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The Inhibitory Action of Copper Oxide Nanoparticles (CuO NPs) against *Escherichia coli* as an Antibiotic

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ABSTRACT

This work shows the ability of copper oxide nanoparticles (CuO NPs) to prevent the growth of bacteria (*Escherichia coli*), and know the amount of concentration in which the effectiveness of these molecules lies against this type of bacteria, and this was done by examining CuO NPs and their spread in dishes petri containing bacteria colonies, with different concentrations (100%, 75%, 50%, 25%, and 10%). The minimum total of these concentrations has been determined, at which the effectiveness of copper nanoparticle oxide molecules is shown to prevent growth, which has a value of 50 %, And then the effectiveness increases as the concentration increases until the concentration reaches 100%, and this complete focus is the ideal focus at Show it with cefotaxime antibiotic, so that it has the same amount of effect in the inhibition. Hence, it became clear to us that the inhibitory area formed was 20 mm. On this basis, so, it is possible to consider nanoparticles of copper oxide nanoparticles (CuO NPs), as appropriate antibiotics for the treatment of infections and all diseases caused by *E. coli* bacteria due to their comparison with the effectiveness of Cefotaxime (CX30) and gave the same result in the inhibitory ability to inhibit bacterial growth, so it was considered a treatment.

Keyword: copper oxide nanoparticles, *Escherichia coli*, antibacterial effect, Cefotaxime

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INTRODUCTION

Nanoparticles have entered into the study of many sciences today, because of their great importance in all medical, biological, pharmaceutical, industrial and military fields as well, and became the concern of most researchers in their various studies in all disciplines witnessed by the scientific, research and applied realities on the reality. Nanoparticles can be classified into different types according to their size, morphology, physical, chemical, and engineering properties. Some such types involve carbon based nanoparticles, ceramic nanoparticles, metal nanoparticles, semiconductor nanoparticles, polymeric nanoparticles and lipid based nanoparticles. In particular, metallic and semiconductor nanomaterial's have gained great popularity in recent years due to their wide fields of application, such as biomedical applications (1), for the release of drugs, sensor devices or cancer treatment, paints (2), electronics (3), and antibacterial functions (4), among others. In this sensation, silver nanoparticles (AgNPs), and copper oxide nanoparticles (CuONPs) are the most used and have been applied in many consumer products owing to their excellent properties, especially antibacterial activity and biochemical detection. There are different chemical, and organic compounds, with antibacterial activity such as, penicillin (β-lactams group) and natural products, which kill microbes, or slow down their growth (5). Among them, nanoparticles, (metallic and semiconductor) have recently obtained more attentiveness (6). Reactive Oxygen Species (ROS) like super oxide anion (O2-), hydrogen peroxide (H2O2), hydroxyl radicals (HO●), and organic hydro peroxides (OHP), NPs deposition on the surface of microorganisms, and (NPs) accumulation in the cytoplasm-periplasmic region of bacteria, must be lead to microorganisms death (7). In the case of bacteria ROS can resulted in denaturation or damage of cellular constituents including lipids, peptidoglycan, proteins, and DNA through generating of ROS by nanoparticles and subsequently physical disruption (8). Metallic and semiconductor nanoparticles are considerable materials for investigation in nanomedicine field. Among various metal oxide nanoparticles CuO has attracted particular attention, because it is the simplest member of the family of copper compounds, and shows a range of useful physical properties, such as high temperature super conductivity, electron correlation effects, and spin dynamics (9). Microbial contamination of air, water, and soil due to different types of microorganisms that lead to many problems in living conditions and is a serious issue in health care. Due to the spread of antibiotic resistant strains of bacteria that cause serious infections, interest in alternative antimicrobial agents, such as small antibiotics, cationic polymers, metal nanoparticles, and antimicrobial peptides, have been given much considerations (10). Researchers have shown in many studies the possibility of using nanoparticles of different minerals to inhibit the growth of bacteria and microbes in general, and from these nanoparticles, of which copper oxide can be used as an antibiotic, the effect of which is to inhibit the growth of bacteria is effective, and clear from experiments conducted in this side.

MATERIALS AND METHODS

There is a set of basic steps, that underlie the current research and that depend on providing a set of materials that are required during the work, including CuO NPs (50 nm), MacConkey's, Nutrient, and Muller hintonagar, and antibiotic disk cefotaxime (CX30). The bacterial isolates, were obtained from the patients who visit al-hindiea Hospital in Karbala, Iraq. All bacterial specimens were cultured on MacConkey's agar plates for 24-48 hours at 37ºC for isolation and purification. All isolates, were confirmed by Viteck (2) compact system (Biomérieux). Antimicrobial activity of the CuO NPs, were tested against human pathogenic bacteria, gram negative bacteria (*Escherichia coli*) that was maintained on nutrient agar slants. The antimicrobial activity was carried out as described by the clinical laboratory. Bacterial sensitivity to antibiotic or CuO NPs is tested using a disk diffusion assay.

CuO NPs were used in dilutions of (100%, 75%, 50%, 25%, and 10%), in sterile deionized water. The isolates were initially incubated, for (15min) at (4ºC) then incubated at (37ºC) over night. The test results were scored when a zone of inhibition, was observed around the well after the incubation period, then the inhibition zone diameter was measured. The bacterial isolates were incubated at (37ºC) overnight, which were used to prepare. About (10ml) tube nutrient broth medium was prepared then each sample was inoculated aseptically with (1 ml), of the respective bacterial suspension. Five dilutions of CuO nanoparticles were prepared (100%, 75%, 50%, 25%, and 10%), in sterile deionized water. The inoculated sets were incubated at (37 ºC). After incubation period, the visible turbidity in each tube, was investigated, show table: 1.

Table 1: The table shows the amount of concentrations and their effect on inhibiting bacterial growth

CuONPs concentrations% with controle	The measure of inhibition zones mm
10%	non
25%	non
50%	5mm
75%	10mm
100%	20mm
Cefotaxime (CX30)	20mm

Among various metal oxide nanoparticles, CuO has attracted particular attention, because it is the simplest member of the family of copper compounds, and shows a range of useful chemical and physical properties such as reactions, and high temperature super conductivity, electron correlation effects, and spin dynamics. Microbial contamination of air, water, and soil due to different types of microorganisms creates problems in living conditions and is a serious issue in health care. Due to the spread of antibiotic resistant strains that cause serious infections, interest in alternative antimicrobial agents, such as small antibiotics, cationic polymers, metal nanoparticles, and antimicrobial peptides have been given much considerations (10).

RESULTS AND DISCUSSION

Antibacterial activity is defined as killing bacteria, or reducing their growth without general toxic , to surrounding tissue of body. Antibacterial activity is defined as killing bacteria or reducing their growth without a general toxicity, to the tissues surrounding any part of the body (11). In our current research, copper oxide nanoparticles were used to prevent and inhibit the growth of *E. coli* bacteria. This was done by testing different concentrations of nanoparticles, for copper oxide, and comparing them with a specific antibiotic, cefotaxine (CX30). There are several reports about antibacterial properties of nanoparticles (12). The different concentrations of nanoparticle of copper oxide that were used in showed that the minimum concentration in preventing bacteria growth , and inhibiting their growth is (50 %). The ideal limit for inhibitory action is the (100 %) concentration, which is identical to that of the antibiotic cefotaxine (CX30). Fig, 1:

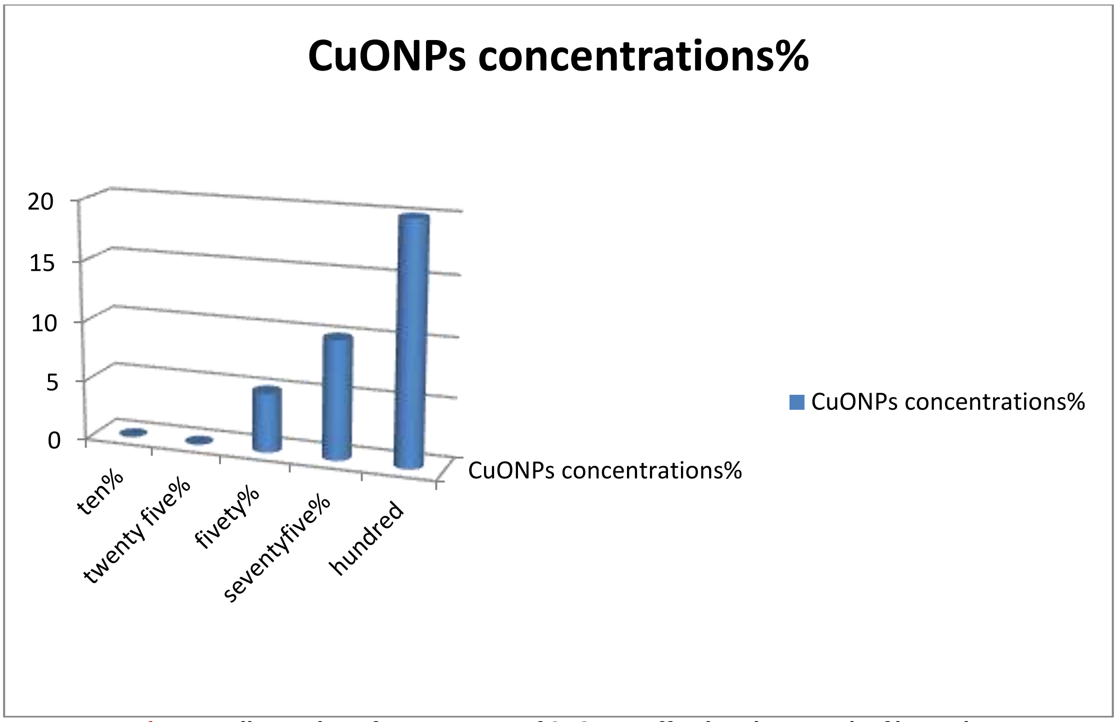

Figure 1: **Illustration of percentages of CuO NPs affecting the growth of bacteria**

Metal nanoparticles comprising oxides of magnesium, silver, copper, zinc, iron and nickel have been reported, with antimicrobial properties, this work investigates the anti-bacterial activity of copper oxide nanoparticles (CuO NPs), against various pathogenic bacterial strains such as *Escherichia coli*. As shown in the above figure, the copper oxide nanoparticle does not perform any activity if it is at a concentration of (10-25%), but its effectiveness starts at the minimum concentration of (50 %), and above. In the figure 2 below it becomes clear to us that the inhibition areas, were non-existent in the small concentration that starts from (10% to 25%), thus the inhibition areas are zero, but when the concentration gradually increases, which starts from (50 %), until it reaches the full concentration 100%, there is a noticeable increase With an area of inhibition areas from (5 mm), until you reach the ideal amount of inhibition (20 mm), Figure 2.

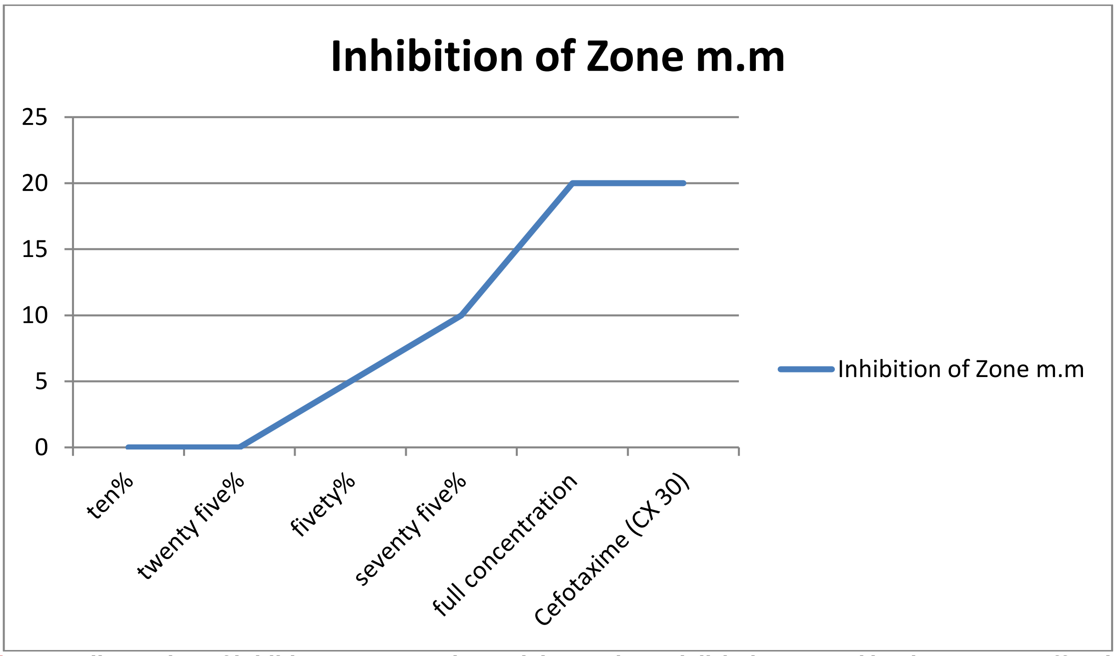

Figure 2: **Illustration of inhibitory zone on a bacterial growth petri dish that caused by the CuO NPs effect by using different concentrations**

Copper oxide nanoparticles (CuO NPs) are one of the widely used nano materials due to their beneficial qualities, including many properties such as large specific surface area, increased electrical conductivity, high toughness and ductility, luminescent efficiency, and antimicrobial property. For example, CuO nanoparticles, have been used in various products such as catalysts, gas sensors, plastics and metallic coatings, batteries for solar energy transformed and in superconductors (13). The increasing utilization of these nanoparticles, has resulted in their increased release to the environment, potentially posing risks to public health and natural environmental systems. For example, as a result of the antibacterial properties of CuO nanoparticles, CuO (NPs), have previously been reported to inhibit the denitrification processes in waste water treatment systems (14). In this case, it became clear to us that the high concentration of the solution works efficiently and with high quality with the same effect of the antibiotic, so that the inhibitory effectiveness in preventing bacterial growth reached approximately (20 mm) altogether, while the rest of the concentrations are less effective in inhibiting the growth of bacteria around discs, on the petri dishes grown in culture media. Due to various important chemical and physical properties of CuO nanoparticles, it is used in a variety of applications such as catalysis, sensors (biological, chemical, and biosensors), electrode materials, field emission (FE) emitters, photovoltaic devices, magnetic storage media, electrode materials for lithium ion battery applications, and so on. As an important heterogeneous catalyst, for many important chemical process CuO nanostructures, or (nanoparticles) are used for degradation of nitrous oxide, selective catalytic reduction of nitric oxide with ammonia, oxidation of CO at low temperature, hydrocarbon and other organic moieties in supercritical water. Various (1D) morphologies of CuO nanostructures, such as nanorods (15), nanowires, nanoneedles, (nanobelts - nanoribbons), nanotubes, and nanofibres were produced using numerous synthetic techniques. In addition to 1D copper oxide nanostructures, or nanoparticles, various complex morphologies of copper oxide, were also synthesized to name a few flower shaped nano ellipsoidal-shaped, ovel nano sheets, nano sheets, nano whiskers, 3D peanut-like patterns, urchin-like structures, nano dendrites, nano ellipsoids, dandelion-shaped hollow structures , nano spheres (16) etc. It is highly recommended using copper oxide nanoparticles as an economic alternative anti bacterial agent especially in treating ectopic infections without any risk of developing, resistant bacterial strains as with antibiotics.

CONCLUSION

The use of copper oxide nanoparticles at the medical, and biological levels and in other fields is due to its specifications that enable it to compare with other materials that are suitable for treating some medical, and industrial problems. Here in this research, a copper oxide nanoparticle was used as a substitute for the antibiotic (cefotaxin), which works to prevent the growth of bacteria E, and thus we concluded that

1. A solution of copper oxide nanoparticles; (CuO NPs) can be used at a concentration of (50-100 %) as an antibiotic due to its effectiveness in inhibiting bacterial *E.Coli* growth.
2. As a result of this inhibitory activity, of copper oxide nanoparticles (CuO NPs, they are therefore suitable as a medicine for the treatment of infectious diseases and pathological infections caused by bacteria *E.Coli*.

CONFLICT OF INTEREST

None

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