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Enhancing the Working Environment of Medical Laboratories by Marking Nano-Coating to Prevent the Growth of Harmful Bacteria

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Annotation: In this work, firstly, Copper oxide nanoparticles (CuONPs and Cu2ONPs) were manufactured by using Plant Extracts method for evaluating antibacterial activtivy of samples. X-ray diffraction measurements confirm appeared the structure of CuONPs and Cu2ONPs with its phases and miller indsies. FESEM measurements of CuONPs that prepared by Senna exhibited NPs assembled in semi-spherical like-shape, and after used Onion plants extract to prepared CuONPs the results showed the most of particles are nanosheets; but after used Ziziphus Spina-Christi plants extract to prepared Cu2ONPs the results showed a cluster of NPs consists of very small NPs.

Secondly, fabricate a stable NPs/PVA nanocoating with antibacterial properties by selected water as solvent without using of surface chemical modification materials, with high chemical stability. The antimicrobial activity test shows that good inhibition percentage for NPs/(PVA) coating against Klebsiella pneumonia (Klebsiella spp.) and Salmonella bacterial.

Therefore, this work is considered a very great introduction to the employment of plant extracts and copper nanostructures in the field of antibacterial coatings, which work to protect workers in health institutions from the risk of infection with various diseases caused by microbes.

Introduction of Nanotechnology

Nanotechnology is one of the most active research areas in modern materials science. During recent times, nanotechnology has become a form of the great and interesting advanced field in physics, chemistry, engineering, and biology. This provides great hope for giving us in the coming fate with several fast starts that will change the way of technological progress in a wide variety of purposes. Nanotechnology can be termed as the fabrication, characterization, and application of nanosized (1-100) nm materials for the development of science [1]. Nanotechnology is an enormously powerful technology, which holds a huge promise for the design and development of many types of novel products with its potential medical applications on early disease detection, treatment, and prevention. Nanoparticles are shown new (improved) properties based on specific characteristics such as size, distribution, and morphology. The nanoscale dimension is important because quantum mechanical properties of electronics, photons, and atoms are appearing at this scale. Nanoscale structures permit the control of fundamental properties of materials without changing materials chemical status. It deals with the study of extremely minute structures and the prefix “nano” is a Greek word which means “dwarf or miniature” [2].

Generally, nanotechnology is playing an important role in many disciplines and becoming the most innovative scientific field. Nanotechnology is mainly.

involved in the NPs fabrication of from varying sizes, shapes, chemical compositions and the potential application of human interests [3]. The main benefits of NPs over larger sized particles are its high surface-to-volume ratio, higher surface energy, different optical, electronic, and unique magnetic characteristics [4]. As the last two decades, nanostructured materials were a widely studied because of their unique features and applications in many fields [5, 6].

The intrinsic properties of metal NPs are mainly determined by size, shape, crystallinity, composition, and morphology. NPs have a small size with distinct properties compared to the bulk form of the same material. In particular, metal NPs have formed a large study because of their optical, electronic and magnetic fields [7, 8]. It has been advised to used (Cu, Ag, and Au) metal in the antimicrobial activity versus many species of microorganisms, including the gram-negative and gram-positive bacteria and as well as fungi [9-11]. The most popular term in the nanoworld is a quantum confinement influence, which is primarily due to differences in atomic structure because of the direct influence of ultra-small length scale of the energy band structure [12]. A quantum- confined structure means a strong spatial localization of carriers (electron and hole) [13]. A quantum confined structure is one in which the motion of the carriers (electron and hole) are confined in one or more directions by potential barriers. Based on the confinement direction, a quantum confined structures will be classified into three categories: one-dimensional (1D, quantum well), two- dimensional (2D, quantum wire), three dimensional (3D, quantum dots) [14].

Nanotechnologies involved in the fabrication of nano materials may be classified as top-down or bottom-up approaches. Top-down (physical methods) or bottom-up (chemical and biological methods) is a measure of the level of advancement of nanotechnology. (Top-down) refers to building nanoscale structures by machining and etching processes, whereas (bottom-up) leads to building organic and inorganic structures atom-by-atom, or molecule-by- molecule. The NPs which can be synthesized by both top-down (physical) and bottom-up (chemical methods and via a green pathway as in this research) approaches is shown in figure (1-1).

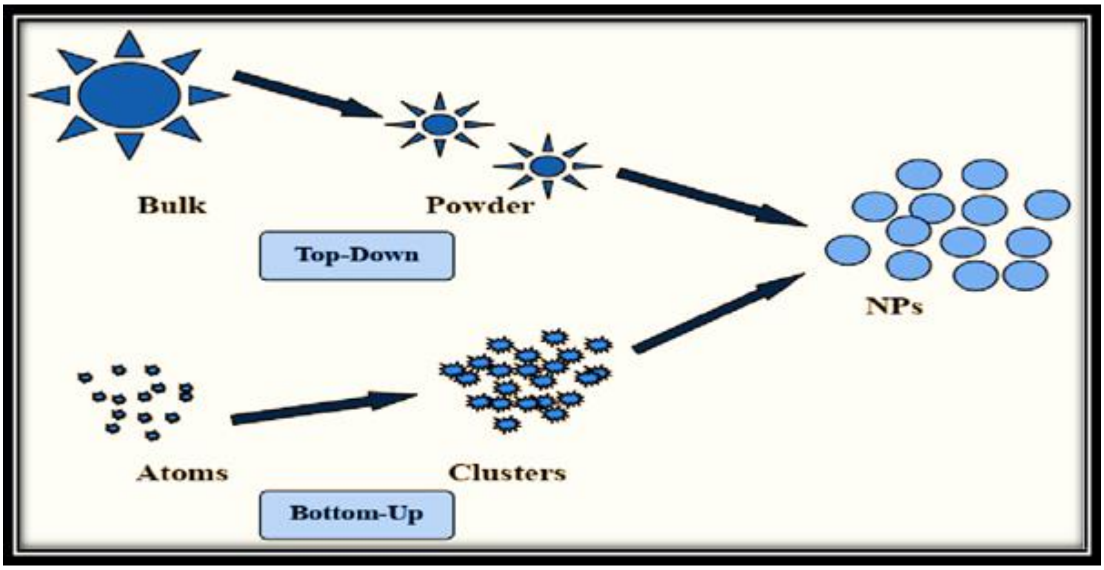


Figure (1-1): Diagrammatic of approaches of synthesis of NPs metal [15].

A number of physical and chemical approaches are explored for the synthesis and stabilization of NPs [16-18]. The physical method involves laser ablation, evaporation, and condensation methods [19]. Whereas in chemical method utilizes chemical reductants, aerosol method, electrochemical or sonochemical deposition, photochemical reduction and laser irradiation technique [20]. Additionally, physical methods are costly because of a waste of energy as well as the great pressure and temperature used in NPs fabrication and needs very advanced equipment. Although in the chemical synthesis of NPs, the generation of hazardous by-products is highlighted as well as the involvement of certain chemicals is costly and may guide to the behavior of toxic chemical varieties involved in the surface of NPs, which may become an adversarial effect on the environment. In addition to, the result of the high surface area to volume ratio of metal nanostructures, the nature of the surface is very important when considering the properties of nanosized particles. There are many methods used to synthesize nanostructures with low-cost methods; like a hydrothermal method, simple evaporation, and chemical method [21]. The increasing application of NPs in different fields, the current situation requires an eco- friendly, clean and economically viable method for the synthesis of NPs. Figure (1-2) shows the different methods of NPs synthesis.

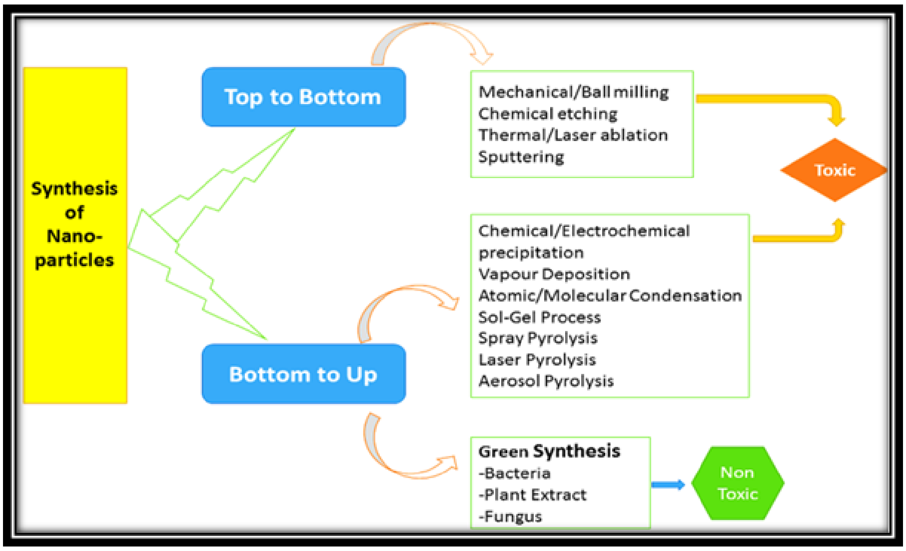


Figure (1-2): Different methods of NPs synthesis [22].

Green Synthesis of NPs:

To synthesis high-quality nanomaterials, it can be achieved through a simple, process (biosynthesis) synthesis of metal nanomaterials compared with the (artificial methods). The biological approach is close to principles of (Nature) and involves natural phenomenon that takes place in biological systems. A lot of interest has been created by the term “green nanotechnology”. Among several biological methods for NPs synthesis, microbial mediated synthesis of is, yet not commercially viable as they involve maintenance of highly hygienic conditions and very complex processes of maintaining cultures [23].

Moreover, the incubation time for the metal ions is longer in contrast the rate of production is rapidly in less time [24]. Green synthesis relies on the different process parameters and reaction conditions such as temperature, pH, solvent medium, stirring time and reducing agents. Plant-mediated synthesis is purely a green synthetic route and is considered better candidates among the different biological entities as they provide clean, eco-friendly, cost-effective, safe, conveniently utilizable and beneficial way for the synthesis of metal NPs for the large-scale production.

Many plants are reported to facilitate the formation of NPs and their potential applications [25, 26]. The value of growth of NPs varies with a reducing potential of ions and the reducing capacity of the plant depends on the presence of several polyphenols including other heterocyclic syntheses [27, 28]. The green synthesis of NPs using plants and their application has become the favorite pursuit of all scientists, including biologist, chemists, and engineers. The major advantage of using plant extracts for biogenesis of NPs is that they are easily available, safe, and nontoxic, and have plenty of metabolites that can contribute to the reduction of metal ions, and are quicker than microbes in the synthesis. The figure (1-3) shows the advantages of green synthesis by the plant extracts.

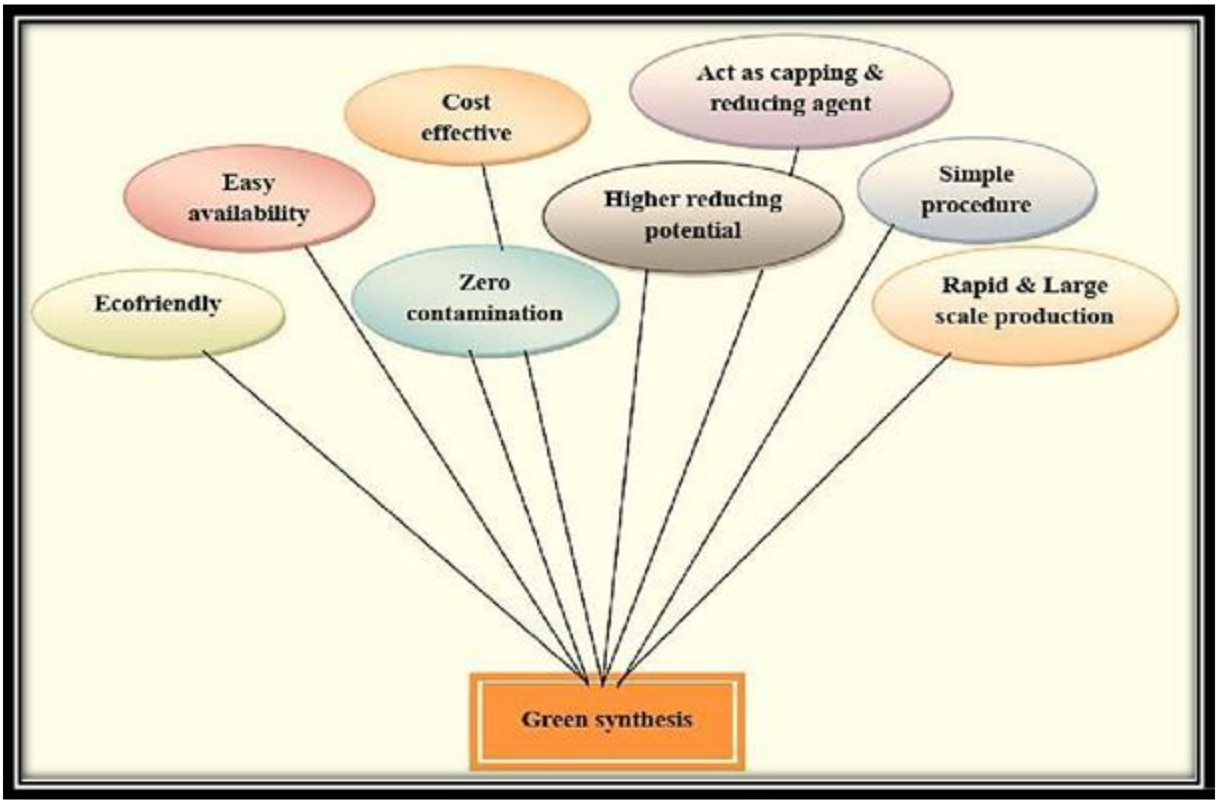


Figure (1-3): Advantages of green synthesis of NPs by plant extracts [29].

The Green Chemistry Method:

Biosynthesis of NPs is a kind of bottom-up approach, where the main reaction happening is reduction/oxidation. The need for biosynthesis of NPs increased as the physical and chemical processes costly [30]. In recent years, the development of efficient green chemistry methods employing natural reducing, capping, and stabilizing agents to prepare metal NPs with desired morphology and size has become a major focus of researchers. Biological methods (microorganisms and plants) can be used to synthesize metal NPs without the use of any harshness, toxic and expensive chemical substances [31].

Increased the interest in the use of plant extracts such as (root, leaves, flower, leg, and fruit) in the medical and biological applications. The safety, short production time, low-slung cost of cultivation and, the ability to up production, volumes make plants an attractive platform, for NPs synthesis. In addition to easily, available, safe to handle, contain a broad viability of metabolites, and phytochemicals (ketones, aldehydes, amides, terpenoids, and flavones). The studies have shown that plants have the ability to diminish metal ions. The bioreduction of metal ions by combinations of biomolecules found in the extracts of certain organisms (enzymes/proteins, amino acids, polysaccharides, and vitamins) is environmentally benign, yet chemically complex [32]. The whole plant and/or their parts such as seed, root, leaf, stem, fruit, pulp, were already reported for green synthesis of different NPs as shown in figure (1-4).

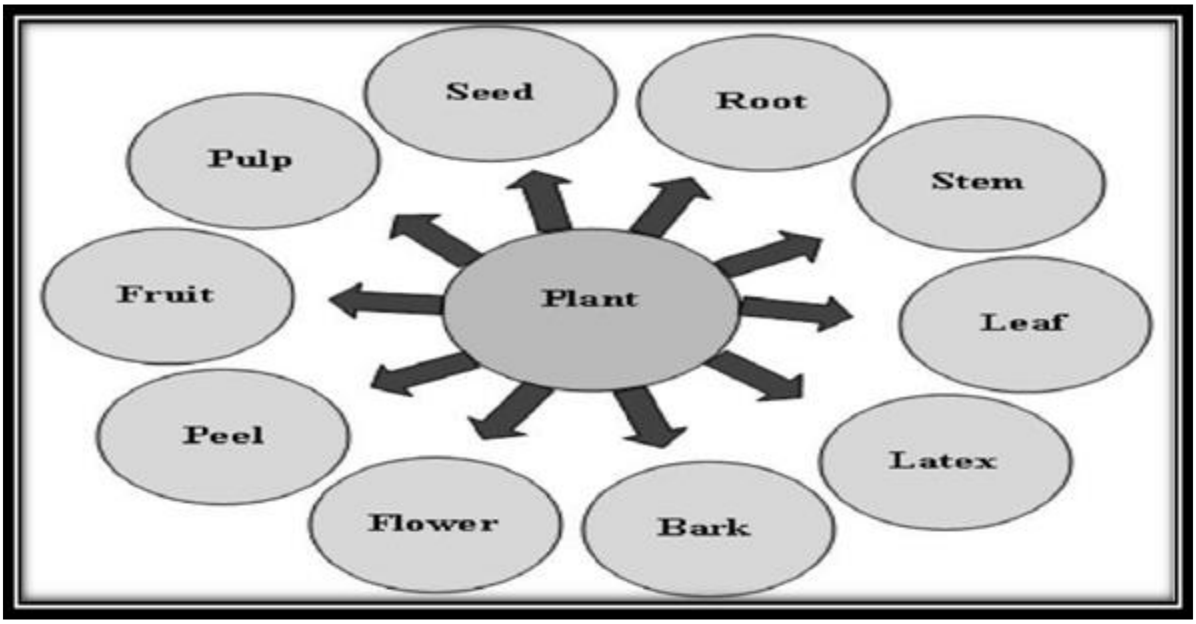


Figure (1-4): Various parts of plants used in the synthesis of NPs [33].

The Role of Plant in Green Synthesis of Metal NPs:

The major advantage of using plant extracts for NPs synthesis is that they are easily, safe, and nontoxic in most cases, have a broad variety of metabolites that can aid in the reduction of metal ions, and are quicker than microbes in the synthesis. Main mechanism considered for process is a plant-assisted reduction due to phytochemicals as shown in figure (1-5). Main phytochemicals involved are terpenoids, flavones, ketones, aldehydes, amides, and carboxylic acids. Flavones, organic acids, and quinones are water-soluble phytochemicals that are responsible for immediate reduction of ions. It suggested that phytochemicals are involved directly in reduction of ions and formation of NPs. Number of biomolecules present in plant extract reduces monovalent metal ion to uncharged atoms and these atoms aggregate to reach nano-size [34].

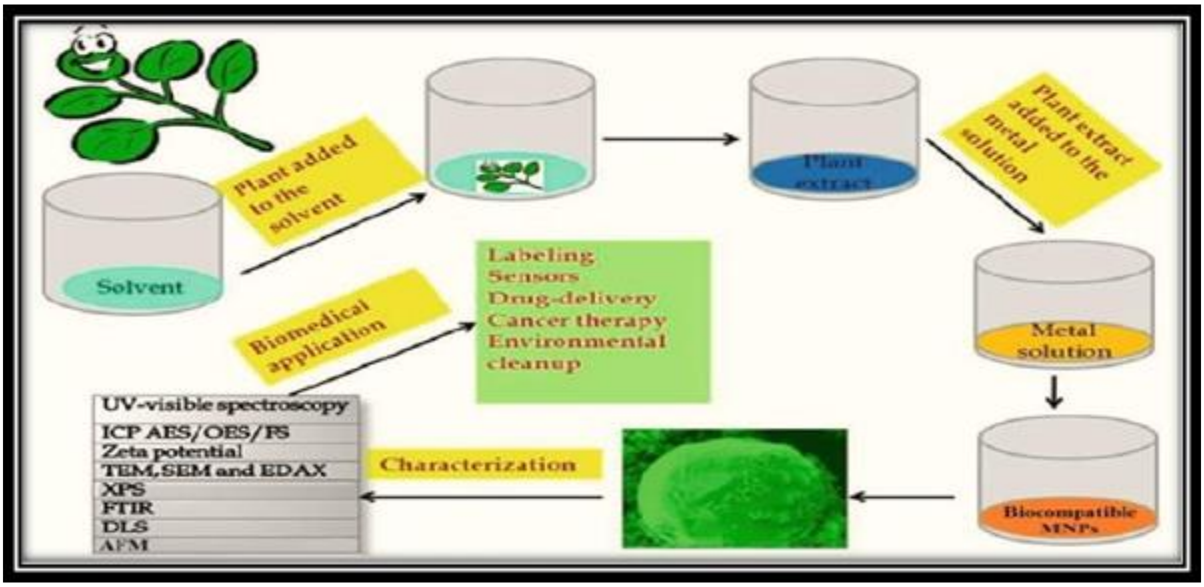


Figure (1-5): A schematic of plants as a source of green nanosynthesis, characterization and biomedical applications [35].

Factors Influencing in Green Synthesis of NPs:

The major physical and chemical parameters that affect on synthesis of NPs are:

1. Reaction temperature.
2. Metal ion concentration
3. Extract contents.
4. PH of the reaction mixture.
5. Duration of the reaction and agitation.

Parameters like metal ion concentration, extract composition and reaction period largely affect the size, shape, and morphology of the NPs [36]. Small and uniform sized NPs were synthesized by increasing pH of the reaction mixture [37]. The nearly spherical NPs were converted to spherical NP by altering pH. The particle size of NPs could be tuned by varying both time and temperature [38].

The Possible Mechanism Involved in Green Synthesis of Metal NPs From Plants Extracts:

The NPs are synthesized with two ways inside the living plants and within the sun-dried biomass, several researchers proposed various mechanistic approaches to understand the hidden pathway behind the green synthesis of NPs. The preliminary mechanism involves the accumulation of NPs after the reduction of metal ion and the reduction process is mediated by some reducing agents or involves some enzymes that are bound to the cell wall [39]. During green synthesis of NPs when plant extract is added to the solution of nitrate metal, the ionization takes place as follows:

Metal NO_3 (aq) + Plant Extract (aq)

Metal $^{+}$

+ NO_3^{-} …… (2-1)

Metal ions act as electron acceptor species, the plant biomolecules acts as an electron donor species. Chlorophyll pigments act as a stabilizing agent between donor and acceptor molecule. These biomolecules act as a chief reducing agent for reduction of metal ion to metal [40]. Apparently, biosynthetic products or reduced cofactors play an important role in the reduction of respective salts to form NPs. During glycolysis NAD (Nicotinamide adenine dinucleotide) formation occurs, which is a co-enzyme found in all living cells. NAD is an oxidizing agent which accepts electrons from other molecules and got reduced, this might have led to the conversion of metal ions to metal NPs. Plants contain a complex network of antioxidant metabolites and enzymes that work together to prevent oxidative damage to cellular components [41]. The green synthesis of metal NPs, reduction, capping, and stabilization steps occur and the biomolecules such as enzymes, proteins, and sugars due to the presence in the plant extract. The mechanism involved in each plant may vary with the types of phytoconstituents, however, the reduction of the ions is the major mechanism take place [42]. Though the exact mechanism involved in the NPs production by plants is not fully deciphered. It is believed that the biomolecules are involved directly in the reduction of the ions and formation of NPs [43]. The biochemical and molecular mechanisms of NPs green synthesis remain undiscovered, the core mechanism for the bioreduction of metal ions was hypothesized to first involve trapping of metal ions on the surface of proteins in the extract by means of electrostatic interactions. The ions are then reduced by proteins, lead to changes in their secondary structure and the formation of metal nuclei, consequently, metal nuclei grow by the reduction of metal ions and their aggregation to NPs [44].

Aims of work:

The study aims to

1. Collecting plants that possess high oxidation and reduction properties.
2. Employment of plant extracts in the preparation of CuONps and Cu2ONps from its aqueous solutions
3. Employing hydrothermal method in improving the shape of nanostructures
4. Employment of nanostructures with the PVA polymer in the manufacture of nano-coatings
5. Study the structural and morphological properties of samples.
6. Study the effect of nano-coatings on inhibiting the growth of Salmonella and Klebsiella bacterial.
7. In the end, the essence of the study is to preserve the health of workers in health institutions by creating a safe and healthy environment

Experimental Work

Introduction:

This chapter will be dealing with experimental part and includes the preparation of CuONPs and Cu2ONPs using simple green synthesis (plant extracts). Subsequently, NPs will mix with PVA polymer to obtain antibacterial **nanocoating**. Then study morphological, structure ad optical properties of NPS. The materials types with source and purity are shown in Table 3-1. The experimental part was carried out in the (i) Al-Esraa University College Medical Instrumentation Techniques Engineering Department, (ii) Physics department/College of science/ Mustansiriya-University/ Baghdad-Iraq.

Substrate preparation*:*

Glass slides have been used as substrates. They were cleaned with ethanol and ultrasonic bath for 10 min respectively, then by using distilledwater to remove both waste and impurities, then dry it carefully.

Table (3-1): Materials types, company and purity which used in this research.

Materials	**Company**	**Purity (%)**
Copper nitrate $Cu(NO_3)_2$	Scharlau	99
Sodium hydroxide (NaOH)	FEN XI CHUN	99
Polyvinyl Alcohol (PVA)	Zhuzhou Chemical	High

Sample Collection and Preparation of Plants:

Our hypothesis is that several factors together to determine the NPs synthesis including plant source, extract, temperature, value of pH, and time reaction. Three of the different plants leaves of Senna, Onion plant, and Ziziphus Spina-Christi using in current study. This plants are rich sources of phenolic substances, especially quercetin, cardiac glycosides, phenolic acids, sulfur, compounds (allicin), alkaloids, flavonoids, terpenes, steroids, vitamins, and minerals.

It is very rich in quercetin, fructose that can reduce ions to be NPs because of the presence of (vitamin C) in these plant extracts. The plants have been collected from the local market in Baghdad – Iraq, as a preliminary work.

Fresh parts (Senna, onion plant, and Ziziphus Spina-Christi) have been used in this study. 20 g of plants have rinsed with distilled water to remove impurities and dust then, cut into fine pieces and grind by the blender. Each plant mixed with 100 mL of distilled water and boiled for 30 minutes at 100 oC. Then filtered with Whatman No.4 filter paper (pore size 20 μm). Then used the centrifuge at 4000 rpm for 25 minutes to get rid of impurities. The filtrate plants extracts were used for bio-synthesis of NPs directly as soon as possible.

Fabrication of CuONPs and Cu2ONPs powder:

In a typical fabrication, 1.5 grams of copper nitrate was dissolved in 70 ml of distilled water following supporting magnetic stirring for 15 min to obtain a homogeneous solution. Afterward, 10 mL of the plant extract (Senna, onion plant, and Ziziphus Spina-Christi) was added to the above solution under magnetic stirring for 30 min. Then 4 M of NaOH was added dropwise in the above solution with continuous stirring to change the color of the solution to green yellowish, the reaction product left overnight, then collected using centrifugation, repeatedly rinsed with the distilled water and pure ethanol, then dried by the oven in 100 ° C for 2 h.

Preparation of CuONPs and Cu2ONPs /PVA nanocomposite:

2 g of PVA powder was added to 50 ml of H_2O with magnetic stirrer at temperature 80 °C until become Thick liquid. Then, about 0.2 g of NPs was added to 4 ml of pure PVA polymer solution under magnetically stirred for 1 h. After this the solution was drop casting on clean glass substrate (2 cm x 2 cm) by using "micro-pipette", and dried at 80 °C to get CuNPs/PVA nanocomposite.

Structural and morphological studies

X-Ray Diffraction (XRD)

CuONPs and Cu2ONPs crystalline phase were analyzed by XRD instrumentation at room temperature by using "Miniflex II Rigaku, Japan" provided with Cu tube for producing Cuk_{α} radiation (λ= 1.5408Å). The incident beam in the 2Θ mode overhead the range of (10^o – 80^o), and worked by (30 kV) voltage and (40 mA) filament current. The data of Specimens in this work was done via matching the intensities and positions of element peak in XRD patterns to those patterns in the database of JCPDS (Joint Committee on Powder Diffraction Standards.

Field Emission Scanning Electron Microscopy (FESEM)

All specimens were studied through FESEM-EDX (Hitachi-S 4160- Japan) and need to be capable to stand the powerful "electric-currents" which generated by using electron beam gun. Any non-conductive specimens can be destroyed due to the build-up of charges on surfaces. Therefore, the coated of non-conductive specimens with the thin layer of conductive-material e.g. gold by using a small sputtering system leads to avoid this problem.

Antimicrobial activity test

The antimicrobial performance of NPs/PVA nanocomposite have been examined against bacterial cultures Gram-negative Salmonella bacteria and Gram-negative bacteria Klebsiella pneumoniae. We have opted these two different kinds of pathogenic microbes because of the following reasons:-

- **Salmonella:-** .They usually invade only the gastrointestinal tract and cause salmonellosis, the symptoms of which can be resolved without antibiotics. However, in sub-Saharan Africa, nontyphoidal Salmonella can be invasive and cause paratyphoid fever, which requires immediate treatment with antibiotics. Typhoidal serotypes can only be transferred from human-to-human, and can cause food-borne infection, typhoid fever, and paratyphoid fever. Typhoid fever is caused by Salmonella invading the bloodstream (the typhoidal form), or in addition spreads throughout the body, invades organs, and secretes endotoxins (the septic form). This can lead to life-threatening hypovolemic shock and septic shock, and requires intensive care including antibiotics [67].
- **Klebsiella Spp.:** Gram-negative Klebsiella bacteria causes a wide range of disease states, urinary tract infections, very pneumonia, meningitis, sepsis, soft tissue infections, and diarrhea; also most infections include infection of an invasive medical device [68].

Results and Discussions

Introduction:

The current chapter will review the outcomes and discussions of structural, morphological and antibacterial activity of CuONPs and Cu2ONPs prepared by using a simple plant extracts method.

Structural Characterization:

The microstructure is defined by the type, crystal structure, number, shape and topological arrangement of phases and defects such as point defects, dislocations, stacking faults or grain boundaries in a crystalline material.

X-ray Diffraction (XRD):

Figure 4-1A (1-3) shows XRD patterns of CuONPs and Cu2ONPs that prepare by plant extracts method, with diffraction peaks, confirm appear all phase of copper oxide. For the samples that prepared by used Senna and Onion plant extracts, the results of XRD spectra showed that, the observed peaks matching well with the reported (JCPDS; 100-005-0661) data of CuONPs and all diffraction peaks in the XRD patterns corresponding to the (110), (002), (-111), (-202), (020), (202) and (- 133) miller indices. But, For the sample that prepared by used Ziziphus Spina- Christi plants extract, the results of XRD spectra showed that, the observed peaks matching well with the reported (JCPDS; 01-077-0199) data of Cu2ONPs and all diffraction peaks in the XRD patterns corresponding to the (110), (111), (200), (220) and (311) miller indices as illustrated in a table (4-1). The crystallite sizes were estimated to be about (9.9-20.9) nm for CuONPs that prepared by Senna, (11.8- 26.7) nm for CuONPs that prepared by Onionn and (22.5-47.3) nm for Cu2ONPs that prepared by Ziziphus Spina-Christi.

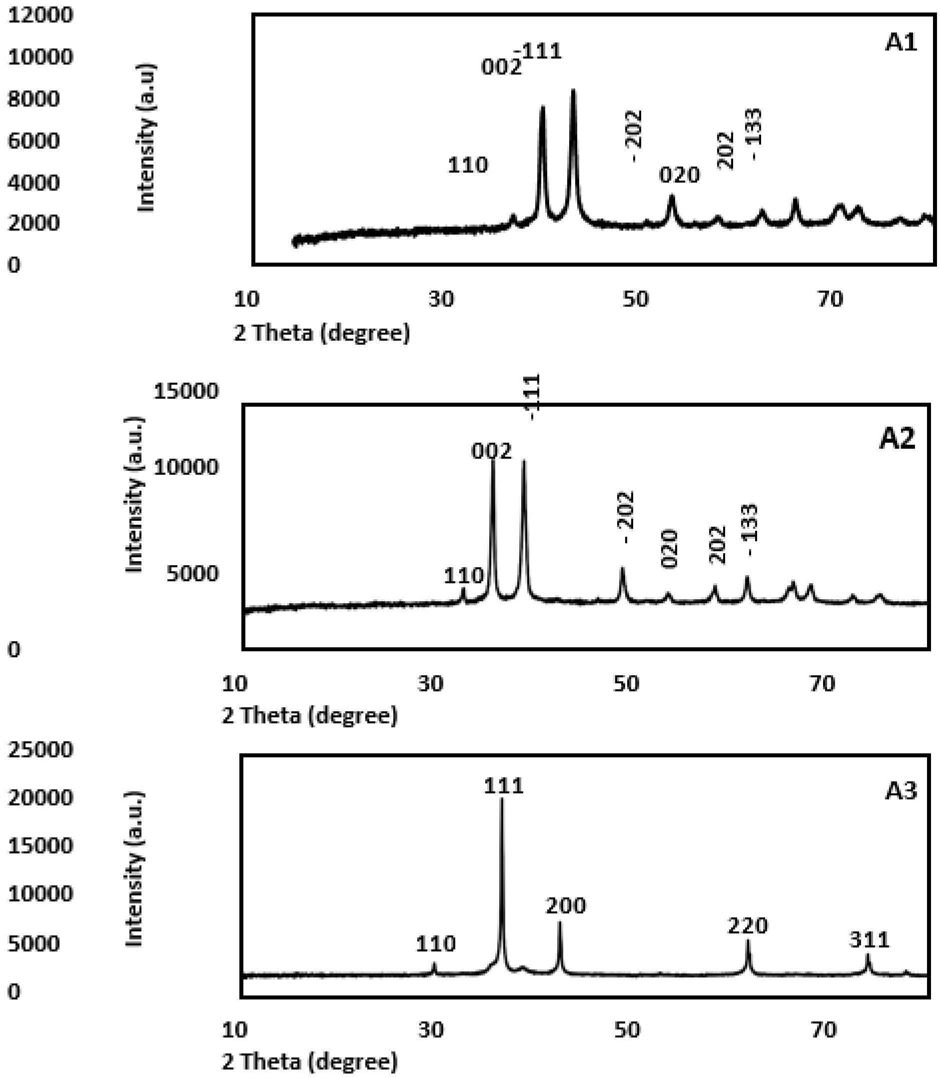


Figure (4-1): XRD pattern of CuONPs as-prepared by using (A1) Senna extract, (A2) Onion, and of Cu2ONPs as-prepared by using (A3) Ziziphus Spina-Christi

Table (4-1): XRD results of CuONPs and Cu2ONPs via plant extracts method.

Material	Plant extract	(hkl)	2θ Exp. (deg.)	2θ JCPDS (deg.)	FWHM (deg.)	Crystallite size (nm)
CuO	Senna	110	32.58	32.51	0.5	16.6
		002	35.61	35.45	0.4	20.9
		-111	38.708	38.73	0.55	15.3
		-202	48.83	48.76	0.64	13.6
		020	53.59	53.4	0.9	9.9
		202	58.15	58.3	0.9	10.1
		-113	61.51	61.57	0.5	18.5
CuO	Onion	110	32.54	32.51	0.31	26.70
		002	35.556	35.45	0.4	20.86
		-111	38.708	38.73	0.44	19.14
		-202	48.76	48.76	0.33	26.44
		020	53.46	53.4	0.6	14.83
		202	58.29	58.3	0.77	11.82
		-113	61.52	61.57	0.41	22.56

Cu2O	**Ziziphus Spina- Christi**	**110**	**29.66**	**29.647**	**0.32**	**25.7**
		111	**36.507**	**36.521**	**0.195**	**42.9**
		200	**42.409**	**42.423**	**0.21**	**40.6**
		220	**61.534**	**61.552**	**0.25**	**37.0**
		311	**73.712**	**73.739**	**0.21**	**47.3**

FESEM of CuONPs and Cu2ONPs

The FE-SEM analysis was used to determine the morphological structure of the reaction products during the biosynthesis of metal NPs. Figure (4-2) shows the morphological variation with used different plant extracts of Senna, onion, and Ziziphus Spina-Christi. Image A1 showed morphological results after used Senna plants extract to prepared CuONPs with relatively a homogenous size and these NPs assembled in semi-spherical like-shape with average diameters ranging between (38-53.59) nm. Image A2 showed morphological results after used Onion plants extract to prepared CuONPs and images show very clearly, that the most of particles are nanosheets with an average diameter of (44.66-37.96) nm. Image A(3-4) showed the morphological of Cu2ONPs after used Ziziphus Spina-Christi plants extract and the results showed a cluster of NPs consists of very small NPs with an average diameter of (40.19-73.93) nm.

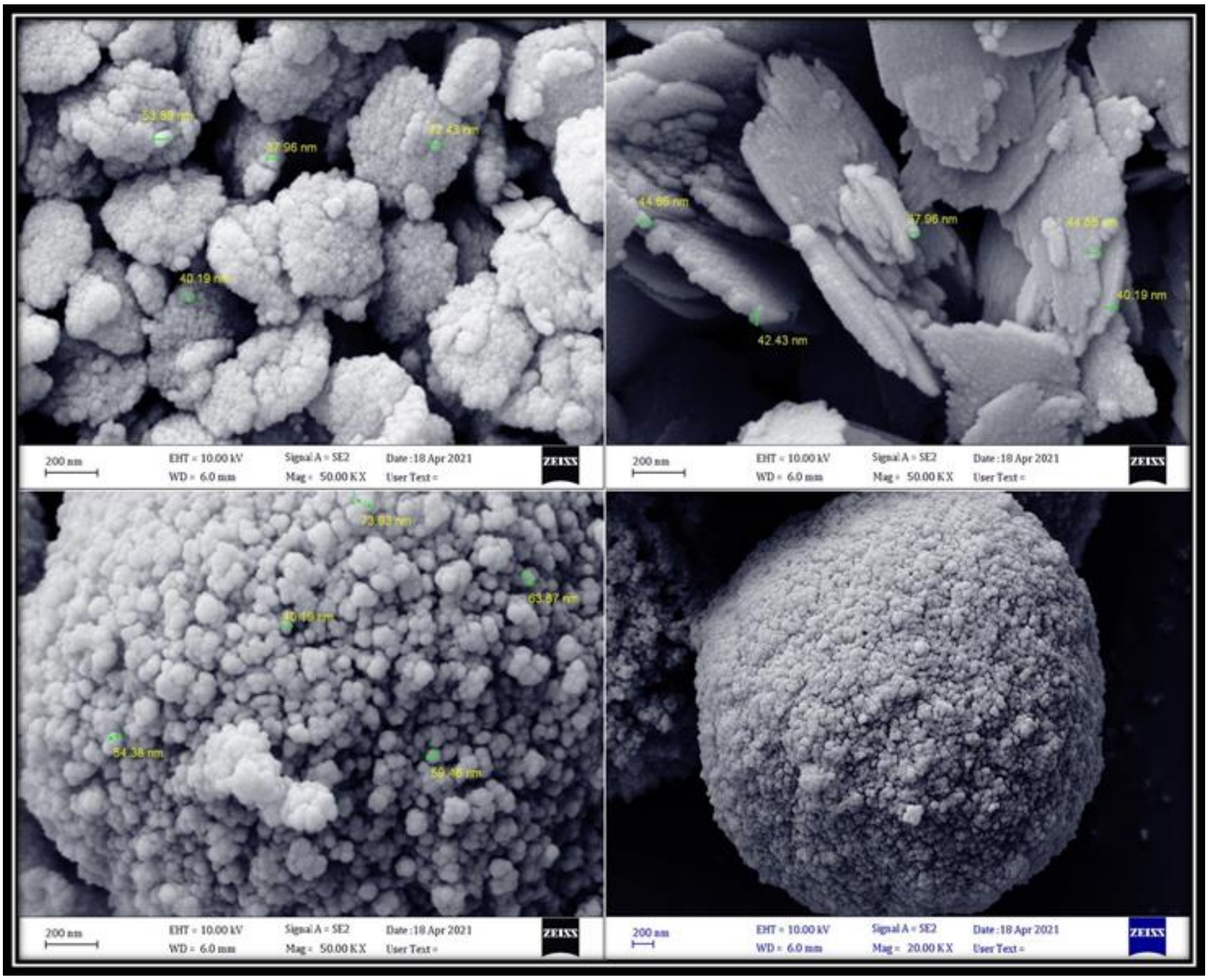
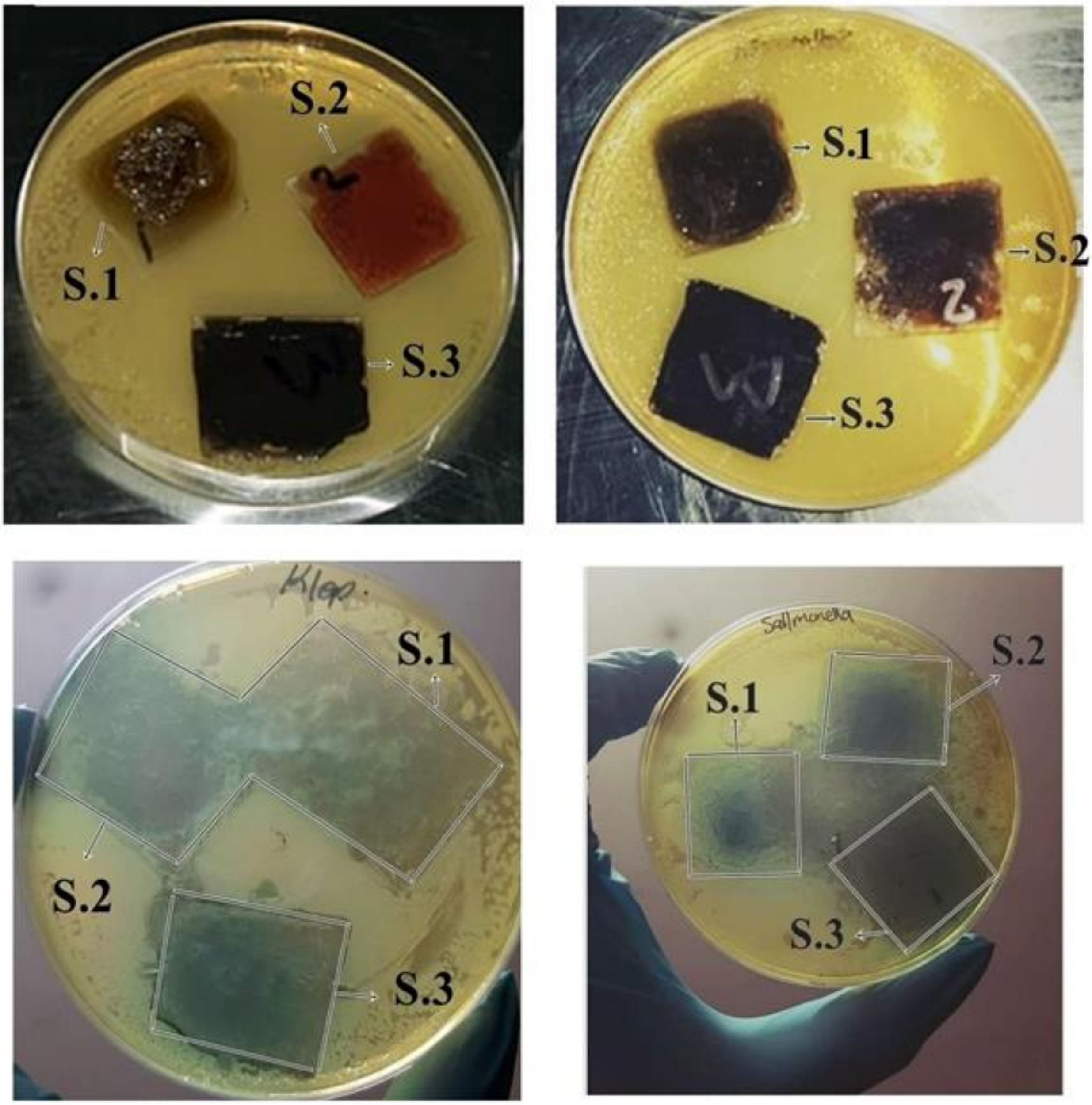


Figure (4-2): FE-SEM images of CuONPs as-prepared by using (A) Senna extract, (A2) Onion, and of Cu2ONPs as-prepared by using (A3,4) Ziziphus Spina-Christi, with scale bar 200 nm.

Antibacterial Activity of CuONPs and Cu2ONPs /PVA surfaces:

The antibacterial activity of surfaces that were prepared has been studied, by using different plant extracts. The efficiency of anti-bacterial activity has been measured against two stander bacterial isolates; Gram-negative bacteria (Salmonella) and Gram-negative bacteria (klebsiella). Figure (4-3) shows the inhibition zones of Cu2ONPs and CuONPs/PVA surfaces hat prepared by used plant extracts of Senna, onion, and Ziziphus Spina-Christi. From this figure, the result shows the high antibacterial activity of all samples against both bacteria. Also, the results showed that CuNPs/PVA nanocomposite that prepared with added plant extracts of Senna has more effective inhibition against both bacteria compared with another sample, and this may be correlated with shaps and structure of Cu2ONPs and CuONPs at this condition.



Klebsiella **Salmonella**

(A) **(B)**

Figure (4-3): The antibacterial activity of CuONPs/PVA surfaces That prepared by used plant extracts of Senna (S1), onion (S2), and of Cu2ONPs that prepared by Ziziphus Spina-Christi (S3) against (A) klebsiella and (B) Salmonella.

To explain antibacterial activity results, the NPs or nanostructures are very small size and have a great surface to volume ratio; and it has physical and chemical properties that differ from those properties in the case of the bulk state of the same chemical composition. However, NPs worked by direct contact with the bacterial cell and the action of this process depending on the aspect ratio (surface area to volume), concentration of NPs and particle shape [70]. Also, The antibactericidal activity of Cu meatillc (NPs) is chiefly due to the release of copper ions (Cu^{+}) and these ions have attached to the bacterial cell wall due to electrostatic attraction. Furthermore, the metal ions are not only interacting with the surface of a membrane but can also penetrate inside the bacteria. NPs may react with the thiol group (-SH) in the cell wall of the bacteria and not allow the transport of nutrients through the cell wall. The protein decreases inside the cell, eventually causing the cellular death. Or the other reason, because the smaller particle size of metal NPs are correlated with a larger band gap and consequently, more available excitons will lead to the generation of a higher concentration of reactive oxygen species (ROS) to enhance antimicrobial activities [71]. However, the activity depended on larger surface area to volume, concentration of NPs, crystalline structure, and particle shape [72].

Conclusions:

According to the results shown in this work, the conclusions are summarized as follows:

1. The simple green synthesis (plant extracts) method used suitable for the fabrication CuONPs and Cu2ONPs, and XRD measurements confirm the crystalline structure of CuONPs and Cu2ONPs with appearing many phases of copper structure in XRD spectra. And this confirms successfully of work.
2. The phytochemicals: proteins, flavonoids, vitamin C, and phenols are responsible for the green synthesis of nano-metal where it converts metal ions to metal NPs. In other words, these compounds worked as reducing and stabilizing agents. The employed of Senna and Onion leads to prepared CuONPs and employed of Ziziphus Spina-Christi leads to prepared Cu2ONPs.
3. FESEM images confirm appear many nanostructures such as (Semi- spherical like-shape after used Senna, nanosheets after used Onion; a cluster of NPs consists of very small NPs after used Ziziphus Spina-Christi plants extract) with an increase of aspect ratio at the nanoscale.
4. For all samples, Antimicrobial activity shows that good inhibition percentage for NPs/PVA surfaces against Klebsiella and Salmonella bacteria.

And this may be attributed to the unique shape and the size of the nanostructure and also the increasing aspect ratio (ratio surface to volume) at a nanoscale.

5. The results achieved in this work give great hope for improving the biological reality of the medical laboratory environment, as it is possible to demonstrate the results of the work in the manufacture of nano-coatings that inhibit the growth of microorganisms in medical laboratories and thus reduce the risk to medical personnel and patients.

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