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Biosynthesis of Silver Nano Particles using *Putranjiva Roxyburghii Wall* Leaves Extract and their Antibacterial Activities

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Abstract

The synthesis of nanoparticles from plant source has to be prove to be effective and alternate method for the noble production of nanoparticles .A number of synthesis technique have been developed including chemical method ,physical method and biosynthetic method. In this research synthesis of silver nanoparticles using putranjiva roxyburghii wall leaves has been investigated .we successfully synthesized uniformly dispersed silver nanoparticles with the size and shape in the range of 3 to 20nm with an average size of 6 nm .The characterization of nanoparticles was done by using UV-VIS spectrophotometer and fluorescent spectrometer.the resulting formation of silver nanoparticles are characterized by using Transmission electron microscopy(TEM) .The TEM study shows the formation of silver nanoparticles .The antibacterial activity of these nanoparticles was studied against E.coli and staphylococcus aurenus .The bacterial property was analysed by measuring the growth curve of bacteria and 50ug/ml concentration of silver nanoparticles was found to be effective antibacterial.

Keywords : *Silver nanoparticles; Biological synthesis; Putranjiva roxyburghii wall; E.coli.*

1. INTRODUCTION

The development of green processes for the synthesis of nano particles is involving in to an important branch of nanotechnology (Manish Dubey et al. 2009; Mukherjee et al., 2001; Sun et al. 2000.) Today, the use of environmentally benign like plant leaf extract, bacteria and fungi for the synthesis of silver nanoparticles offers numerous benefits ecofriendliness and compatibility of pharmaceutical and biomedical application as they do not any toxic chemicals in the synthesis protocol (Han et al. 2005). Chemical synthesis method leads to the presence of some toxic chemical species absorbed on the surface that may have adverse effect in medical application Han et al., 2005. Bioinspired synthesis of nanoparticles provides advancement over physical and chemical method as it is cost effective and ecofriendly. In this method there is no need to use high pressure ,energy ,temperature and toxic chemicals (Leela et al. 2008; Vyom et al. 2009). Here, we report the synthesis of silver nanoparticles, reducing silver ions (Ag^+) present in aqueous solution of silver nitrate by the help of putranjiva roxyburghii wall leave extract .we also study the antibacterial properties of silver nanoparticles towards *E.coli* and *Staphylococcus aurenus* .Although several previous reporters have studied the antibacterial activity of chemically synthesized silver nanoparticles (Upendra Kumar et al. 2009). But here we study the biologically (using putranjiva roxyburghii wall leave extract) synthesized silver nanoparticles.

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2. EXPERIMENTAL DETAILS

2.1 Material

For the synthesis of silver nanoparticles healthy leaves of putranjiva roxyburghii collected from ccs university ,Hissar ,Haryana. The chemical silver nitrate ($AgNo_3$) is purchased from Merck limited, India. The lyophilized culture of *E.coli* and *Staphylococcus aurenus* are procured from Biotechnology laboratory ,LPU.

2.2 Preparation of extract

Extract has been prepared by bringing fresh leaves of putranjiva roxyburghii wall from ccs university , Hissar, Haryana to the laboratory .leaves weighing 25gm washed throughly thrice in distilled water for fifteen minutes ,dried ,cut in to fine pieces and were boiled in to 500ml flask with 100 ml of sterile distilled water up to five minutes and were filtered.

2.3 Synthesis of silver nanoparticles

Silver nitrate was purchased from Mreck India company and used as same. 50 ml of fresh leave extract was added to aqueous solution of 1mM silver nitrate.The average size of the synthesized particle was 10 nm with the range of 5 to 20nm with irregular shape.Due to our interest to get much smaller particles above solution was centrifuged at the rate of 1200rpm up to fifteen minutes and investigated that particle present in the supernatant were nearly homogeneous with average particle size of 6nm.

3. CHARACTERIZATION

3.1 UV-VIS spectroscopy

Ultra violet visible spectroscopy(UV-1601 pc shimadzu spectrophotometer) or ultra violet visible spectrophotometer (UV-VIS) refers to the absorption spectroscopy in the UV-Visible spectral region. This means it uses light in the visible and adjacent (near – UV and near- infrared ranges) the absortion in the visible range directly effect the perceived color of the chemicals involved. In this region ,electromagnetic spectrum molecules undergo electronic transistion.

3.2 Transmission electron microscopy

Transmission electron microscopy (TEM) (Philips CEM -10) is a microscopy technique where by a beam of electron is transmitted through an ultrathin specimen, interacting with the specimen as it passes through .An image is formed through the interaction of electron transmitted through the specimen; the image is magnified and focused to an imaging device.

3.3 Analysis of interaction of silver nanoparticles with bacteria

3.3.1 Bacterial growth curve

The antibacterial activity of silver nanoparticles against *E.coli* and *staphylococcus aurenus* was analysed by their growth curve. We have inoculated fresh colonies of agar medium in to 10 ml of broth (luriabertani) media. The media is supplemented with 10 to 50*u*g/ml silver nanoparticles and bacterial culture was incubated at 37°c with continous shaking at 150 rpm .The growth of *E.coli* and *staphylococcus aurenus* in the broth media was indexed by measuring the optical density at lambda=600 nm at regular intervals using UV-VIS spectrophotometer whereas control does not contain any exposure of silver nanoparticles synthesized from putranjiva roxyburghii wall leave extract.

4. RESULT AND DISCUSSION

4.1. UV –VIS Technique

Figure 1 represents the UV-VIS spectra of aqueous component as a fraction of time variation of leaf broth with 1mM aqueous silver nitrate solution .Metal nanoparticles have free electrons,

which gaves surface Plasmon resonance (SPR) absorption band, due to the combine variation of metal nanoparticles in resonance with light wave. Sharp bands of silver colloids were absorbed at 438nm and change in color were observed from colorless to yellow shown in fig 2. These characterization variations in color due the excitation of surface Plasmon resonance in metal nanoparticles.

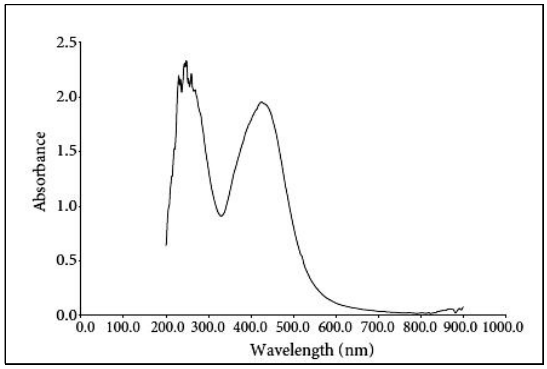

Fig 1: UV –VIS spectra of silver nanoparticles

Fig 2: Picture of aqueous solution of 1mM Silver nitrate with *putranjiva roxyburghii* wall leaf extract before adding the leaf extract and after addition of leaf broth.

4.2 Transmission electron microscopy

Transmission electron microscopy image of silver nanoparticles derived from putranjiva roxyburghii wall leave extract was shown in fig 3.The morphology of the nanoparticles was spherical in nature .The obtained nanoparticles in the range of sizes 3nm to 20nm .The average mean size of silver nanoparticles was 6nm .

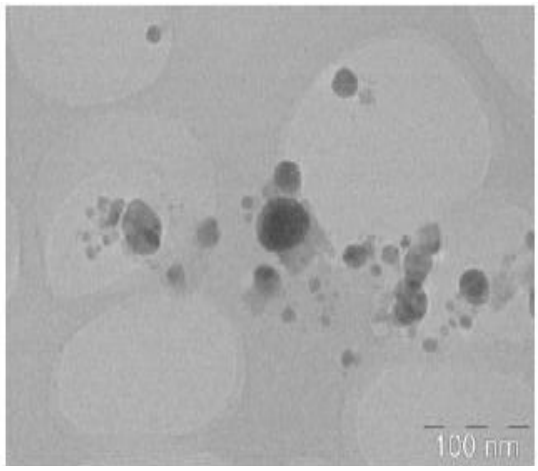

Fig 3: TEM image of silver nanoparticles from *putranjiva roxyburghii* wall leaf

4.3 Analysis of growth curve

It was well that silver nanoparticles exhibits strong antibacterial activity due to their well developed surface which provide maximum contact with the environment .Here ,antibacterial effect of silver nanoparticles were studied by using optical intensity as function of time for 25 hours with varying concentration of silver nanoparticles .From fig 4.and 5,we can conclude in the absence of silver nanoparticles there is increase in optical density showing bacterial growth but as the concentration of silver nanoparticles increases there is reduction in the bacterial growth of *E.coli* and *staphylococcus urenus* .

5. CONCLUSION

We used putranjiva leaf extract as a reducing and caping agent to minimize the serious environmental pollution problem. silver nanoparticles were synthesized by this method

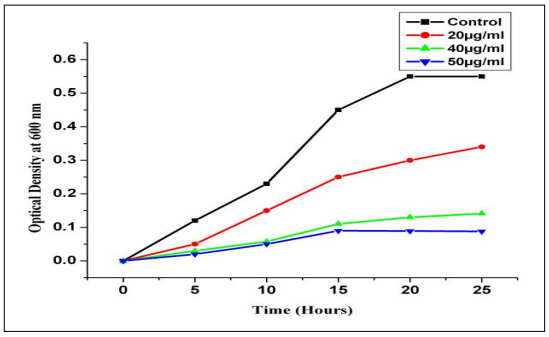

Fig.4: Effect of Silver Nanoparticles on *E.Coli* growth

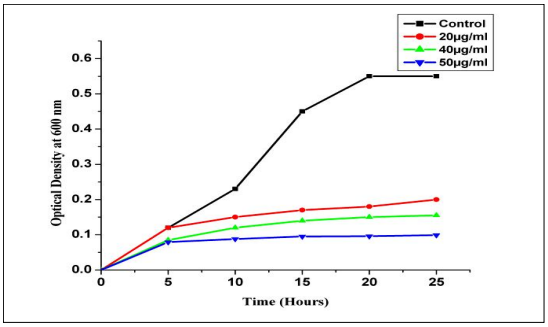

Fig.5: Effect of Silver Nanoparticles on gowth rate of *Staphylococcus Aurenus*

having 6nm average mean size .The preparation of silver nanoparticles by using putranjiva leaf extracthas desired quality with low cost and convient method. These nanoparticles at concentration 50*ug*/ ml were showed complete antibacterial activity against *E.coli* and *staphylococcus aurenus.*

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