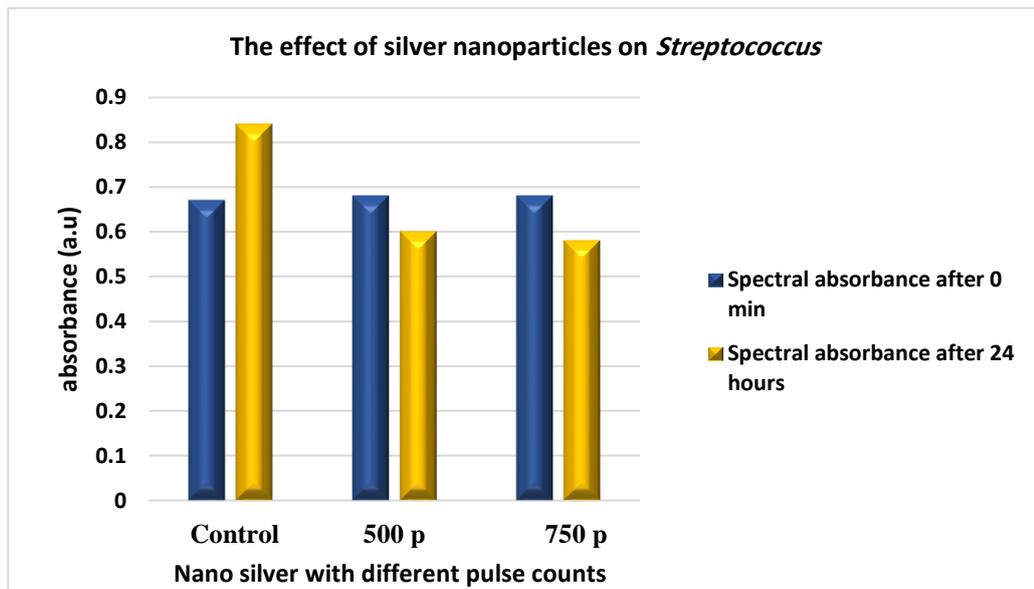


Figure (4-18) Killing and inhibiting *Streptococcus* bacteria with silver nanoparticles concentrations (500, 750 pulse)

# **Chapter Five**

## **Conclusions and Recommendation**

**(5-1): Conclusions**

The efficacy of spherical nanoparticles, specifically silver and gold, proves to be more pronounced in affecting *Escherichia coli* bacteria compared to rod-shaped nanoparticles.

Despite the alignment of the absorption peak with the laser employed, it is evident that the 405 nm (20 mW) laser outperforms the 532 nm (8 mW) laser in eradicating bacteria in the presence of nanomaterials.

The combined impact of the laser and nanomaterial is superior to the sole influence of nanomaterials on bacteria. Optimal results were achieved with nanoparticles exhibiting a pulse number of 750 for both gold and silver. Furthermore, the antimicrobial effect of silver nanoparticles surpasses that of gold nanoparticles on both *Escherichia coli* and *Streptococcus* bacteria.

The lasers utilized (405 nm, 532 nm) exhibit a distinct influence on the eradication and inhibition of bacteria (*Escherichia coli*, *Streptococcus*) when various concentrations of nanomaterials are employed. The degree of killing and inhibition correlates directly with the duration of irradiation and the concentration of the nanomaterial.

**(5-2): Future studies**

1. Using a nano polymer materials of different sizes and shapes.
2. Using a laser with a wavelength that matches the absorption spectrum of the material used.
3. Study of the effect of laser as an anti-cancer

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## الخلاصة

يمثل شعاع الليزر وتكنولوجيا النانو الحيوية نقطة اتصال بين مجالات علم الأحياء وتكنولوجيا النانو، وهي طريقة حديثة تستخدم كوسيلة لمكافحة البكتيريا بدلاً من الأدوية التقليدية. تمت في الدراسة الحالية دراسة تأثير الجسيمات النانوية (الذهب والفضة) وكذلك تأثير الجسيمات النانوية والليزر معاً لتحديد تأثيرها على بكتيريا الإشريكية القولونية سالبة الجرام والمكورة العقدية الرئوية موجبة الجرام. الطريقة المستخدمة لتحضير الجسيمات النانوية هي الاستئصال بالليزر Nd:YAG النبضي مع الماء المقطر لتحضير جزيئات الذهب والفضة ذات الشكل الكروي. وبالمثل، تم استخدام طريقة الاستئصال بالليزر من خلال تطبيق فرق الجهد الكهربائي لتكوين جزيئات الذهب والفضة على شكل قضيب. بالإضافة إلى ذلك، تتم دراسة الخصائص التركيبية والهيكلية من خلال فحوصات مثل المجهر الإلكتروني النافذ والتحليل الطيفي للمنطقة فوق البنفسجية والمرئية.

هناك ثلاث طرق تم استخدامها لتحديد تأثير الجسيمات النانوية على البكتيريا: طريقة عد المستعمرات البكتيرية، والطريقة الثانية هي قياس مساحة القتل والتثبيط، والطريقة الثالثة هي التحليل الطيفي.

وفي جميع الطرق وجد أن جزيئات الفضة النانوية كانت الأفضل مقارنة بجزيئات الذهب النانوية. وكانت الجسيمات النانوية ذات الشكل الكروي أكثر فعالية من الجسيمات النانوية على شكل قضبان. وباستخدام خاصية هذه الجسيمات النانوية في امتصاص أشعة الليزر، تبين أن أعلى انخفاض في الخلايا البكتيرية حدث عند تشعيها بالليزر 405 نانومتر وبوجود الفضة النانوية. تأثرت كل من بكتيريا الإشريكية القولونية والمكورات العقدية بشكل واضح بجسيمات الفضة النانوية، وكانت الجسيمات ذات عدد النبض 750 هي الأكثر فعالية. لقد ثبت أن بكتيريا الإشريكية القولونية تظهر استجابة أكبر مقارنة ببكتيريا العقدية في هذا السياق.

عند استخدام الجسيمات النانوية مع الليزر، حصلنا على أفضل تأثير عند تعريض البكتيريا لمدة 5 دقائق. وهذا يدل على أن الجمع بين الجسيمات النانوية والليزر له تأثير إيجابي في القضاء على البكتيريا.



جمهورية العراق  
وزارة التعليم العالي والبحث العلمي  
جامعة بابل  
كلية العلوم للبنات  
قسم فيزياء الليزر وتطبيقاته

## تأثير الليزر مع جسيمات الذهب والفضة النانوية على الفعالية البيولوجية للبكتيريا المقاومة للمضادات الحيوية

رسالة

مقدمة الى قسم فيزياء الليزر وتطبيقاته في كلية العلوم للبنات / جامعة بابل وهي جزء من  
متطلبات نيل درجة الماجستير في علوم فيزياء الليزر

من قبل

زهراء مسافر عبد

بكالوريوس علوم فيزياء الليزر 2020

بإشراف

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