# Silver Nanoparticles as a Potent Antimicrobial Agent: A Review

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**Summary**

Nanotechnology is expected to open some new aspects to fight and prevent diseases using atomic scale tailoring of materials. The ability to uncover the structure and function of biosystems at the nanoscale, stimulates research leading to improvement in biology, biotechnology, medicine and healthcare. The size of nanomaterials is similar to that of most biological molecules and structures; therefore, nanomaterials can be useful for both in vivo and in vitro biomedical research and applications. The integration of nanomaterials with biology has led to the development of diagnostic devices, contrast agents, analytical tools, physical therapy applications, and drug delivery vehicles. In all the nanomaterials with antibacterial properties, metallic nanoparticles are the best. Nanoparticles increase chemical activity due to crystallographic surface structure with their large surface to volume ratio. The importance of bactericidal nanomaterials study is because of the increase in new resistant strains of bacteria against most potent antibiotics. This has promoted research in the well known activity of silver ions and silver-based compounds, including silver nanoparticles. This effect was size and dose dependent and was more pronounced against gram-negative bacteria than gram-positive organisms. Also, Biosynthesis of nanoparticles is under exploration is due to wide biomedical applications and research interest in nanotechnology. The biological approach for the synthesis of nanoparticles is considered as more eco-friendly and cost-effective as compared to the other chemical and physical approaches. This review presents a detailed view about the antimicrobial properties of silver nanoparticles. We have also investigated the futuristic view of research on this aspect which is the subject of ongoing investigation in our research group.

## Introduction

The studies of bactericidal nanomaterials are gaining importance because of increase in new resistant strains of bacteria against most potent antibiotics. Effect of activity of silver ions and silver based compounds including silver nanoparticles was the size and dose-dependent nature which was more pronounced in gram negative as compared to gram positive bacteria. Metallic nanoparticles are best nanoparticles with antibacterial properties as these increase chemical activities due to crystallographioc surface structure with their large surface to volume ratio.

**Silver nanoparticles (AgNPs)**

Silver (Ag) is a transition metal element having atomic number-47 and atomic mass-107.87. The medicinal uses of silver have been documented since 1000 B.C. Silver is a health additive in traditional Chinese and Indian Ayurvedic medicine. Its action as an antibiotic comes from the fact that it is a non-selective toxic "biocide." Silver based antimicrobial biocides are used as wood preservatives. In water usage, silver and copper based disinfectants are used in 119 hospital and hotel distribution systems to control infectious agents (for example, Legionella). Silver together with copper, is commonly used to inhibit bacterial and fungal growth in chicken farms and in post harvested cleaning of oysters. Silver used to sterilize recycled water aboard the MIR space station and on the NASA space shuttle (1). Microdyn (colloidal silver in gelatin) is sold in supermarkets to disinfect salad vegetables and drinking water. Johnson Mathey Chemicals (Nottingham, UK) developed an inorganic composite (immobilized slow-release silver product) for use as a preservative in cosmetics, toiletries, and similar retail hygine-sensitive products. In Japan, a new compound (Amenitop, silica gel microspheres containing a silver-thiosulfate complex) is mixed into plastics for lasting antibacterial protection. Silver halide is often incorporated into prescription eye glasses for reversible “photochromatic” protection, as it decreases transmitted visible light. Silver resistance is important to monitor because modern technology has developed a wide range of products that depend on silver as a key microbial component. In the late 1970s, Robert O. Becker discovered that silver ions promote bone growth and kill surrounding bacteria. Silver kills some 650 different disease organisms. Gupta et al, 1998). Silver based topical dressing has been widely used as a treatment for infections in burns, open wounds and chronic ulcers. The Silver nanoparticles and Ag+ carriers can be beneficial in delayed diabetic wound healing as diabetic wounds are affected by many secondary infections. These nanoparticles can help the diabetic patients in early wound healing with minimal scars. Silver nitrate is still a common antimicrobial used in the treatment of chronic wounds (2).

**Biological approach for synthesis of silver nanoparticles (Green synthesis)**

Biological approach for the synthesis of silver nanoparticles (Green synthesis) is an eco-friendly and cost-effective method as compared to the other chemical and physical methods.

This has promoted research in the well known activity of silver ions and silver-based compounds, including silver nanoparticles. This effect was size and dose dependent and was more pronounced against gram-negative bacteria than gram-positive organisms.

Lok and his co-workers synthesized spherical nanosilver of diameter 9.3 nm using borohydride reduction method. Proteome approaches (2D and MS identification) were conducted parallel to analysis involving solutions of silver ions to investigate antibacterial action against E. coli. Data revealed by proteomic approaches is that short exposure of *E. coli* cells to antibacterial concentration of nano silver resulted in the accumulation of envelope protein precursor, indicative of dissipation of proton motive force. Nano silver were also found to show destabilization of outer membrane, collapse of plasma membrane potential and depletion of levels of intracellular ATP. Nano silver appears to be an efficient physicochemical system conferring antimicrobial activities (3).

In a study, silver nanoparticles which are biologically synthesized by *Fusarium oxysporum* were found to possess antibacterial properties. These nanoparticles were incorporated in materials and cloth, making them sterile and can be used in hospitals where often wounds are contaminated by micro-organisms. Marcato and his co-workers observed antibacterial effects when silver nanoparticles were incorporated in the cotton cloth. However, in silk cloth antibacterial effects was not observed due to less incorporation of silver nanoparticles because of less pore size (2).

The antimicrobial effect of biologically synthesized silver nanoparticles from *Fusarium oxysporum* was observed when incorporated in cotton fabrics against *S. aureus* (4).

In an another study, low concentration of silver nanoparticles was found to exhibit microbicidal effect on yeast and *E. coli*, However, on *Staphylococcus aureus* (gram positive bacteria) the antibacterial effect of silver nanopartciles was mild (5).
In another study, the bactericidal action of silver nanoparticles along with amoxicillin on E. coli was studied. Silver nanoparticles (0-40 ug/ml) and amoxicillin (0-0.525 mg/ml) showed high antimicrobial effect in Luria Bertani medium. *E. coli* showed different bactericidal sensitivity to the silver nanoparticles. As compared to the individual treatment, when amoxicillin and silver nanoparticles were combined, greater bactericidal activity of silver nanoparticles has been observed. Delay in synergistic effect of silver nanoparticles and amoxicillin and decrease in stationary and exponential phases were indicated in dynamic tests on bacterial growth on pre-incubating *E. coli* cells with silver nanoparticles antimicrobial effects were observed. Thus solutions with more silver nanoparticles have showed better antimicrobial effect (6).
In a very interesting study, antibacterial effects of silver nanoparticles synthesized by the sodium borohydride method was evaluated on recombinant *E. coli* bacteria expressing green florescent protein (GEP) was used as the model system. It was observed that silver nanoparticles above a certain concentration were not only bactericidal but also found to reduce sizes of the treated bacteria compared to untreated ones. However, no direct effect on DNA/protein profile was observed in electrophoretic studies (7).
Several antibiotics use silver compounds such as metallic silver, silver nitrate, silver sulfadizine for treatment of burns, wounds and several bacterial infections but has been declined remarkably (8). Sharma and his co-workers in 2009 also observed the antibacterial activity of silver nanoparticles and their modified form by the surfactant and polymers against various gram positive and negative bacteria (9). In another study, starch stabilized silver nanoparticles were deveopled from X- ray synthesis and they are found to possess antibacterial activity against *E. coli*. Their antibacterial property shown to be dependent on the X-ray doses (10). Similarly, oleic acid stabilized silver nanoparticles was obtained by the simple green chemical synthetic methods, shown to possess high antibacterial activity against gram negative *E. coli* and gram positive *Staphylococcus aureus* bacteria (11). Similarly, gamma irradiation of silver ions in aqueous solutions containing piolyvinyl pyrrolidone (PVP) resulted in synthesis of silver nanoparticles, which are found to possess antibacterial potential against *E. coli* indicating their potential application as biocidal material (12). When silver ions exchanged with the titanium phosphate film by ion exchange process, was effective in prohibiting growth of *E. coli* and was expected to be used as antibacterial coatings (13).
In a study, silver nanocrystals encapsulated in mesoporous silica nanoparticles was found to possess antibacterial activity against both gram positive and gram negative bacteria. This finding also promotes the antibacterial use of silver nanoparticles (14). Similarly, like antibacterial activity, antifungal activity of silver nanoparticles prepared by modified Tollens process was evaluated for pathogenic *Candida* sp. By means of determination of MIC (minimum inhibitory concentration), MFC (minimum fungicidal concentration) and time dependency of yeast growth inhibition. It was observed that at concentration as low as 0.21 mg/l of silver, silver nanoparticles exhibited inhibitory effects of silver nanoparticles against tested pathogen. It was also interesting to note here that silver nanoparticles were able to exert inhibitory effect at a concentration that is below their cytotoxic limits. So they were regarded as safe to be used as antimicrobials (15). Antimicrobial activity of silver nanoparticles consisting of silver nitrate and titanium dioxide was evaluated on gram negative and gram positive bacteria. It was observed that 1/128 and 1/512 were the minimum inhibitory concentrations of nanoparticles against *E. coli* and *Staphylococcus*

*aureus* respectively. Similarly, surgical masks coated with silver nanoparticles confirmed protection by reducing the risk of transmission of infection agents (16). Silver nanoparticles produced by chemical reduction from aqueous solution of silver nitrate containing a mixture of hydrazine hydrate and sodium citrate as reductants and sodium dodecyl sulfate as stabilizer was found to possess antibacterial activity against *E. coli*, *Staphylococcus aureus* and *Pseudomonas aeruginosa* by Kirby-Bauer method (17). However in another study, the effect of silver nanoparticles was found to be more on *S. aureus* and *E. coli* but less on the *B. subtilis* (18). Silver nanoparticles were also synthesized from the water soluble organics of *Curcuma longa* tuber and extract. The microbial bactericidal concentration (MBC) for E. coli BL-21 strain was found to be 50 mg/lt (19). A fungal strain KSU-09 isolated from the roots of date palm (Phoenix dactylifera) was identified as *Amylomyces rouxii* was found to synthesize silver nanoparticles and possess antimicrobial activity against *Shigella dysenteriae* type 1, *S. aureus*, *Citrobacter* sp., *E. coli*, *P. aeruginosa*, *B. subtilis*, *C. albicans*, *Fusarium oxysporum* (20). Similarly, silver nanoparticles synthesized from Mentha piperita leaves extract was found to possess antimicrobial activity against clinically isolated human pathogens (21). Again, biologically synthesized silver nanoparticles from *Acalypha indica* leaf extracts were found to possess antimicrobial activity against *Vibrio cholera*, *E. coli* (water borne pathogens). MIC was 10 ug/ml for both of the pathogens (22).

In addition some studies observed that biologically synthesized silver nanoparticles exhibited a potentional antibiofilm formation activity that was tested in vitro on bio-films formed by *P. aeruginosa* and *S. epidermidis* during 24-hrs treatment. Treating these organisms with silver nanoparticles resulted in more than 95% inhibition in biofilm formation (23). Antifungal activity of silver nanoparticles prepared by modified tollen process was evaluated for the pathogenic Candida sp. By MIC, MFC etc. Silver nanoparticles exhibited MIC of 0.21 mg/lt which again get lowered to 0.05 mg/lt of AgNPs (24). The antimicrobial effect of colloidal silver nanoparticles loaded over the cotton fabrics was investigated against *E. Coli*, *S. aureus* and *Candida albicans* (25). The antimicrobial effects of silver nanoparticles were compared with chemical disinfectants like phenol and sodium hypochlorite. The MBC of silver nanoparticles was found to be 0.6 ppm within 6 hours which were much better than corresponding chemical disinfectants (26). Further study were conducted based on microtitre assay suggested that at about 0.5 mg/lt silver, the inhibition of growth of *E. coli* by silver nanoparticles, silver ions and silver chloride were 55.8%, 100% and 66.66% respectively (27). Antibacterial activity of silver nanoparticles synthesized from psychrophillic bacteria were analyzed against *Arthrobacter kergulensis*, *A. gangotriensis* and *B. indicus*. The lowest concentration of silver nanoparticles is 2 ug/ml at which they arrest the bacterial growth as indicated by agar diffusion method (28). In another study, silver nanoparticles capped with sodium alginate were deposited layer by layer on surgical sultures and characterized for their antimicrobial activity against S. aureus. The concentration of sodium alginate varies from 5mM to 0.1mM and was found to have effect on the final antimicrobial activity of the fiber. It was observed that lowest alginate concentration produced the highest antimicrobial efficiency (29). Mirzajani et al, also investigated the antibacterial activity of silver nanoparticles on *S. aureus* PTCC1431 and suggested that concentration of silver nanoparticles above 8ug/ml resulted in release of muramic acid (MA) into the medium which causes cell wall distraction (30).

**Mechanism of antibacterial effect of silver nanoparticles**

Silver nanoparticles exerts their antibacterial effects by anchoring to and penetrating the bacterial cell wall and modulating cellular signaling by dephosphorylating putative key peptide substrates on trypsin residues is main mechanism by which silver nanoparticles exhibit antibacterial properties**.** In gram negative bacterial, silver nanoparticles act in three ways.

1. Silver nanoparticles attach to the cell membrane surface and distrupts its function.
2. They are able to penetrate inside bacteria so there they tend to bind to sulphur and phosphorous containing compounds like DNA and damage them.
3. They release silver ions with additional contribution to the bactericidal effects.

Bacterial cell lysis could be one of reason for observed antibacterial property. Nanoparticles modulated phosphotyrosine profile of bacterial peptide that in turn affects signal transduction and inhibited growth of micro-organisms. Antibacterial effect is dose-dependent and is independent of acquisition of resistance by bacteria against antibiotics.

Sondi and his co-workers demonstrated that E. coli cells treated with silver nanoparticles found to be accumulated in the bacterial membrane which results in the increase in permeability and resulted in dealth of cell. They suggested the silver nanoparticles are the most suitable bactericidal agent (31, 32). DVM and his co-workers used electron spin resonance spectroscopy to investigate the effect of silver nanoparticles to microbes and found that they exert their effect by generation of free radicals (5).

## Conclusion

The emergence and spread of antibiotic resistance pathogen is an alarming concern in clinical practice. Many organisms such as MRSA, HIV-1, Hepatitis-B Virus, and Ampicillin resistant E.coli are difficult to treat. There is a need of a cheap broad-active agent that canbeusedagainstvarietyofpathogen. TheAgNPs have been found to be effective against many viruses and bacterial species. The use of noble metals at nano-sizes to treat many conditions is gaining importance. The recent development in nanotechnology has pro-vided tremendous impetus in this direction due to its capacity of modulating metals into nanosizes and various shapes, which drastically changes the chemical, physical and optical properties and their use. The efficacy of AgNPs against HIV-1 has been reported by many laboratories including ours [19,26]. It has been shown that AgNPs have got anti-HIV-1 activity and can help the host immune system against HIV-1. This has laid ground for the development of new, potent antiviral drugs capable of preventing HIV infection and controlling virus replication. Recently, it has been demonstrated thatAgNPsfunctionasbroad-spectrumvirucidal and bactericidal agents, and in addition, increase wound healing. Nonetheless, conclusive safety has not been demonstrated extensively in animal models, and therefore, additional testing of AgNPs is needed before they can be used in clinical applications.

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**References**

1. Mishra M, Kumar H and Tripathi K. 2008. Digest Journal of Nanomaterials and Biostructures. 3(2): 49.
2. Gupta A and Silver S. 1998. Nat. Biotechnol. 16: 888.
3. Lok C, Ho C, Chen R, He Q, Yu W, Sue H, Tam P, Chin H and Che C. 2006. Proteomic analysis of mode of antibacterial action of silver nanaoparticles. J. Proteome Research. 5(4): 916-924.
4. Duran N, Marcato PO, Priscyla D, DeSouza A, Gabriel IH, Oswaldo L, Esposito E. 2007. Antibacterial effect of silver nanoparticles by fungal process on textile fabrics and their effluent treatment. J. Biomedical Nanotechnology. 3(2): 203-208.
5. DVM j, Kuk E, Yu KN, Kim J, Park SJ, Lee HJ, Kim SH, Park SJ, Park YH, Hwang C, Kim Y, Lee Y, Jeong D and Cho M. 2007. Antimicrobial effects of silver nanoparticles. Nanomedicine: Nanotechnology, Biology and Medicine. 3(1): 95-101.
6. Li P, Li J, Wu C, Wu Q and Li J. 2005. Synergistic antibacterial effects of beta lactam antibiotic combined with silver nanaoparticles. Nanotechnology. 16(9): 12.
7. Gogoi SK, Gopinath P, Paul A, Ramesh A, Ghosh SS and Chattopadhyay A. 2006. Green fluorescent protein-expressing E. coli as model system for investigating antimicrobial activities of silver nanoparticles. Langmuir. 22(22): 9322-9328.
8. Rai M, Yadav A and Gade A. 2009. Silver nanoparticles as new generation of antimicrobials. Biotechnology Advances. 27(1): 76-83.
9. Sharma VK, Yngard RA and Lin YT. 2009. Silver nanoparticles: green synthesis and their antimicrobial activities. Advances in Colloid and Interface Science. 145(1-2): 83-96.
10. Kassace MZ, Akhavan A, Sheikh N and Beteshobabrud R. 2008. Gamma-rays synthesis of starch stabilized silver nanoparticles with antibacterial activities. Radiation physics and chemistry. 77(9): 1074-78.
11. Le AT, Tam LT, Tam PD, Huy PT, Huy TQ, Alieu NV, Kudrinskiy AA and Krutyakov YA. 2010. Materials Science and Engineering: 30(6): 910-916.
12. Sheikh N, Akhavan A, Kassace MZ. 2009. Synthesis of antibacterial silver nanoparticles by gamma irradiation. Low Dimensional Systems and Nanostructures. 42(2): 132-135.
13. Wang Q, Yu H, Zhong L, Liu J, Sun J and Shen J. 2006. Incorporation of silver ions in to ultrathin titanium phosphate films: in situ reduction to prepare silver ions and their antibacterial activity. Cem-Mater. 18(7): 1988-1994.
14. Liong M, France B, Bradley KA and Zink JI. 2009. Antimicrobial activity of silver nanocrystals encapsulated in mesoporous silica nanoparticles. Advance of Material. 21(17): 1684-1689.
15. Panacek A, Kolar M, Vercerova R, Prucek R, Soukupova J, Krytof V, Hamal P, Zboril R and Kvitek L. 2009. Antifungal activity of silver nanoparticles against Candida sp. Biomaterial. 30: 6333-6340.
16. Li Y, Leung P, Yao L, Song QW and Newton E. 2006. Antimicrobial effect of surgical masks coated with nanoparticles. J Hospital Infection. 62(1): 58-63.
17. Guzman M, Dille J and Godet S. 2011. Synthesis and antibacterial activity of silver nanoparticles against gram-positive and gram negative bacteria. Nanomedicine: Nanotechnology, Biology and Medicine.
18. Ruparelia JP, Chatterjee AK, Duttagupta SP and Mukherjee S. 2008. Strain specificity in antimicrobial activity of silver and copper nanoparticles. Acta Biomaterialia. 4: 707-716.

19. Muthuswamy S, Krishnamurthy S and Yeoung-Sang Y. 2010. Immobilization of silver nanosynthesized using Curcuma longa tuber powder and extract on cotton cloth for bactericidal activity. Bioresource technology. 101: 7958-7965.
20. Musarrat J, Dwivedi S, Sinh BR, Al-KhedhairyAA, Azam A, Naqvi A. 2010. Production of antimicrobial silver nanoparticles in water extracts of fungus Amylomyces rouxii strain KSU-09. Bioresource Technology. 101: 8772-8776.
21. Mubarakali D, Thajuddin N, Jegananthan K and Gunasekaran M. 2011. Plant extracts mediated synthesis of silver and gold nanoparticles and its antibacterial activity against clinically isolated pathogens. Colloids and Surfaces B: Biointerfaces. 35: 360-365.
22. Krishnaraj C, Jagan EG, Rajasekar S, Selvalumar P, Kalaichelvan PT and Mohan N. 2009. Synthesis of silver nanoparticles using Acalypha indica leaf extracts and its antibacterial activity against water borne pathogens. Colloids and Surfaces B: Biointerfaces. 76: 50-56.
23. Kalimuthu K, Selvaraj B, Pandian SRK, Venkataraman D and Sangiliyandi G. 2010. Silver nanoparticles impede the biofilmformation by P. aeruginosa and S. epidermidis. Colloids and Surfaces B: Biointerfaces. 79: 340-344.
24. Marcato O, De Souza GIH, Alves OL, Esposito E, Duran N. 2001. Antibacterial activity of silver nanaoparticles synthesized by Fusarium oxysporum strain. 2nd Mercosur congress on Chemical Engineering.
25. Ilic V, Saponjic S, Vodnik V, Potkonjak B, Janancic P, Nedelz KJ and Redetic M. 2009. The influence of silver content on antimicrobial activity and color of cotton fabrics functionalized with silver nanoparticles. Carbohydrate Polymers. 78: 564-569.
26. Chamakura K, Perez-Ballestero R, Luo Z, Bashir S and Liu J. 2011. Comparison of bactericidal activities of silver nanoparticles with common chemical disinfectant. Colloids and Surfaces B: Biointerfaces. 84: 88-96.
27. Choi O, Deng KK, Nam-Jung K, Ross L, Surampalli R and Hu Z. 2008. The inhibitory effects of silver nanoparticles, silver ions and silver chloride colloids on microbial growth. Water research. 42: 3066-74.
28. Shivaji S, Madhu S and Singh S. 2011. Extracellular synthesis of antibacterial silver nanoparticles using psychrophillic bacteria. Process Biochemistry. 46: 1800-1807.
29. Dubas ST, Wacharanad S and Potiyaraj P. 2011. Tunning the antimicrobial activity of surgical sutures coated with silver nanoparticles. Colloids and Interfaces A: Physicochem, Eng Aspects. 380: 25-28.
30. Mirzajani F, Ghassempour A, Aliahmadi A and Esmaeili M. 2011. Antibacterial effect of silver nanoparticles on *Staphylococcus aureus*. Research in Microbiology. 162: 542-549.
31. Sondi I and Sondi BS. 2004. Silver nanoparticles as antimicrobial agent: a case study on E. coli as a model for gram negative bacteria. J. Colloid and Interface Science. 275(1): 177-182.
32. Wright JB, Lam K, Hanson D, Burrell RE. 1995. Am J Inf Cont. NA 7:627.