وزارة التعليم العالي و البحث العلمي

جامعـــــــة بابـــــل/كلية الصيدلة

The effect of nanoparticles on bacterial DNA

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Abstract

The antibiotic resistance pathogens have become a serious health issue and thus, numerous studies have been reported to improve the current antimicrobial therapies The past decade has witnessed a substantial upsurge in the global use of nanomedicines as innovative tools for combating the high rates of antimicrobial resistance. Antibacterial activity of metal and metal oxide nanoparticles (NPs) has been extensively reported. The microbes are eliminated either by microbicidal effects of the NPs, such as release of free metal ions culminating in cell membrane damage, DNA interactions or free radical generation, or by microbiostatic effects coupled with killing potentiated by the host's immune system. NPs possess many mechanisms of antimicrobial activity against bacteria like: alteration of bacterial cell membrane, respiratory chain disruption, Protein and DNA damage and oxidative stress by free radical production). NPs exhibite effect of genetic materials and there are many types of these damage such as sugar lesions, base lesions,protein and DNA crosslinks, single and double strand breaks are produced by free radical induced reactions and inhibition of DNA replication by binding to the DNA. The aim of this review is to study the effects of nanoparticles on bacterial DNA. The current status of nanoparticle use in pharmacology and therapeutics and their effects on bacterial DNA.

Introduction

Nanotechnology refers to the creation and utilization of materials whose constituents exist at the nanoscale ; and by convention be up to 100 nm in size. Presently, different metallic nanomaterials are being produced using copper, zinc, titanium, magnesium, gold, alginate and silver. Nanoparticles are being used for diverse purposes, from medical treatments, using in various branches of industry production such as solar and oxide fuel batteries for energy storage, to wide incorporation into diverse materials of everyday use such as cosmetics or clothes [1]

Nanoparticles (NPs) are materials that are small enough to fall within the nanometric range, with at least one of their dimensions being less than a few hundred nanometers. This reduction in size brings about significant changes in their physical properties with respect to those observed in bulk material [2].
They exhibit unique physical properties (such as particle aggregation and photoemission, and electrical and heat conductivities) and chemical properties (such as catalytic activity), and hence have received much attention from scientists and researchers in different areas of biological sciences.[3] Nanoparticles (NPs) have unique physicochemical properties which make them promising platforms for drug delivery. However, immune cells in the bloodstream (such as monocytes, platelets, leukocytes, and dendritic cells) and in tissues (such as resident phagocytes) have a propensity to engulf and eliminate certain nanoparticles in the situation when specific delivery to immune cells is not desired, the ideal nanoparticle platform is the one whose integrity is not disturbed in the complex biological environment, which provides extended circulation in the blood to maximize delivery to the target site, is not toxic to blood cellular components, and is "invisible" to the immune cells which can remove it from circulation.[4]

The worldwide escalation of bacterial resistance to conventional medical antibiotics is a serious concern for modern medicine. High prevalence of multidrug-resistant bacteria among bacteria-based infections decreases effectiveness of current treatments and causes thousands of deaths. New improvements in present methods and novel strategies are urgently needed to cope with this problem. Owing to their antibacterial activities, metallic nanoparticles represent an effective solution for overcoming bacterial resistance. However, metallic nanoparticles are toxic, which causes restrictions in their use. Recent studies have shown that combining nanoparticles with antibiotics not only reduces the toxicity of both agents towards human cells by decreasing the requirement for high dosages but also enhances their bactericidal properties. Combining antibiotics with nanoparticles also restores their ability to destroy bacteria that have acquired resistance to them. Furthermore, nanoparticles tagged with antibiotics have been shown to increase the concentration of antibiotics at the site of bacterium–antibiotic interaction, and to facilitate binding of antibiotics to bacteria. Likewise, combining nanoparticles with antimicrobial peptides and essential oils generates genuine synergy against bacterial resistance.

NPs need to be in contact with bacterial cells to achieve their antibacterial function. The accepted forms of contact include electrostatic attraction, van der Waals forces, receptor–ligand and hydrophobic interactions. NPs then cross the bacterial membrane and gather along the metabolic pathway, influencing the shape and function of the cell membrane. Thereafter, NPs interact with the bacterial cell's basic components, such as DNA, lysosomes, ribosomes, and enzymes, leading to oxidative stress, heterogeneous alterations, changes in cell membrane permeability, electrolyte balance disorders, enzyme inhibition, protein deactivation, and changes in gene expression. The following mechanisms are the most frequently proposed in current research: oxidative stress, metal ion release, and non-oxidative mechanisms.

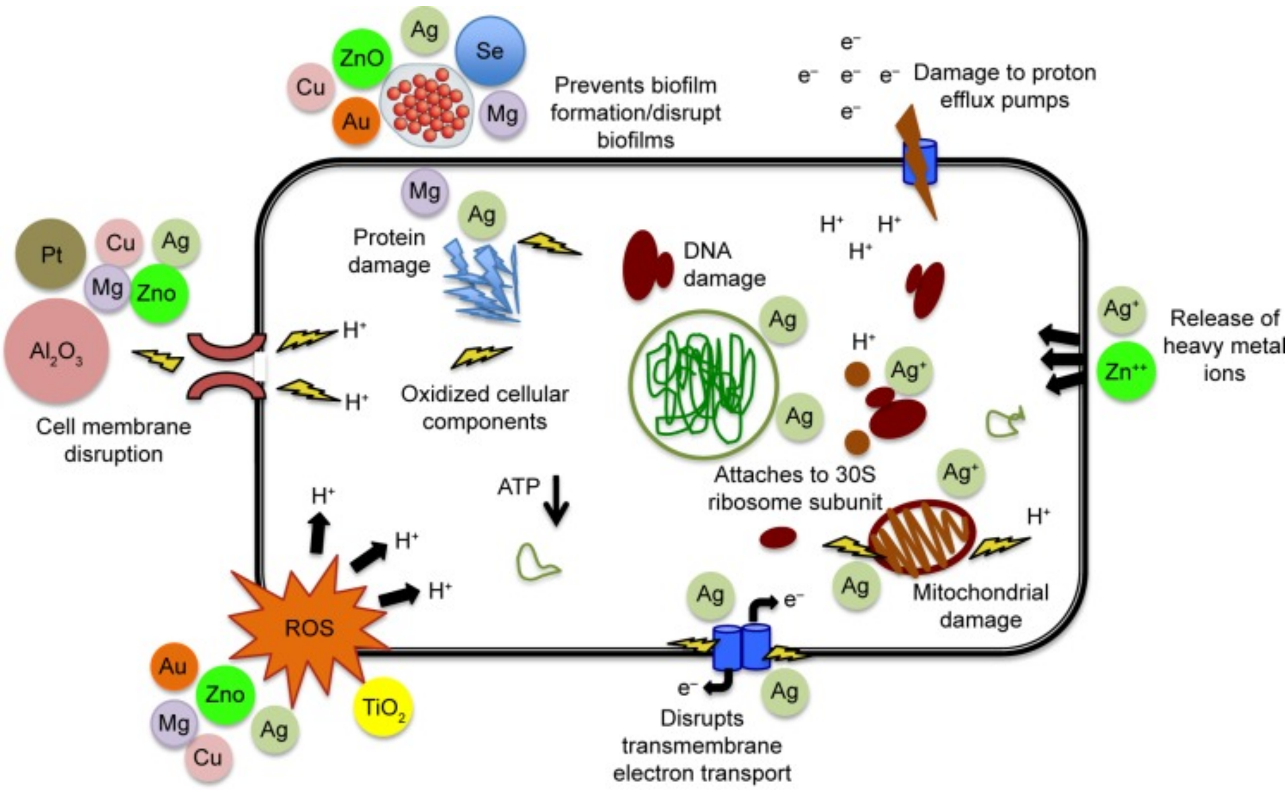

Fig 1: Probable nanomaterials-based bactericidal effects

Applications

Nanoparticles are portable, cheaper, safer, and easier to administer[5].
They have been used in vivo to protect the drug entity in the systemic circulation, restrict access of the drug to the chosen sites and to deliver the drug at a controlled and sustained rate to the site of action[6]

A list of some of the applications of nanomaterials to biology or medicine is given below:
Fluorescent biological labels
Drug and gene delivery
Bio detection of pathogens
Detection of proteins
Probing of DNA structure
Tissue engineering
- Tumour destruction via heating (hyperthermia)
- Separation and purification of biological molecules and cells
- MRI contrast enhancement
Phagokinetic studies
Protein detection [7]

The aim OF THE PROJECT

The aim of this review is to study the effects of nanoparticles on bacterial DNA. The current status of nanoparticle use in pharmacology and therapeutics and their effects on bacterial DNA.

The effect of nanoparticles on genetic materials

The DNA of most bacteria is contained in a single circular molecule, called the bacterial chromosome. The chromosome, along with several proteins and RNA molecules, forms an irregularly shaped structure called the nucleoid. This sits in the cytoplasm of the bacterial cell.
In addition to the chromosome, bacteria often contain plasmids – small circular DNA molecules. Bacteria can pick up new plasmids from other bacterial cells (during conjugation) or from the environment. Plasmid help bacteria to survive stress
Many plasmids contain genes that, when expressed, make the host bacterium resistant to an antibiotic (so it won't die when treated with that antibiotic). Other plasmids contain genes that help the host to digest unusual substances or to kill other types of bacteria. Plasmid DNA (pDNA) can appear in one/ or some of five conformations, Nicked open- circular, Relaxed circular, Linear, Supercoiled and Supercoiled denatured, in the as given order of electrophoretic mobility from slowest to fastest, respectively.
Mechanisms by which NPs exhibit their antimicrobial activity against bacteria include:

(i) Disruption of bacterial cell membrane integrity
(ii) Induction of oxidative stress by free radical formation
(iii) Mutagenesis
(iv) Protein and DNA damage
(v) Inhibition of DNA replication by binding to DNA
(vi) Respiratory chain disruption

ROS-induced oxidative stress is an important antibacterial mechanism of NPs. ROS is a generic term for molecules and reactive intermediates that have strong positive redox potential, and different types of NPs produce different types of ROS by reducing oxygen molecules. The four ROS types are the superoxide radical , the hydroxyl radical (·OH), hydrogen peroxide (H2O2), and singlet oxygen (O2), which exhibit different levels of dynamics and activity. For example, calcium oxide and magnesium oxide NPs can generate O_2^-, whereas zinc oxide NPs can generate H2O2 . [8]
Several types of damage, including base lesions, sugar lesions, protein and DNA crosslinks, single-strand breaks and double strand breaks are produced by free radical induced reactions [9].
ROS cause Base oxidation, particularly guanine, and block lesions or strands break which may be lethal unless they are repaired. iron-sulfur cluster- containing proteins are also vulnerable to ROS damage and may substantially restrict metabolic pathways even if the damage is not microbicidal. The presence of SOD in the periplasm has suggested the existence of extracytoplasmic O2. targets [10] .
Another proposed mechanism is AgNPs can result in DNA damage through shrinkage of the cytoplasm membrane or its detachment from the cell wall. As a consequence, the DNA molecules are condensed and their ability to multiply is reduced. [11]

NPs exposed to bacterial cells have been shown to cause changes in the genomic and proteomic profiles, suggesting that the presence of NPs primes an adaptation of the cells to the new NP-containing environment. For example, when Ag-NPs and Ag+ were exposed to bacterial cells, an upregulation of a shared 161 genes and downregulation of 27 genes in E. coli were observed. Interestingly, Ag-NPs and Ag+ exclusively regulated 309 and 70 genes, respectively. Another study reported that E. coli treated with Ag-NPs upregulated many genes covering a wide range of functions such as membrane structure and biofilm formation (bolA), the citric acid cycle (sdhC), electron transfer (sdhC), cellular transport (mdfA), protein efflux (fsr, yajR, emrE), and DNA repair (recN, uvrA, ybfE, yebG, ssb, sbmc, and nfo).[12]

Table(1) The different types of metal nanoparticles and their effect on bacterial DNA

	NP type	Bacterial type	Results	Sources
1	**Copper based Nanoparticles**	Gram-positive (*Bacillus subtilis*, *Bacillus cereus*, *Staphylococcus aureus*) and Gram-negative(*Xanthomonas campestris*, *E.coli*)	Inhibition of bacterial growth by The Cu-based NPs induce pDNA degradation in a dose- dependent manner as well as extensive ds CT-DNA degradation	[13]
2	**Copper oxide Nanoparticles**	*B. subtilis*, *B.cereus*, *S.aureus* and *E.coli*	NPs' cleavage efficiency translates to mimicking topoisomerases' activity. The supercoiled DNA generally is first transformed in relaxed (by DNA single-stranded cleavage of one phosphodiester bond) and the supercoiled and relaxed may be then transformed in linear (by DNA double-stranded cleavage of two phosphodiester bonds) of the plasmid DNA. DNA damage induced by NPs is considered as toxicity on the genetic material (genotoxicity)	[14]

3	**Silver Nanoparticles**	*E.coli* and Micrococcus	interaction of silver nanoparticles (AgNPs) withdifferent types of Deoxyribonucleic acid (DNA), mammalian and bacterial, having different base pair compositions. Binding of spherical silver nanoparticles (AgNPs) to Calf thymus (CT) DNA	[15]
4	**Silver Nanoparticles**	*Pseudomonas aeruginosa* and *Staphylococcus aureus*	The release Ag+ bind to the cell wall to damage it by fragmenting the strands of peptidoglycan , releasing the amino sugars to the media. Consequently, AgNPs accumulate and bind to the inner layers and change the peptide part and glycan strands to form large pits, leading to the destruction of the cell wall and inhibition of respiratory chain dehydrogenases and cellular growth	[16]

5	**Silver Nanoparticles**	*E.coli*	the antibacterial activity of silver nanoparticles by a mechanism interfering with DNA replication.The genetic information was mostly stored in the form of DNA. Ag NPs turn DNA into a condensed form that loses replication ability. Silver nanoparticles were able to induce gene mutations.	[17]
6	**Silver Nanoparticles**	*E. coli*	Ag ions are capable of stopping the DNA replication function and/or making proteins become inactivated, after the treatment with Ag nanoparticles, *E. coli* cells were damaged and a so-called formation of "pits" in the cell wall of the bacteria was observed. Ag nanoparticle accumulation on to the bacterial membrane leads to a significant increase in permeability and thereby is the cause of cell death , although free-radical generation caused by the free Ag(I) ions is the widely accepted explanation for understanding the mechanism of growth-inhibition by Ag nanoparticles	[18]

7	**Titanium Dioxide Nanoparticles**	*Streptococcus mutans* and *Porphyromonas gingivalis*	Mechanism of action of TiO2 NPs against the bacteria could be ROS generation, DNA damage after inter-nalization, peroxidation of membrane phospholipids, and inhibition of respiration	[19]
8	**zinc oxide Nanoparticles**	*Deinococcus radiodurans*	Zinc concentration beyond critical limits can severely inhibit the activity of functional enzymes of bacteria such as NADH dehydrogenase, glutathione reductase, and peroxidase. These events lead to the oxidative stress in bacterial cells	[20]
9	**Chitosan Nanoparticles**	*E.coli*	The ability of different molecular weights of chitosan to form Nanoparticles with a plasmid, and particulated polymers to stabilize a plasmid in a supercoiled form, transformation of the plasmids with incubated nanoparticles	[21]

10	**Magnesium Oxide Nanoparticles**	*Ralstonia solanacearum*	MgONPs possessed significant concentration-dependent antibacterial activity, and the minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) were measured as 200 and 250 μg/mL, respectively. Reactive oxygen species (ROS) accumulation could also be an important reason for the antibacterial action, inducing DNA damage.	[22]
11	**Porous silicon Nanoparticles**	*E.coli* and *S.s aureus*	loss of bacterial binding power and weakened cell membrane integrity. PSNPs binding to the free radicals inside the microbes, and to observe the nuclear breakdown.	[23]
12	**Platinum Nanoparticles**	*Staphylococcus aureus*	The effect of the platinum complexes is most probably based on their covalent binding with DNA bases. forming intra- and interstrand crosslinks, DNA —protein crosslinks, and monoadducts with DNA. DNA secondary structures can block transcription and replication with subsequent apoptosis	[24]

13	**cerium oxide Nanoparticles**	*Pseudokirchneriella subcapitata*	The oxidative activity of CeO2 particles is mediated by hydroxyl radical production and initiation of lipid peroxidation.	[25]
14	**Aluminum oxide Nanoparticles**	*Pseudomonas putida*	This study showed a decrease in the genetic stability of DNA (GTS, %) after treatment of the bacteria nano-Al2O3. The results showed that the nano-Al2O3 can induce modifications of the genetic material to a greater extent than the same compounds in the macro form.	[26]
15	**Iron Nanoparticles**	*E.coli* and *Pseudomonas aeruginosa*) and *Staphylococcus aureus* and *Enterococcus faecalis*	Oxidative stress generated by ROS is the leading player in the antibacterial activity of these NPs. These ROS can damage bacterial proteins and DNA. They can also bind and penetrate the bacterial cell wall, causing structural changes in the cell membrane, such as cell membrane permeability and cell death	[27]

16	**Fullerene Nanoparticles (CNP)**	*E. coli*	Cytotoxic properties of C60 were a result of its ability to generate reactive oxygen species (ROS) in the presence of oxygen and light. The presence of elevated levels of ROS in cellular environments causes severe oxidative stress via damage to essential cellular structures such as DNA, proteins and lipids	[28]
17	**graphene quantum dots (CNP)**	methicillin-resistant *Staphylococcus aureus* and *Escherichia coli*	generate reactive oxygen species when photoexcited (470 nm, 1 W), and kill the two strains of pathogenic bacteria	[29]
18	**Nickel hydroxide Nanoparticles**	*Escherichia coli P.aerugenosa* and *S. aureus*	Denaturation of protein and also believed to have caused damage to the bacterial cell by interacting with phosphorous and sulphur containing compounds such as DNA causing death of bacterial cell	[30]

19	**cadmium and iron oxide Nanoparticles**	*Pseudomonas putida* KT2442	Exacerbated conjugative transfer of antibiotic resistance genes (ARGs) were involved in the enhancement of cell membrane permeability, antioxidant enzyme activities, and mRNA expression levels of the conjugation genes by the co-effect of Cd2+ and nano Fe2O3. This study confirmed that the simultaneous exposure to Cd2+and nano Fe2O3 exerted a synergetic co-effect on plasmid-mediated conjunctive transfer of ARGs	[31]
20	**palladium Nanoparticles**	*E. coli, L. pneumophila* and *P. aeruginosa* *S. aureus*, *E. hirae*, *B. cereus*)	The supercoiled plasmid DNA was converted into circular form. we concluded that Pd NPs/ Urtica acted as effective chemical nuclease for double strand DNA cleavage. This result showed that Pd NPs can be used an alternative cancer drug as a DNA target agent after following toxico- logic test systems.	[32]

21	**Gold Nanoparticles**	*Escherichia coli* and *Staphylococcus aureus*	DNA conjugated gold nanoparticles indicates that the genomic DNA could stabilize the particles against aggregation owing to negatively charged phosphate backbone (binding with genomic and plasmid DNA of microorganisms)	[33]
22	**iron oxide and gold Nanoparticles**	*Escherichia coli*	The reactive oxygen species (ROS) along with superoxide radicals (O2-), hydroxide radical (OH-) and singlet oxygen (1O2) generated by the iron oxide nanopaticle is thought to be the reason behind the inhibition . gold nanoparticle was prepared and used as a vector for plasmid DNA transport within bacterial cell	[34]

CONCLUSION

It is evident in the literature that NPs exhibit antibacterial activity. The exact mechanism through which this activity occurs is only hypothesized and needs to be studied further. Although the multiple pathways that seem to be simultaneously activated by NPs make elucidation a difficult task, they are also the reason why NP exposure is so effective. The combination of ROS production, gene regulation changes, cell wall penetration, and metabolite binding are most outcomes that happens due to interation of NPs with bacteria. Although these mechanisms would also be toxic to human cells because of the similarity of the biomolecules (lipids, proteins and DNA), potential treatments of bacterial infections could be targeted focally by using specific ligands and bacterial cell receptors. Nowday, we need more research should be done to gain a further understanding of how NPs interacted with genetics contents and causess antibacterial damage and most commen mechanisms.

References

[1] Dubchak, S., Ogar, A., Mietelski, J.W. and Turnau, K.(2010), Influence of silver and tianium nanoparticles on arbuscular mycorhiza colonization and acumulation of radiocaesium in Helianthus anus, Span. J. Agric. Res., 8(1): 103-108.

[2]Raju.D, Vishwakarma R.K, Khan B.M,Mehta U.J, and Ahmad.A, 2014, Biological synthesis of cationic gold nanoparticles and binding of plasmid DNA, Materials Letters 129: 159-161

[3]Liu W. T ; 2006, Nanoparticles and their biological and environmental applications of Bioscience, Journal of bioscience and bioengineering and Bioengineering; 102(1)1-7

[4] Dobrovolskaia M. A, Aggarwal. P, Hall J. B, Mcneil.S.E , 2008 ;Preclinical studies to understand nanoparticle interaction with the immune system and its potential effects on nanoparticle biodistribution, Molecular pharmaceutics 5 (4), 487-495

[5] Hasan, S.R., Recent J.;2015;A review on nanoparticles: their synthesis and types;Research Journal of Recent Sciences; 4(2014):1-3

[6] Mohanraj, V.J and Chen, Y. ;2006; Nanoparticles-a review ;Tropical Journal of Pharmaceutical Research.,5(1):561-573

[7]Salata, O.V; 2004; Applications of nanoparticles in biology and medicine; Journal of Nanobiotechnology;2(1):3

[8]Linlin,W. Chen,H. and Longquan,S. ;2017; The antimicrobial activity of nanoparticles: present situation and prospects for the future;Int J Nanomedicine ; 12: 1227–1249

[9]taruna,H and Parihar, M. S.;1998; reactive Oxygen Species And Oxidative DNA Damage;Indian J Physiol Pharmacol; 42(4):440-452

[10]Ferric C.F. ;October 2011; Antimicrobial Actions of Reactive Oxygen; mBio journals 2(5):141-11.

[11]Abbas, W.S.; Atwan, Z.; Abdulhussien, Z.; M. A. Mahdi, M.A.(2019). Preparation of silver nanoparticles as antibacterial agents through DNA damage. Materials Technology, 1-13

[12]Slavin, Y. N.; Asnis, Ja.; Häfeli, U.; Bach, H. (2017). Metal nanoparticles: understanding the mechanisms behind antibacterial activity. Journal of Nanobiotechnology, 15(1):65

[13]Giannousi, K., Lafazanis, K., Arvanitidis, J., Pantazaki, A., Dendrinou-Samara, A.;(2014) ;Hydrothermal synthesis of copper based nanoparticles: Antimicrobial screening and interaction with DNA; Journal of Inorganic Biochemistry; 24–32

[14]Antonogloua. O, Lafazanisb. K,Mourdikoudisd.S ,Vourliasf.G,Lialiarisc.T,Pantazakib. A,Dendrinou-Samaraa.C; Biological Relevance of CuFeO2 Nanoparticles: Antibacterial and Anti-inflammatory Activity, Genotoxicity, DNA and Protein .Interactions

[15] Pramanik. S, Chatterjee. S, Arindam. S, Devi. P. S, and Kumar. G. S, 2016; Unraveling the Interaction of Silver Nanoparticles with Mammalian and Bacterial DNA, The Journal of Physical Chemistry B;120 (24):5313-5324

[16] Abbas. W, Atwan. Z, Abdulhussein. Z, and Mahdi.M; 2019, "Preparation of silver nanoparticles as antibacterial agents through DNA damage" , Materials Technology 34 (14), 867-879.

[17] Yang.W, Shen. C, Ji. Q, An. H, Wang. J, Liu. Q, and Zhang. Z;2009; Food storage material silver nanoparticles interfere with DNA replication fidelity and bind with DNA, Nanotechnology 20 (8), 085102.

[18] Takeshima.T, Tada.Y, Sakaguchi.N, Watari.F ,and Fugetsu.B, 2015, "DNA/Ag Nanoparticles as Antibacterial Agents against Gram-Negative Bacteria" Nanomaterials 5 (1), 284-297,

[19] Tabrez, S.K. , Abdulaziz, A. Javed, M.; 2015 ;" ZnO and TiO2 nanoparticles as novel antimicrobial agents for oral hygiene: a review "J Nanopart Res ;17:276

[20] Singh, R., Cheng, S. & Singh, S. ;2020 ; Oxidative stress-mediated genotoxic effect of zinc oxide nanoparticles on *Deinococcus radiodurans; 3 Biotech* 10.

[21] Bozkir.A and Saka.O.M,2004, Chitosan–DNA Nanoparticles: Effect on DNA Integrity, Bacterial Transformation and Transfection Efficiency;Journal of Drug Targeting 12 (5), 281-288

[22] Cai L, Chen J, Liu Z, Wang H, Yang H and Ding W; (2018) ;Magnesium Oxide Nanoparticles: Effective Agricultural Antibacterial Agent Against Ralstonia solanacearum; Front. Microbiol. 9:790

[23]Majid, S.J. ,Uday, M.N., Kareem, H.J., Zainab, J.T., Buthenhia, A. and Nada, R.A. :2018; ”Porous silicon nanoparticles prepared via an improved method: a developing strategy for a successful antimicrobial agent against Escherichia coli and Staphylococcus aureus”;
International Conference on Materials Engineering and Science;454.

[24] Lukas,N., Jiri,K. ,Amitava,M. ,Dagmar,H., Branislav,R.N. ,Jaromir,G. ,Kristyna,G. ,Kristyna,S. ,Simona,D. ,Sona,K. ,Marie,N. ,Pavel,K. and Vojtech,A. ; July 12, 2017 ;Platinum nanoparticles induce damage to DNA and inhibit DNA replication; pols one

[25] Rogers NJ, Franklin NM, Apte SC, Batley GE and Angel BM, Lead JR; 2010. Physico-chemical behaviour and algal toxicity of nanoparti- culate CeO2 in freshwater. Environ Chemistry 7:50–60

[26]Monika, Z.R and Nina, D. ;12 Jan 2015; DNA changes in *Pseudomonas putida* induced by aluminum oxide nanoparticles using RAPD analysis; Taylor & Francis; 57(3):1573-1581.

[27] Leili,S.A. , Vahid, A. ,Ali, M.T. ,Ali, M.A. and Saeed,T. ; 7 (2021); Green synthesis of iron-based nanoparticles using Chlorophytum comosum leaf extract: methyl orange dye degradation and antimicrobial properties; Heliyon; 7(2):2405-8440.

[28]Anna, A. ,Joyce, C. ,Kevin, G. and Madison, L. ; April 2010; The Effect of a Fullerene Water Suspension on the Growth, Cell Viability, and Membrane Integrity of Escherichia coli B23; Journal of Experimental Microbiology and Immunology (JEMI); 14: 13-20.

[29]Biljana, Z.R., Marina, M.M., Ivana, R.D., Biljana M.T., Momir S.M. ,Milica D.B. ,Verica G.P., Miroslav D.D., Zoran M.M. and Vladimir S.T.; May 2014; Photodynamic antibacterial effect of graphene quantum dots; Biomaterials; 35(15): 4428-4435

[30]Kumar,H. , Rani, R. and Salar, R.K. ; Dec. (2011); Synthesis of Nickel Hydroxide Nanoparticles by Reverse Micelle Method and its Antimicrobial Activity ; research journal of Chemical Sciences; 1(9): 42-48.

[31] Qiang, P., Xiao-Ting, F., An-Qi, S., Ting, P. , Hu, L. , Simon,B.L., Xin- Li, A. and Jian, Q.S. ; 6 February 2021; Co-effect of cadmium and iron oxide nanoparticles on plasmid-mediated conjugative transfer of antibiotic resistance genes; Environment International; 152:160-4120

[32]Gulbagca F, Aygün A, Gülcan M, Ozdemir S and Gonca S, Şen F. ;2021 Aug; Green synthesis of palladium nanoparticles: Preparation, characterization, and investigation of antioxidant, antimicrobial, anticancer, and DNA cleavage activities. Applied Organometallic Chemistry ;35(8):6272.

[33]Mishraa.A ,Tripathy.S.K. and Yun.S, 2021 , “Fungus mediated synthesis of gold nanoparticles and their conjugation with genomic DNA isolated from Escherichia coli and Staphylococcus aureus” , Process Biochemistry, 47 (5), 701-711, 2012

[34]Chatterjee.S , Bandyopadhyay.A, and Sarkar. K; 2011;Effect of iron oxide and gold nanoparticles on bacterial growth leading towards biological application, Journal of Nanobiotechnology 9 (1), 1-7.