



## MiniReview

### Research Trends in Nanotechnology with Microbes in Nepal

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#### Abstract

Nanotechnology is an emerging field of modern science. It is a multi-disciplinary approach involving engineering, medicine, biology, chemistry, and physics. This involves the researchers from different fields which results in the rapid growth of this technology and exploring its application. The biological approach of synthesis of nanoparticles (NPs) and its antimicrobial property opened the door for researchers to deal with microbes. In this article, we have reviewed the research trends in nanotechnology involving microbes in Nepal. In Nepal, synthesis of NPs and dealing with microorganisms dates back only a decade ago and many research were carried out and ongoing in this field in search of its proper application.

**Keywords:** nanotechnology, microorganisms, metal nanoparticles, antimicrobial activity

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#### 1 INTRODUCTION

Nanotechnology is the study of tiny structures called nanoparticles (NPs) measuring 0.1 to 100nm derived from the bulk material. These NPs display different properties from bulk materials such as electrical conductance chemical reactivity, magnetism, optical effects and physical strength. The development in this field started in 1959 with thought process initiated by physicist Richard Feynman in his lecture stating that “There’s plenty of room at the bottom - an invitation to enter a new field of physics” at American Physical Society. But the term “Nanotechnology” was coined by Norio Taniguchi in 1974 during the description of ‘how the dimensional accuracy has improved with elapsing time’<sup>[1]</sup>. National

nanotechnology initiative was launched for the first time in 2001 and with this, era of molecular nanotechnology started from 2011<sup>[2]</sup>. Currently, this technology is rapidly growing as an important scientific field of modern research. It is a multi-disciplinary approach involving engineering, biology, chemistry, and physics<sup>[3]</sup> and has applications in various areas like medical (health care, drug-gene delivery, cosmetics, biomedical sciences), physics (electronics, space industries, energy science, optoelectronics, catalysis, single electron transistors, light emitters, nonlinear optical devices), environmental health, chemical industries, food and feed etc.<sup>[4]</sup>.

NPs can be synthesized by various methods. But

for the human welfare, the development of clean, nontoxic and eco-friendly synthesis procedures are more emphasized and green synthesis approaches are evolved. The main objective of this review article is to present an overview of research trends in nanotechnology involving microorganisms in Nepal.

## 2 NPs

The word 'Nano' is the Greek word which means 'dwarf'. So, NPs represent very small particles measuring in nanometer ( $10^{-9}$ m). Because of their defined chemical, optical and mechanical properties, they have many useful applicable values in the present century. The metallic NPs show good antibacterial properties due to their large surface area to volume ratio. There are different types of metal NPs like copper, zinc, titanium, magnesium, gold, alginate and silver<sup>[5]</sup>. Among them, silver nanoparticle has been considered as the most research interest NPs due its specific properties such as antimicrobial applications, biosensor materials, composite fibers, cryogenic superconducting materials, cosmetic products, and electronic components<sup>[4]</sup>. Since the rapid development of resistant strains of microorganisms leaving limited sensitive antibiotics, the researchers have current interest on metal NPs as an alternative. In Nepal, research was carried on different metal NPs like zinc<sup>[6]</sup>, copper<sup>[7,8]</sup>, Iron<sup>[9,10]</sup> and silver NPs (AgNPs), but most of the researches were focused upon silver NPs<sup>[11-18]</sup>.

### 2.1 Production of NPs

NPs can be produced by various methods like physical, chemical and biological methods. Physical methods include majorly include evaporation-condensation approaches. The main advantages of physical methods are mainly contamination free solvent in the prepared thin films and the uniform distribution NPs. But disadvantages may be high consumption of energy, large space acquisition and time consumption to achieve thermal stability.

Chemical methods include electrochemical synthetic method, microwave assisted synthesis, microemulsion technique, UV-initiated photoreduction, photo induced reduction etc. The advantages of chemical methods are mainly availability of many precursor chemicals and mechanisms while major disadvantages are high cost and non-ecofriendly.

Biological method is evolved focusing upon the disadvantages of physical and chemical methods. This method is also termed as "green chemistry technology" or green synthesis<sup>[4]</sup>. Green synthesis has more advantages among three methods due to being more environmentally and economically friendly processes, avoiding the use toxic chemicals in the

synthesis protocols and problem free while scaling up for producing in large quantity<sup>[13]</sup>. In the green synthesis, either biological microorganisms like bacteria, fungi or plant extracts based have been explored. But phyto mediated or plant extract based method is more preferred due to difficulty in maintaining aseptic condition and preservation of microorganisms<sup>[19]</sup>. Some examples of green synthesis are production of silver, gold, palladium, zinc oxide, platinum, and magnetite NPs applying different plants, gold, silver, cadmium, zinc, magnetite, and iron NPs production using bacteria; silver, lead and cadmium NPs from yeasts; gold, silver and cadmium NPs from fungi and silver and gold NPs from algae<sup>[20]</sup>.

The research carried out in Nepal majorly used biological method followed by chemical methods for the NPs synthesis. Since Nepal is rich in medicinal plants, researchers used the extracts of different parts of the traditional medicinal plant as precursor during NPs synthesis. For the most studied silver NPs (AgNPs), researchers chose different plants like *Justicia adhatoda*, *Aloe vera Barbaden sis*, *Artemisia vulgaris* L., *Psidium guajava* L., *Azadirachta indica*<sup>[16]</sup>, *Choerospondias Axillaris*<sup>[11]</sup>, *Brassica oleracea* L. var. *italica* Plenck, *Capsicum annum* L. *Parthenium hysterophorus* L.<sup>[18]</sup>, *Berberis asiatica*, *Cassia fistula*<sup>[13]</sup>, *Aloe Vera Azadirachta indica*<sup>[21]</sup> etc. Besides plants, some researchers used medicinally important fungi like *Ganoderma lucidum*<sup>[15]</sup> and bacteria like *Bacillus subtilis*<sup>[22]</sup> for the AgNPs production. But for zinc<sup>[6]</sup> and copper NPs<sup>[7,8]</sup>, chemical method was preferred while Neupane et al.<sup>[9]</sup> produced Himalayan honey loaded iron oxide nanoparticle for the study.

### 2.2 Application of NPs

NPs have wide applications in different areas like biosensing, drug delivery, bioimaging, catalysis, nanomanu-facturing, lubrication, electronics, textile manufacturing, and water treatment<sup>[23]</sup>.

The metallic NPs are most promising due to their wide applications. Among them, silver nanoparticle has gained more interests due to specific properties like possessing antibacterial, antiviral, antifungal and anti-inflammatory activities as well as chemical stability and good conductivity. Because of these properties, AgNPs are widely used in medical products like topical creams, antiseptic sprays and in wound dressings and also applied in synthesis of composite fibres, cryogenic superconducting materials and cosmetic products.

In Nepal, it seems that more than 60% of the research was carried on AgNPs with one of its objective as study of antimicrobial activity. Both gram positive as well as gram negative organisms were used for antimicrobial activity study. *Staphylococcus aureus* and *Escherichia*

**Table 1. Summary of Some Research in Nanotechnology Involving Microbes in Nepal**

| Research Area                                                     | Nanoparticle Involved                                                                 | Precursor of Nanoparticle                                                                                                                                    | Antimicrobial Activity Against                                                                                                                                         | Control Antibiotics         | Ref. |
|-------------------------------------------------------------------|---------------------------------------------------------------------------------------|--------------------------------------------------------------------------------------------------------------------------------------------------------------|------------------------------------------------------------------------------------------------------------------------------------------------------------------------|-----------------------------|------|
| Biological approach and antimicrobial activity                    | AgNPs                                                                                 | <i>Justicia adhatoda</i><br><i>Aloe vera</i> Barbaden sis<br><i>Artemisia vulgaris</i> L.<br><i>Psidium guajava</i> L.<br><i>Azadirachta indica</i><br>Local | <i>Escherichia coli</i><br><i>Salmonella typhi</i>                                                                                                                     | Ampicillin                  | [16] |
| Chemical approach and antimicrobial activity                      | Zinc oxide nanoparticles                                                              | Zinc nitrate hexahydrate                                                                                                                                     | <i>Staphylococcus aureus</i> <i>Escherichia coli</i>                                                                                                                   |                             | [6]  |
| Biological approach and synergistic effect                        | AgNPs                                                                                 | <i>Choerospondias Axillaris</i>                                                                                                                              | <i>Escherichia coli</i> <i>Salmonella typhi</i>                                                                                                                        | Streptomycin                | [11] |
| Chemical approach and water purification                          | Ag-iron oxide/ fly ash                                                                | Fly ash, ferrous chloride, and silver nitrate                                                                                                                | <i>Escherichia coli</i>                                                                                                                                                |                             | [24] |
| Biological approach and effects on plant development              | Biogenic silica nanoparticles                                                         | Rice husk                                                                                                                                                    | <i>Oryza sativa</i> , <i>Amomum subulatum</i> <i>Paulownia tomentosa</i>                                                                                               |                             | [25] |
| Biological approach and antibacterial activity                    | AgNPs                                                                                 | <i>Ganoderma Lucidum</i>                                                                                                                                     | <i>Klebsiella pneumoniae</i> , <i>Escherichia coli</i> , <i>salmonella typhi</i> , <i>Bacillus subtilis</i> , <i>Bacillus cereus</i> and <i>Staphylococcus aureus</i>  | Gentamicin and Streptomycin | [15] |
| Biological approach and antibacterial activity                    | AgNPs                                                                                 | <i>Bacillus subtilis</i> T (DA-12)                                                                                                                           | <i>Escherichia Coli</i> , <i>Staphylococcus aureus</i> , <i>Klebsiella pneumoniae</i> , <i>Pseudomonas aeruginosa</i> <i>Bacillus subtilis</i>                         |                             | [22] |
| Chemical approach and antimicrobial and immunostimulatory effects | AgNPs                                                                                 | Silver nitrate solution                                                                                                                                      | <i>Staphylococcus aureus</i> , <i>Staphylococcus epidermidis</i> , <i>Streptococcus pneumoniae</i> , <i>Klebsiella pneumoniae</i> <i>Escherichia coli</i>              |                             | [26] |
| Biological approach and antimicrobial activity                    | AgNPs                                                                                 | <i>Brassica oleracea</i> L. var. <i>italica</i> Plenck, <i>Capsicum annuum</i> L. <i>Parthenium hysterophorus</i> L.                                         | <i>Staphylococcus aureus</i> <i>Escherichia coli</i>                                                                                                                   | Streptomycin                | [18] |
| Biological approach and antimicrobial activity                    | Cu <sub>2</sub> O NPs                                                                 | Copper sulphate pentahydrate salt                                                                                                                            | <i>Staphylococcus aureus</i> <i>Escherichia coli</i>                                                                                                                   | Tetracycline                | [7]  |
| Chemical approach and antimicrobial effect                        | Copper nanoparticles                                                                  | Copper sulphate pentahydrate                                                                                                                                 | <i>Alternaria spp</i><br><i>Staphylococcus aureus</i><br><i>Escherichia Coli</i>                                                                                       |                             | [8]  |
| Biological approach and antimicrobial activity                    | IO-NPs                                                                                | Himalayan honey                                                                                                                                              | <i>Staphylococcus aureus</i> <i>Escherichia coli</i>                                                                                                                   | Amikacin                    | [9]  |
| Chemical approach and antimicrobial effect                        | AgNPs                                                                                 | Activated carbon powder                                                                                                                                      | <i>Staphylococcus aureus</i><br><i>Escherichia Coli</i>                                                                                                                |                             | [14] |
| Microbial fuel cell                                               | Fe <sub>2</sub> O <sub>3</sub> / graphite and MnFe <sub>2</sub> O <sub>4</sub> / PANI |                                                                                                                                                              |                                                                                                                                                                        |                             | [10] |
| Biological approach and antimicrobial activity                    | AgNPs                                                                                 | <i>Berberis asiatica</i> <i>Cassia fistula</i>                                                                                                               | <i>Staphylococcus aureus</i> <i>Escherichia coli</i>                                                                                                                   | Streptomycin                | [13] |
| Biological approach and antimicrobial activity                    | AgNPs                                                                                 | <i>Ageratina adenophora</i> leaf extract                                                                                                                     | <i>Bacillus subtilis</i><br><i>Escherichia Coli</i>                                                                                                                    |                             | [17] |
| Biological approach and antimicrobial activity                    | AgNPs                                                                                 | <i>Aloe Vera</i><br><i>Azadirachta indica</i>                                                                                                                | <i>Staphylococcus aureus</i> , <i>Klebsiella pneumoniae</i> , <i>Pseudomonas aeruginosa</i> , <i>Escherichia coli</i>                                                  |                             | [21] |
| Biological approach and antimicrobial activity                    | AgNPs                                                                                 | Root extracts of <i>Rubus ellipticus</i> Sm. (RERE)                                                                                                          | <i>Enterococcus faecalis</i> (ATCC 29212), <i>Escherichia coli</i> (ATCC 25922), <i>Staphylococcus aureus</i> (ATCC 25923), <i>Klebsiella pneumoniae</i> (ATCC 700603) | Neomycin                    | [12] |

*coli* are the most commonly used gram positive and gram negative organism respectively. The researchers also used pathogenic organisms like *Klebsiella pneumonia*, *Pseudomonas aeruginosa* and *Salmonella typhi*. The activity was evaluated comparing the activity of plant extract with or without AgNPs and also with standard antibiotic. Though, the organisms used were common, the procedure followed modified according to objective.

Bisht et al.<sup>[11]</sup> mentioned the synergistic effect of AgNPs with streptomycin increasing the antibacterial activity in combination. Khadka et al.<sup>[13]</sup> and Gautam et al.<sup>[17]</sup> showed that AgNPs are more effective against gram positive than gram negative organisms. Subba et al.<sup>[16]</sup>, Shahi et al.<sup>[18]</sup> and Prasain<sup>[21]</sup> compared the antimicrobial activity of AgNPs derived from different plants helping to screen the appropriate plant source. Khanal et al.<sup>[12]</sup>, Aryal et al.<sup>[10]</sup>, Bajracharya et al.<sup>[22]</sup> and Poudel et al.<sup>[15]</sup> experimented upon various gram positive and gram negative organisms including pathogenic and opportunist pathogens and their result also showed the possession of antimicrobial activity by AgNPs.

Besides AgNPs, researchers also used copper, zinc and iron oxide NPs for antimicrobial activity. Hona et al.<sup>[8]</sup> determined the minimum inhibitory concentration of copper NPs against bacteria and fungi while Regmi et al.<sup>[7]</sup> concluded that antimicrobial activity of copper NPs was concentration dependent. Singh et al.<sup>[6]</sup> determined the synergistic effect of zinc NPs with various antibiotics against both gram positive and gram negative organisms. So, all these research indicated the possible application of metal NPs as antimicrobial agents and may help to provide an alternative for the resistant antibiotics.

Along with antimicrobial activity, the research by Pandey et al.<sup>[14]</sup> and Joshi et al.<sup>[24]</sup> applied metal NPs for the waste water treatment. Pandey et al.<sup>[14]</sup> synthesized silver NPs doped activated carbon (AC) composite and Joshi et al.<sup>[24]</sup> synthesized a fly ash based composite, Ag-iron oxide/fly ash. In both research, the antimicrobial property had been determined and mentioned for their efficient application in waste water treatment.

### 3 RESEARCH IN NANOTECHNOLOGY INVOLVING ORGANISMS IN NEPAL

In Nepal, Nepalese Association of Medical Microbiology and Nepal Polymer Institute had organized the “First International Conference on Infectious Diseases & Nanomedicine-2012 (ICIDN-2012)” in Kathmandu from December 15-18, 2012 in association with Kathmandu University, Kavre. The distinguished scientists and delegates from more than 20 countries around the world had attended the conference. The proceedings of this conference published a single full research article from Nepal related with NPs by Subba et al.<sup>[16]</sup>

and authors claimed that ‘biogenesis of AgNPs using plant extracts from Nepal has not been investigated so far’. The publicity of NPs developed the curiosity in researchers from multiple fields like chemistry, botany, biotechnology etc. and more research in this field had been carried out. Table 1 shows summary of some research carried out in Bachelor and Master’s program which illustrates the increase in interest of researchers in this field.

### 4 CONCLUSION

Nanotechnology is an emerging technology, growing rapidly in modern science field. In Nepal, since a decade ago, the researches were carried out focusing more on the biological approach of synthesis of NPs and its antimicrobial property. Due to immense interest of researchers in this area, more research was carried out resulting more publications. The research in this area is beginning from Bachelor level and this may help in future to expertise in this field.

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### Conflicts of Interest

The authors declared no conflict of interest.

### Author Contribution

Shakya Hada MS developed the concept and design, acquired the data and wrote the main manuscript and revised the manuscript. Shrestha A edited the manuscript and prepared plagiarism report. Manandhar P and Manandhar S helped in literature search.

### Abbreviation List

AC, Activated carbon  
AgNPs, Silver NPs  
NPs, Nanoparticles

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