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Appraisal of antimicrobial activities of ZnO and Pb (NO₃)₂ Nanoparticles

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Abstract

Sensitivity to nanoparticles has been found to vary according to microbial activity. Agar and disc diffusion studies with *P aeruginosa* and *S aureus* showed better performance of zinc oxide nanoparticles compared with lead nitrate nanoparticles. *S aureus* describes the maximum concentration of nanoparticles compared to other pathogens and then affected by lead nitrate nanoparticles. Good correlation was observed by using MIC measurement in liquid water. MIC of zinc oxide nanoparticles against *P. aeruginosa* and *S. aureus* was found to be 5.1 µg/ml, 2.5 µg / ml, significantly lower than that compared to Pb (NO₃)₂ nanoparticles 6.5 µg/ml, 12.5 µg/ml, respectively. It can be concluded that ZnO nanoparticles increase a person's immune system resistance against microorganisms compared to other nanoparticles. The current research opens up new avenues for the biological production of Nano-materials and ZnO NPs with the potential to act as antimicrobials and control microbial pathogens such as those that appear tested by multidrug-resistant pathogens.

Keywords: Oxidative stress induction, Metal ion release, Non-oxidative mechanisms, Microbial membranes, biomedical applications, Minimum inhibitory concentration

Introduction

The emergence of Nano science and nanotechnology in the last decade presents opportunities for the exploration of the microbial effect of metal nanoparticles. Nanoparticles (NPs) are increasingly being used for disinfection as an alternative to antibiotics (Linlin, *et al.*, 2017) [1]. Nanotechnology may be particularly effective in the treatment of infectious diseases. The functional particles of the metal nanoparticles have been identified for their size and surface area to volume ratio, which allows them to interact with microbial micro-organisms and not just by releasing the metal ions in solution.

Examples include the use of NPs in the prevention of infection in medical devices and devices for preventing the spread of disease and promoting healing, in the use of drugs in the treatment of diseases, in relation to drugs for diagnostic tests, and with regard to drugs for disease control. The anti-inflammatory properties of NP are not well understood, but conventional practices include induction of oxidative stress, iron release, and non-oxidative processes. The microphone activity of iron nanoparticles is designed according to their size and surface to attract relative proportions, allowing them to interact with microbial particles, rather than releasing iron ions into solution.

Infectious symptoms, or hospital-acquired infections, are associated with hospital treatment or health care facilities. Many species of bacteria, fungi and viruses are responsible for such diseases (Horan, *et al.*, 1993) [2]. It is considered an infectious disease if it occurs within 48 hours of hospitalization or within 30 days of discharge. Improved hygiene and infection control programs have been very effective in reducing nosocomial infections.

There are many living things in a human relationship that affect a person's identity and body. The rapid and uncontrollable spread of these viruses can affect health and safety. One way to control the inflammation and spread of nosocomial infection is to wear non-bacterial clothing. When it comes to transportation, clothing is important to the chain of death penalty. All types of clothing are considered to be highly resistant to contaminants (Tinker *et al.*, 2010) [3]. Thus, the administration of antibiotics can help in the treatment of patients, people suffering from autoimmune disease, purebred patients and vulnerable infants (Tinker *et al.*, 2010) [3]. Antibiotics can help those who come in contact with patients, such as visitors, nurses, doctors, and other medical staff.

The Sonochemistry center at the University of Coventry is one of a group of 17 industrial firms working on a funded project, aimed at developing a pilot line for industrial practice for the coating of antimicrobial nanoparticles on fabric surface by ultrasound. The technology being developed is based on a laboratory scale son chemical system developed at Bar Ilan University (BIU), Israel. The method uses a single step to blend and disintegrate the antibacterial nanoparticles into the fabrics using ultrasonic waves. "Ultrasonic stirring has been found to be an effective method for dispersing nanoparticles to the surface of various metals including clays and polymers" (Abramov 2009) [4]. This process results in a smooth and homogeneous coating and enables the production of nanoparticles to the fabric surface at very high speed. The speed of the strong city forced them to cling tightly to the platforms.

We hope to be able to produce this fabric at an affordable price for general hospital use such as bandages, hospital flyers and uniforms. Synchronous synthesis and coating of antimicrobial agents reduces the production steps and therefore energy and production costs. Many chemicals can be recycled instead of being reused. These types of treatments are relatively practical and durable because the antimicrobial agent is physically attached to the fabric structure and is released slowly during use. Antimicrobial agents are natural or synthetic compounds that inhibit microbial growth. The textile industry uses a wide variety of antimicrobial agents, mainly biocides.

Antibacterial activity of metal nanoparticles

The use of metal nanoparticles has been rapid due to the amount of work involved in the preparation and modification of biomedical particles. Many heavy metals and iron oxides are toxic to germs in the free zone or at very low levels (Padmavathy, *et al.*, 2008) [20] these abnormal substances kill bacteria in a variety of ways, such as binding and blocking cellular proteins, the formation of active oxygen species and damage to cell walls (Gao, *et al.*, 2008) [6]. Zinc oxide (ZnO), copper oxide (CuO), magnesium oxide (MgO), titanium dioxide (TiO₂), and silver (Ag) are the most commonly used antimicrobials in the production of antimicrobial clothing. Nanoparticles are particles the size of 100 nanometers. The formation of nanoparticles alters the properties of many common materials (Reddy, *et al.*, 2007) [7].

For example, the large surface area of nanoparticles can cause significant interactions with bacterial cell walls. Of the raw materials, iron and iron oxides have been of great interest to antimicrobials due to their durability, selectivity and heat resistance. One of the reasons why these non-functional materials (iron and iron oxides) receive a lot of attention is their ability to withstand extreme working conditions (Fu, *et al.*, 2005) [8]. They contain toxins that secrete bacteria but have minimal side effects on human cells (Reddy, *et al.*, 2007) [7].

Zinc oxide (ZnO) belongs to a group of oxides against photo oxides and photo catalytic chemicals in biological species. This inorganic white powder and dirt is insoluble in water. Nanoparticles of ZnO and Pb (NO₃)₂ have shown their effectiveness in inhibiting microbes and antifungals when used in tissue coatings and materials. Zinc is an important component of ZnO nanoparticles and is considered non-toxic. Toxic studies have shown that zinc ions do not damage the DNA of human cells (Yamada, *et al.*, 2007) [10]. Study and evaluating the antibacterial activity of ZnO and its derivatives, found that ZnO lions (10–50 nm) had a better

antimicrobial effect than ZnO (2µm). The antibacterial activity of ZnO nanoparticles is small due to their interaction with electrostatic precipitators. It showed that under the cytostatic effect of ZnO nanoparticles in contact with bacteria, the bilayer layer of lipids is destroyed, disrupting the content of the cytoplasm.

Antibacterial test

The research efforts and research materials of leading scientists and technologists in the most complex sciences and nanotechnologies have succeeded in creating highly organized nanoparticles of Nano constructions of all shapes and sizes. He provided an approach to the destruction of nanomaterials that can produce biological properties in biological systems. It showed several studies that allow the use of nanoparticles as an effective bactericidal agent (Tiller, *et al.*, 2001) [11].

The antibacterial properties of iron oxide (Pb (NO₃)₂ and ZnO) Nano powders in many media have been measured (Sawai 2004) [12]. It is believed that the active forms of oxygen produced by these metal nanoparticles may be the main mechanism / cause of their antibacterial action. In the case of mycosis, Nano fragments can be considered antibacterial, anti-fungal. However, only a few studies have been conducted to evaluate the effect of Nano-structured materials on *P. aeruginosa* and *S. aureus*. Therefore, the aim of this study was to evaluate the toxic effects of Pb (NO₃)₂ and ZnO Nano element (chemical compounds used) in laboratory conditions. Among the selected items such as Gram negative / Gram positive, the risk assessment and the environmental and related environmental significance should be most effective.

The interaction of biomolecules with microbial molecules containing nanoparticles is an expanding research that is currently unknown. In this study, Pb (NO₃)₂ and ZnO nanoparticles showed a wide range of antibiotics for a wide range of pathogens, including critical pathogens, under normal conditions. This study examines the toxicity of Pb (NO₃)₂ and ZnO (salt based) nanoparticles and their resistance to bacterial pathogens.

Materials and methods

1. Nutrient agar
2. LB broth
3. Bacterial culture:
 - *Pseudomonas aeruginosa*
 - *Staphylococcus aureus*
4. Nanoparticle salt solution
 - ZnO and
 - Pb(NO₃)₂ NPs

The aggressive strains used here are Gram negative *Pseudomonas aeruginosa* and Gram positive *Staphylococcus aureus*. Bacteria purified from these test bacteria were purchased from microbial culture kits, the Institute of Microbial Technology and Chandigarh. Cultures in the NA (nutrient) system were administered at 37°C. Both cultures were maintained by taking a phase based on NA medium (equivalent) and storage (4°C). Previously used in the experiment. Antibacterial resistance of each bacterium was performed using a therapeutic medium, agar in a Petri dish, to investigate absorption and excretion. Using the agar / vegetative agar / NA system, we have developed surface colony growth for bacterial screening. For bacterial cell proliferation, suspension cultures were prepared using 2% Lauria Broth (w / v) collected for review.

1. LB media material – (for 1000 ml solution)

Peptone	5gm
Beaf extract	3gm
NaCl	5gm
pH	7.2

LB media suitable for bacterial culture.

2. Nutrient agar - (for 1000 ml solution) 40gm**Agar well diffusion and Disc method**

Biosynthetic ZnO and Pb (NO₃)₂ nanoparticles were also used to measure cell activity in vitro. Antibacterial properties of iron oxide nanoparticles were accomplished by methods and well-qualified synthesis testing. Kirby-Bauer is a semi-quantitative study used for bacterial pathogens. This method is simple and relatively fast to ensure the antibacterial property of diffuse antibacterial agents treated with fibrous materials (Bauer, *et al.*, 1959) [14]. In this experiment, a useless Petri cup was used and approximately 20 ml of fresh agate (NA) was poured into the cup. The plates were left overnight at 37°C to check for damage. Each bacterium (*Pseudomonas aeruginosa* and *Staphylococcus aureus*) was treated with Loria broth. Each sterilizer expanded for 4 h was used to wipe the nutrient agar plate with the help of a non-abrasive detergent.

The sterilized discs (10 mm in diameter) test cloth with two different sizes (0.5mM and 2mM over ZnO NPS) and control cloth (paper disks without ZnO nanoparticle) were applied and then gently pressed N on the plate on top. Tablets were then incubated for 18–24 hours at 37°C. The antibacterial suggests that the travel of the virus was expressed through the diameter of the interfering branches compared to the control of the fabric.

To test the efficacy of Pb (NO₃)₂ NP, make 2 wells (10 mm wide) on all covered plates (20 ml purified agar (NA) in clean non-gas canisters) using driving training. Approximately 10–80 µl at different concentrations (0.5mM and 2mM) Pb (NO₃)₂ (prepared) NP was carefully added to wells controlled by pipes and allowed to soak for 2 hat room temperature. Experiments were performed with Pb (NO₃)₂ NP injection. The plates were left to soak for 18–24 h at 37°C for bacterial germination. Closed charts were checked to block areas that appeared to be an upper ring around discs and wells. Using a mechanical scale, the area of each region is measured and the value of each region is recorded, sorted, and expressed in millimeters. After measuring the width of the limiting areas (mm), The besots index was also determined. The three components were saved correctly and the test was tested three times. According to the order in which each image was displayed, the comparison was made in three subtractions and the total values were given.

Preparation and incubation of test in Coolum's

The micro dilution method was used to determine the antibacterial test. In this method, the antibacterial identifications of certain compounds were tested against several pathogenic bacteria.

- Two bacterial suspensions used in this experiment (*Pseudomonas aeruginosa* and *Staphylococcus aureus*) were prepared by picking a colony from each bacterial culture store with a loop button and inoculating a valuable 20 ml (sterile) pregnancy nutrient broth into a 100 ml Erlenmeyer flask.
- After inoculation, the flasks were incubated using a 110 rpm shaker incubator at 37°C for 24 ± 2 hours.

- Both inoculums after overnight incubation, 0.4 ml each were transferred to 20 ml nutrient broth pregnant in 100 ml Erlenmeyer flasks (two) and incubated again with shaking the incubator at 110 rpm at 37°C for 3 hours.
- then a 3-fold serial dilution of the AIT ammunition of each sample obtained was carried out in a pregnant nutrient medium, observation of pregnancy compared to the approximate target concentration of host tissue., Between 1 k 105 colonies which make up the units / AIT (UFC / AIT) and 3 k 105 UFC / AIT.
- At 3 hours of incubation Culture, the number of bacteria was estimated by measuring the D.O. (Optical density) in culture / sample at approximately 660 (Corning 253 colorimeter). Nodding its head between 0.1 and 0.3 was almost equal to the conception concentration between 1 k 108 CFU / al and 3 k 108 CFU / al.
- The inoculum is prepared, stored and stored (4°C) until use. In order to fundamentally verify the absence of contamination and pregnant women to verify the validity of the inoculum, the dilutions of the inoculum are properly grown on a solid medium. All steps of this experiment were performed in duplicate and repeated three times.

Determination of Minimum Inhibitory Concentration (MIC)

MIC (minimum inhibitory concentration) is the lowest concentration of drugs / chemicals that can prevent the apparent growth of a microorganism on cultural plates. The MIC culture can be determined from the readings obtained on the plates after night incubation. The most commonly used methods of controlling concentration are agar dilution methods and tube dilution methods. Serial dilutions of the product occur in the bacterial environment. The test organisms / bacteria are scored to be added to the product dilutions, incubated and grown. The method used is the standard antimicrobial test. MIC is important in many applications, such as in diagnostic laboratories, where MIC enhances the resistance of microorganisms to antimicrobial agents. It has also been used to monitor the activity of new antimicrobials. MIC (minimum inhibitory concentration) is generally considered to be a very basic laboratory measurement related to the activity of an antimicrobial agent against an organism.

Resazurin (indicator) preparation

Resazurin sodium salt powder according to Sigma Aldrich, the product solution was prepared 0.02% (w / v) by using distilled water and stored at 4°C for weeks 1 (Reham, *et al.*, 2013) [16]. A sequential dilution technique was used to stabilize the microphone in the laboratory. Lightly modified 96-fine microplates were used in this method (Sarker, *et al.*, 2007) [17]. The dishes were prepared under aseptic conditions. Bacterial strains (*Pseudomonas aeruginosa* and *Staphylococcus aureus*) were grown overnight in equilibrated plants at 37°C. Muller Hinton broth was used in this experiment.

A 1 mg/ml solution of each NP (ZnO and Pb (NO₃)₂ NP) was prepared in methanol. Test bacteria (*Pseudomonas aeruginosa* and *Staphylococcus aureus*) at a concentration of 3 x 10⁸ CFU / ml were prepared as described in section 3.6 above. In a 96-well plant titration vessel, up to 11 wells were mixed with 50 µl of Mueller Hinton's broth (MHB) and 50 µl (NP) solutions (each) and then mixed with 10 µl of cell-treating bacteria at 37°C, 24 hours.

The final volume of each well is 110 μ l. The 12 springs were used as regulators containing the inoculum. After incubation, 10 μ l of resazurin was added to each well. The plates were then reattached to their temperature for 2 hours. A color change was observed after 2 hours. Each dish was packed in pictures to ensure it was contaminated. After 72 hours at 28°C, the plates were monitored for results. In the presence of appropriate cells, resazurin blue is reduced to pink. The low-order sequence inhibited the color change, a sequence that acted as an inhibitory agent for each cell. The experiment was performed in pairs and the standard deviation was calculated from the standard deviation.

Results and discussion

Antimicrobial activity of ZnO nanoparticles

The ability of an antibacterial agent to inhibit the growth of bacteria or the antibacterial property of zinc oxide nanoparticles (via a fungus) was evaluated using a disk diffusion method. Two different concentrations of 0.5mM and 2mM ZnO nanoparticles were taken. 10mm paper discs with and without zinc oxide nanoparticles (control) were tested and the results obtained are shown in Table 1. The antibacterial property of zinc oxide NC synthesized by *Escherichia coli* (bacteria) and *Aspergillus niger* (fungus) extract was tested against *Pseudomonas aeruginosa* (Gram negative) and *Staphylococcus aureus* (Gram positive), with maintenance control (paper discs without zinc oxide coating). The antibacterial effect of (0.5mM and 2mM) ZnO NP was determined based on the zone of inhibition (mm) shown in Figures 1 and 2.

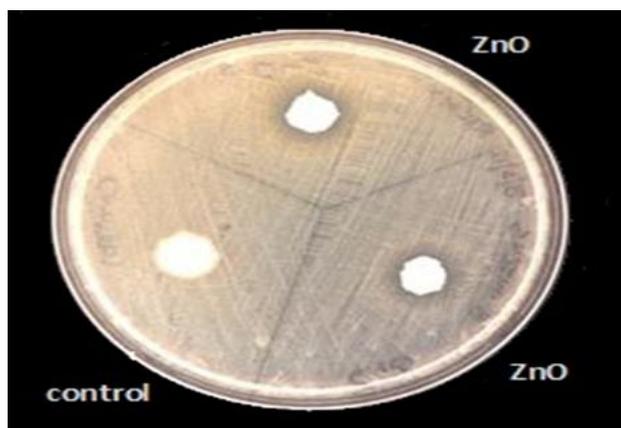


Fig 1: Antibacterial activity of ZnO NPs against *S. aureus*

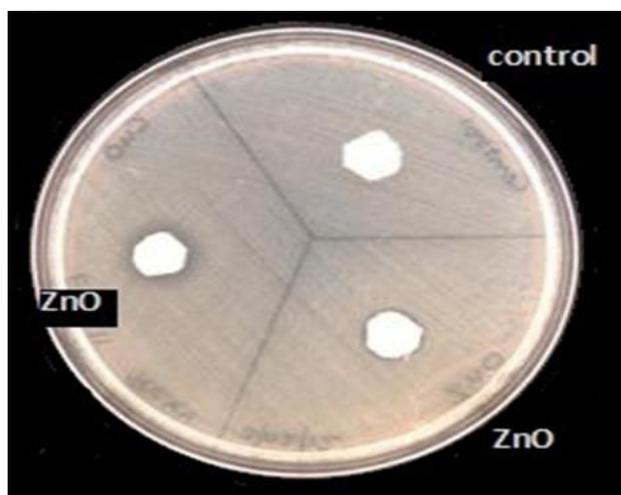


Fig 2: Antibacterial activity of ZnO NPs against *P. aeruginosa*

Table 1: Antibacterial assessment by Disc diffusion method

Fabrics Treated	Organism	Compound In mm		Zone of Inhibition (mm) Diameter	
		0.5	0.2		
ZnO NPs	<i>S aureus</i>	0.5	0.2	15	19
	<i>P aeruginosa</i>	0.5	0.2	13	14
Control	<i>S aureus</i>	0	0	0	0
	<i>P aeruginosa</i>	0	0	0	0

The ZnO NP suspension used in this test is different from TEM and SEM (Figures 3A, 4A). ZnO NPs vary in size from 30 to 70 nm and have a spherical or spherical structure. In the absence of ZnO, capillaries had no effect on bacterial growth and a common colony was observed. To evaluate the effect of ZnO NP on *S. aureus* and *P. aeruginosa*, corresponding bacterial cultures were cultured in NA (food agar) dishes containing different concentrations of zinc oxide NP (0, 5 mm and 2 mm) were introduced overnight in 37°C before counting Cells.

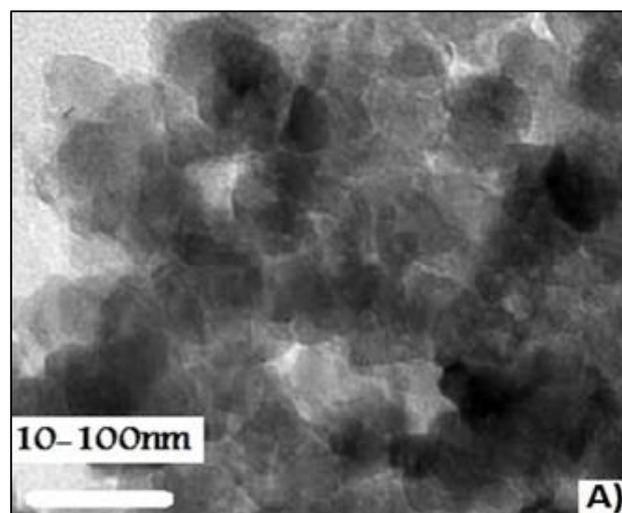


Fig 3A: TEM micrograph of ZnO

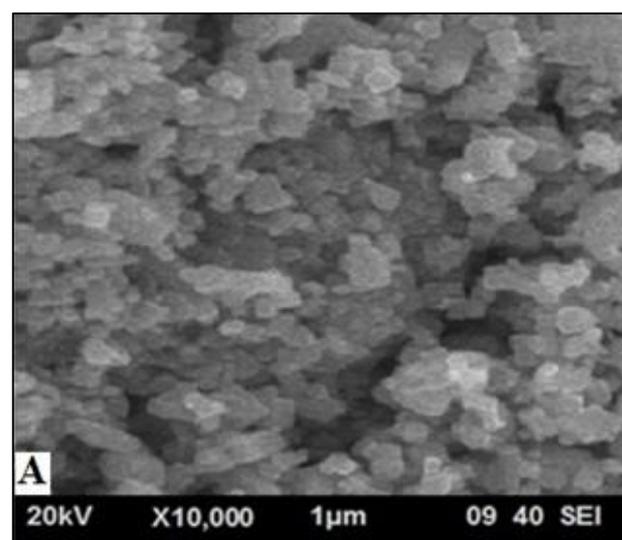


Fig 4A: SEM micrograph of ZnO

The figures show that ZnO nanoparticles improve antimicrobial activity against different pathogens as they increase in size. It concludes by drawing with two different types of zinc oxide nanoparticles showing the high area of the limit. *Staphylococcus aureus* was a gram-positive bacterium that showed a peak of 19 mm ZnO NP, and *Pseudomonas aeruginosa* was a gram-negative bacterium that showed a 14

mm inhibition area with ZnO NPs. Thus, the greatest impact was received against *Staphylococcus aureus*, followed by *Pseudomonas aeruginosa*.

MIC (Minimum Stress Depression) was defined as the highest / lowest concentration of zinc oxide nanoparticles that prevented the growth of pathogens. MIC is important in many applications, such as laboratory diagnostics, where MIC confirms the resistance of microorganisms to an antimicrobial agent. It is also used to monitor new

The minimum content of ZnO NPs that make up the complete pathogen, primarily obtained by MIC, is provided in Table 2.

Table 2: Determination of MIC for ZnO (Zinc oxide) nanoparticles

S. No	Name of the Micro organism	MIC ($\mu\text{g/ml}$)
1	<i>S. aureus</i>	2.5
2	<i>P. aeruginosa</i>	5.1

According to a study by Yamamoto *et al.* In 2000, zinc oxide nanoparticles could form ROS (reacted oxygen species) responsible for their inflammatory actions (Yamamoto, *et al.*, 2000) [18]. It was also further investigated that the interaction of the reaction with hydrogen peroxide is responsible for the antimicrobial action of zinc oxide nanoparticles. As well as the bactericidal properties of nanomaterials may be due to the interaction of other compounds formed in the presence of zinc oxide nanoparticles and the outer lipid of the bilayer Produced hydrogen peroxide enters cell membranes and allows them to

kill. This study found that ZnO complex nanoparticles are responsible for inhibiting the growth of bacteria (Li, *et al.*, 2007) [19].

Other bacterial nanoparticles from zinc oxide have been further elucidated. This study showed that when hydrogen peroxide was formed from ZnO NP, the nanoparticles were in good contact with dead bacteria to prevent infectious diseases as well as to release hydrogen peroxide in moderation (Padmavathy and Vijayaraghavan, 2008) [20]. The results of the study are related to the author's findings and suggest that zinc oxide nanoparticles used in animals have good antimicrobial properties.

Antimicrobial activity of Pb (NO₃)₂ nanoparticles

The antibacterial behavior of Pb (NO₃)₂ NP synthesized by a fungal extract (*Aspergillus Niger*) was controlled using the agar well diffusion method (10 μl shown in Table 3) 2 different concentrations of 2 NP-free LB pulp. The antibacterial activity of (0.5mM and 2mM) Pb (NO₃)₂ NP was determined from the inhibition zones (mm) shown in Figures 5 and 6. From the figure, it can be concluded that Pb (NO₃)₂ NP showed a high inhibition zone as a standard for streptomycin antibiotics. *Staphylococcus aureus* is a gram-positive bacterium, and the maximum inhibition zone is 19 mm with Pb (NO₃)₂ NP, and *Pseudomonas aeruginosa* is a gram-negative bacterium with an inhibition zone of Pb (NO₃)₂ NP of 10 mm.

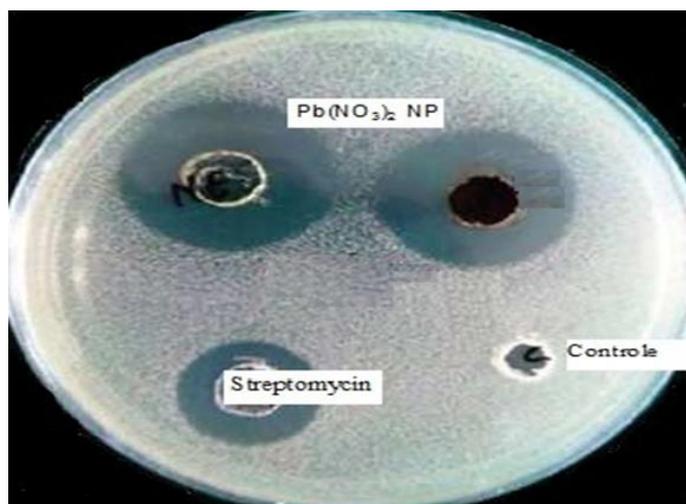


Fig 5: Antibacterial activity of Pb (NO₃)₂ NPs against *S. Aureus*

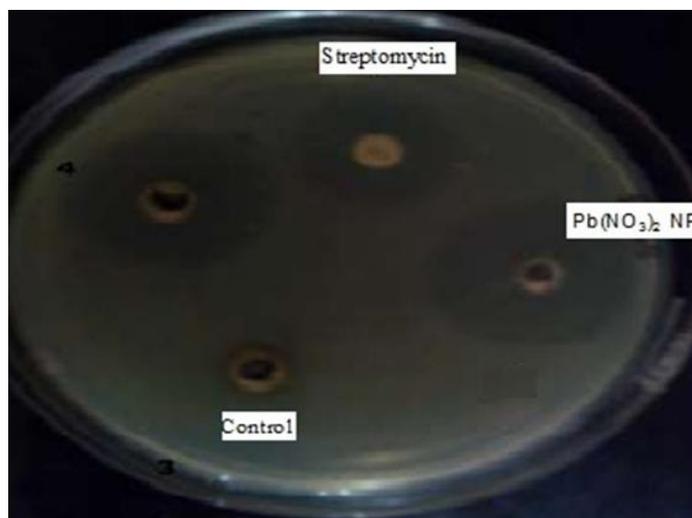
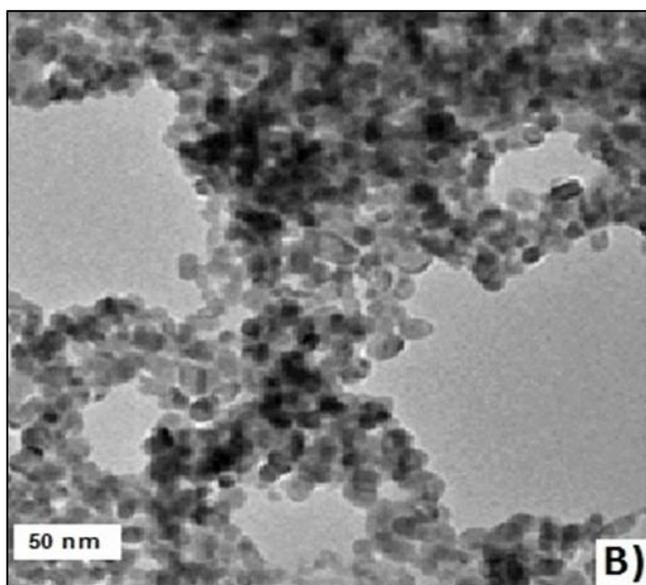
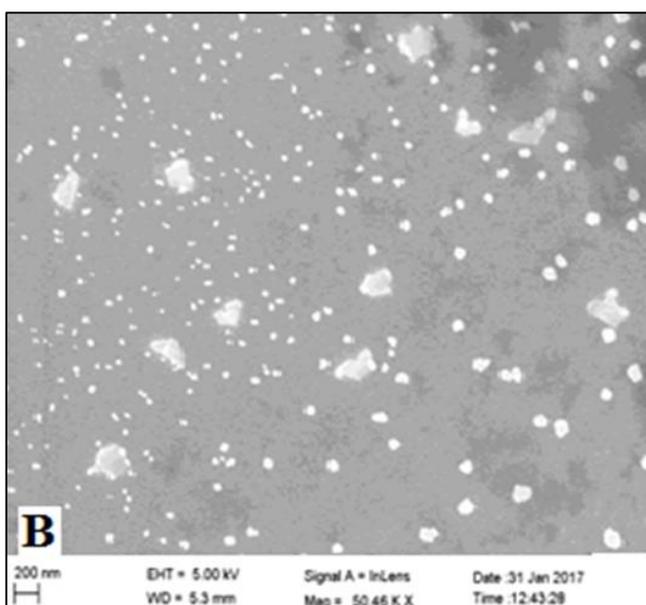


Fig 6: Antibacterial activity of Pb (NO₃)₂ NPs against *P. Aeruginosa*

Table 3: Antibacterial assessment by agar well diffusion method

Fabrics Treated	Organism	Compound In mm		Zone of Inhibition (mm) Diameter	
Pb(NO ₃) ₂ NPs	S aureus	0.5	0.2	19	15
	P aeruginosa	0.5	0.2	10	11
Control	S aureus	0	0	0	0
	P aeruginosa	0	0	0	0

TEM Figure, Figure 7 (B) shows that Pb (NO₃)₂ nanoparticles (synthesized by the fungus *A. Niger*) are almost spherical and stick-shaped. The particle size is in the range of 10-50 nm. Large estimate of the synthesized nanoparticles was characterized by a transmission electron microscope using a Jeol Jsm 100cx instrument (Jeol Ltd., 1400, Tokyo, Japan). SEM analysis clearly shows the existence of synthesized lead nitrate Pb (NO₃)₂ nanoparticles. Pb (NO₃)₂ NPs are very small, in the range of 10-45 nm, and some of them are aggregated, as shown in Figure 8 (B).

**Fig 7B:** TEM micrograph of Pb (NO₃)₂**Fig 8B:** SEM micrograph of (B) Pb (NO₃)₂

MIC (Minimum Inhibitory Concentration) is defined as the highest / lowest concentration of lead nanoparticles that inhibits the growth of an organism. In diagnostic laboratories,

the determination of MIC is important because of the resistance of microorganisms to a confirmed antimicrobial agent. MIC is able to monitor the properties of new antimicrobial agents. The minimum concentration of Pb (NO₃)₂ NP that completely killed the bacteria was taken as MIC respectively, shown in Table 4.

Table 4: Determination of MIC for Pb (NO₃)₂ NPs

S. No	Name of the Micro organism	MIC (µg/ml)
1	<i>S. aureus</i>	6.5
2	<i>P. aeruginosa</i>	12.5

The findings of the current investigation are consistent with past investigations managing the antimicrobial effects of nanomaterials. Nanoparticles induce peroxidation of phospholipid polycyclic compounds in the lipid layer of microorganisms, so the probability of cell membranes decreases, and ordinary cells move in solid cellular structures. (Shrivastava, *et al.* 2010) [21] Analyzed the Nano silver antimicrobial effect on *S. aureus* in 2010 and demonstrated that the Nano silver antimicrobial effect of the particles relies on measurement and is more successful against gram-negative microorganisms than against gram-positive microorganisms (Shrivastava, *et al.*, 2010) [21]. This is consistent with the results of this test.

Matthews *et al.* Can be used to treat and completely suppress high levels of Ag nanometer-scale Gram-positive and Gram-negative outbreaks in a study entitled "Application of Nano medicine to Antibacterial Treatment and Diagnosis", 2010. I expressed it as. Bacterial species consistent with the sequelae of silver exploration (Matthews, *et al.*, 2010) [22].

A study entitled "Nano-is a Colloidal Fusion and Antibacterial Effect" found that most of the microorganisms were crushed by treatment with Ag nanoparticles, and most of the microorganisms were crushed due to the low concentration of Ag nanoparticles. Developmental deficits resulted in a remarkable reduction in the measurement of living microorganisms as opposed to contrast tests comparing the antibacterial effects of NPs with the microbial copper of *P. aeruginosa* and *S. aureus* 2545. (Guangyin, 2007) [23].

Consistent with the results of this test. The cooperation of conceivable reaction components, nanomaterials and organic macromolecules along these lines releases particles from which the nanomaterial reacts with proteins from the surface of bacterial cell (-SH) Thiols. This protein swells in the bacterial cell layer and replaces supplements in the cell splitter. Nanomaterials interfere with these proteins, reduce film permeability, and facilitate cell passage during long-distance travel.

Conclusion

Some metal oxide nanoparticles have demonstrated excellent antibacterial activity. The biological approach used in this study appears to be a cost-effective alternative to the chemical and physical methods of producing existing nanoparticles. Therefore, it is suitable for the development of commercial large-scale microbial processes. Production of metal oxide nanoparticles. The microorganisms used for antibacterial

action are *Staphylococcus aureus* and *Pseudomonas aeruginosa*. The antibacterial activity performance of ZnO and Pb (NO₃)₂ nanoparticles was performed using the disc method and the agar diffusion assay. The method used for the antibiotic susceptibility test was the Kirby Bauer method, and the medium prepared was a trophic agar medium that was rigorously tested for pH and composition. The depth of the agar in the plate is a factor and is also taken into account by the Kirby-Bauer method [Bauer, *et al.*, 1959]^[14]. This method is well documented and standard inhibition zones have been determined for resistance and susceptibility values.

Some metal oxide nanoparticles have demonstrated excellent antibacterial activity. The biological approach used in this study appears to be a cost-effective alternative to the chemical and physical methods of producing existing nanoparticles. Therefore, it is suitable for the development of commercial large-scale microbial processes production of metal oxide nanoparticles

The synthesized metal oxide nanoparticles showed effective antimicrobial activity against several pathogens. Size and concentration are two important factors affecting the antimicrobial properties of Pb (NO₃)₂ and ZnO NP. According to, it's possibility to exist in the form of aggregates during the synthesis of nanoparticles (He, *et al.*, 2007)^[24]. In this study, nanoparticles can be deposited in the broth, but the location agar medium in the broth makes it difficult to estimate the actual exposure concentration of Pb (the disk method and agar diffusion analysis were selected for the antibacterial properties of synthetic nanomaterials) And ZnO NP. Mixing the NP dispersion with the desired medium solution destroyed the dispersion system in most cases and was shown to allow for the slow settling of the NP of the medium solution. Agar medium was used very much to overcome this problem in this experiment. This takes into account that the sedimentation process of nanoparticles is much slower compared to the intermediate solidification process. In this experiment, a prepared broth culture of several pathogens aged 4 hours was used to wipe the nutrient agar plate using a sterile cotton swab to add nanoparticles of each of the following two concentrations (0.5mM and 2mM). Because the agar system is well suited for a variety of experiments, few previous studies have used the agar system (Brayner, *et al.*, 2006)^[25].

Microorganisms used for antibacterial action are *Staphylococcus aureus* and *Pseudomonas aeruginosa*. The antimicrobial activity performance of ZnO and Pb (NO₃)₂ nanoparticles was performed using the disk method and agar diffusion assay. The method used for the antibiotic susceptibility test was the Kirby Bauer method, and the prepared medium was a nutrient agar medium that was rigorously tested for pH and composition. The depth of the agar in the plate is a factor and is also considered by the Kirby Bauer method [Bauer, *et al.*, 1959]^[14]. This method is well documented and standard areas of inhibition for the values of tolerance and susceptibility have been determined.

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