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Green Synthesis of Silver Nanoparticles using *Brassica Oleracea* (Cauliflower) and *Brassica Oleracea Capitata* (Cabbage) and the Analysis of Antimicrobial Activity

*R. Tamileswari , M. Haniff Nisha[1] , Sr. S. Jesurani [2]
PG and Research Department of Physics
Jayaraj Annapackiam college for women(Autonomous)
Theni, India

Abstract:- The use of plants and vegetables for the synthesis of nanoparticles (AgNPs) has more advantages because it is safe to handle and easy availability. The present investigation reports the green synthesis of silver nanoparticles using the vegetable cauliflower and cabbage and analysis of its antibacterial activity. The presence of Silver nanoparticles was initially confirmed by the colour developed and then by UV-Vis Spectrometer, XRD, SEM and EDAX. The antibacterial activity was evaluated against the pathogenic strains *Klebsiella 2neumonia, Bacillus subtilis, Staphylococcus aureus and Escherichia coli* and the maximum zone of inhibition was obtained for *Bacillus subtilis.*

Keywords: Green Synthesis, brassica oleracea, silver nanoparticles, SEM , antibacterial activity.

I.INTRODUCTION

A material when in a nano sized form can assumes properties which are very different from those when the same material is in a bulk form. Nanomaterials or structures in typically ranging from subnanometers to several hundred nanometers [1]. The noble metal nanoparticles have more unique properties that are substantially diverse from those of bulk materials [2]. Among them silver nanoparticles have numerous physiognomies that make it extensively used one in science and also it have been demonstrated as an effective biocide against an eclectic range of bacteria including both gram-positive and gram negative. There are several methods to synthesis a silver nanoparticles [3]. The chemical methods expended for the synthesis of nanoparticles are too expensive and also involve the use of toxic, hazardous chemicals that are responsible for various biological risks. So the synthesis of nanoparticles using plants or vegetables and their extracts Rosa damascene and Punica granutum[4], Raphanus sativus leaves[5], Securinga leucopyrus[6], Allmania nadiflora[7], Borassus flabellifer[8] can be advantageous over other synthesis. This is the most adopted method of green, eco-friendly production of nanoparticles and also has a special advantage that the plants are widely distributed, emphatically obtainable, much secure to handle and act as a source of several metabolites. The vegetable itself act as a reducing and capping agent in the synthesis process.

The vegetable cauliflower *(Brassica oleracea) as* sample A and cabbage (*Brassica oleracea capitata)* [9] as sample *B* be used to synthesis the silver nanoparticles. These are the vegetables of the Brassicaceae family. It is originally from Europe and Asia. These vegetables has potent biological and immunological activities. It also has various chemical constituents. They are very good source of electrolytes, minerals, vitamins and dietary fibers, isothiocyanate anti-oxidant compound called sulforaphane [10] , phytochemicals like indoles which are detoxifying agents and zea-xanthin, lutein which are flavonoid antioxidants . Eventhough AgNps have been synthesized from various biological sources, the synthesis of AgNP from cauliflower and cabbage is not much reported in literature. So this investigation is designed to synthesis the AgNPs from cauliflower and cabbage and to evaluate the antibacterial activity.

MATERIAL AND METHODS

To obtain silver nanoparticle the deionized water was used as a reaction medium, the reducing agent used was vegetable extract, i.e sample A and B extract and the reagent used for the synthesis was silver nitrate which does not evolve any toxic hazard on the atmosphere.

Fig.1: The vegetables

Cauliflower

Cabbage

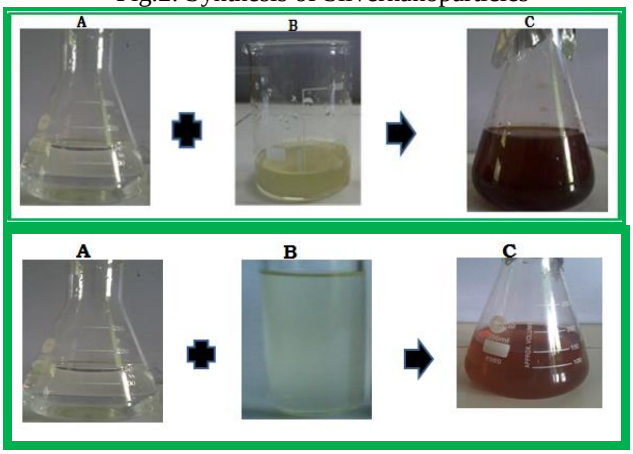
Preparation of Fresh Radish Extract

Vegetables were procured from the local market (Nov, 2014) . Aqueous extract of cauliflower and cabbage was prepared using 25 gm. It was washed thoroughly in deionised water, dried, cut into small pieces and were boiled in 150 ml of deionized water for 5-10 minutes. The extract was filtered through Whatmann No.1 filter paper and used for further research.

Synthesis of silver nanoparticle

1mM aqueous solution of Silver nitrate (AgNO3) was prepared and used for the synthesis of silver nanoparticles. 10 ml of the vegetable extract was added into 90 ml of aqueous solution of 1 mM Silver nitrate and incubated 15 minutes at room temperature. After 15 minutes, the colour of the solution changed from colourless to dark brown and honey brown respectively indicating the formation of silver nanoparticles.

Fig.2: Synthesis of Silvernanoparticles



A - Silver nitrate solution;
B - Vegetable extracts;
C - Colour change after adding A&B respectively cauliflower and cabbage

Characterization of Silver Nanoparticles

The extract was centrifuged twice to isolate the AgNPs and to eliminate the unwanted surplus. It was dried in Hot air oven for 30 minutes at temperature 100°C. Synthesized silver nanoparticles were confirmed by UV–Vis spectroscopy and it was carried out using UV-Vis spectrophotometer in 200–1100nm range. Detailed analysis of the morphology, size and distribution of the nanoparticles was documented by Scanning Electron Microscopy (SEM) and the presence of elemental silver signal was confirmed in the sample by using EDAX.

Assessment Of Antibacterial Activity

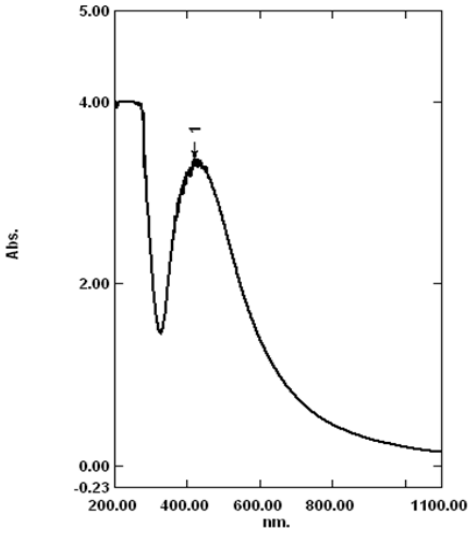
Antibacterial activity of synthesized silver nanoparticles against Gram negative (*Escherichia coli* and *Klebsiella pneumoniae*) and Gram positive (*Bacillus subtilis*, *Staphylococcus aureus*) bacteria was evaluated using disc diffusion method and zone of inhibition was measured.

RESULTS AND DISCUSSION

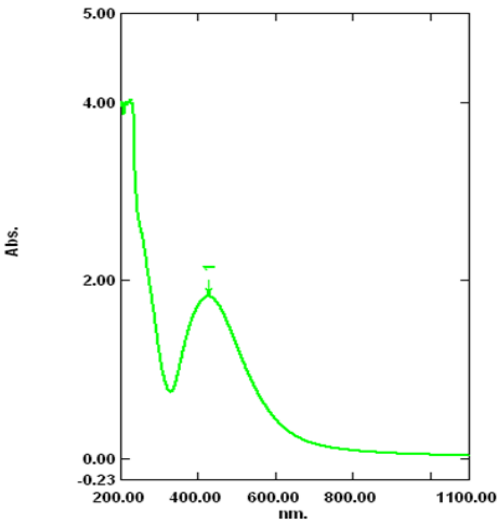
UV-Vis Spectroscopy

The confirmation of formation and stability of silver nanoparticles was studied by using UV-Visible spectrum. The uv-vis spectrum was recorded using aqueous solution of Ag nanoparticles. The silver nanoparticle showed the sharp peak around 420 nm for sample A and 428nm for sample B (Fig.3) with high absorbance which is very specific of silver nanoparticles.

Fig.3: UV-Vis Spectra of the sample



Sample A : 420nm



Sample B : 428 nm

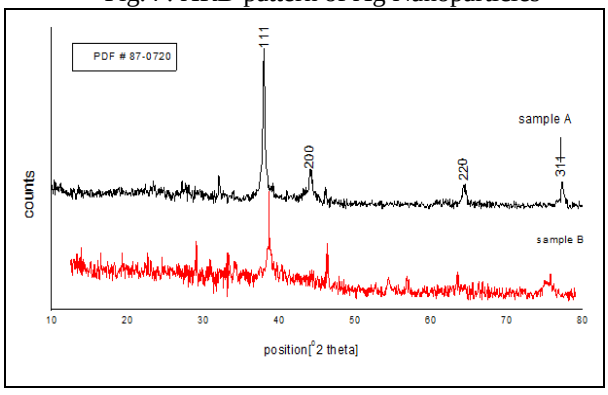
XRD analysis of silver nanoparticles

The synthesized Ag nanoparticles using Radish extracts was further confirmed by the characteristic peaks observed in XRD pattern. The fig.4 showed the XRD pattern for different intensity peaks in the whole spectrum of 2θ values ranging from 10 to 80. The silver nanoparticles produced in our experiments were in the form of nanoparticles as evidenced by the peaks at 2 theta values of 37.98, 46.072, 64.38, 76.61 which was matched with the JCPDF # 87-0720 (which confirms the silver). And we confirm that this pattern shows the cubic system and face centered lattice parameter. Average particle size of the silver nanoparticles formed in this process was determined using Scherer's formula,

$$d = 0.9\times\lambda/\beta * \cos\theta$$

For this the calculated particle size was 36nm and 42nm for sample A and B respectively. XRD pattern thus clearly illustrate that the silver nanoparticles formed .

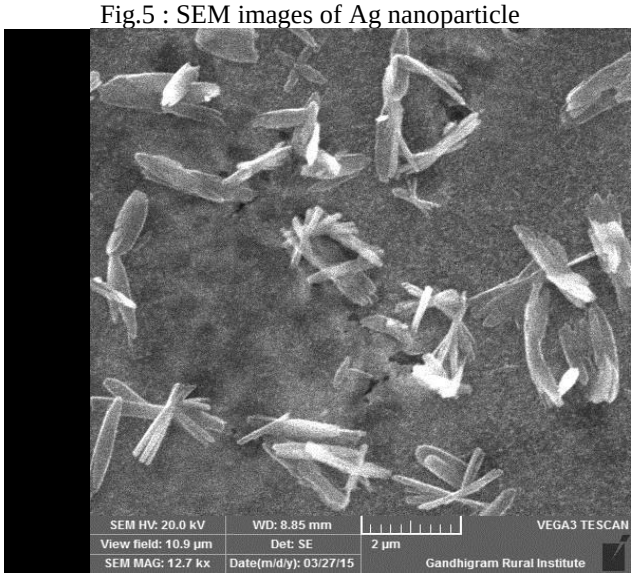
Fig.4 : XRD pattern of Ag Nanoparticles



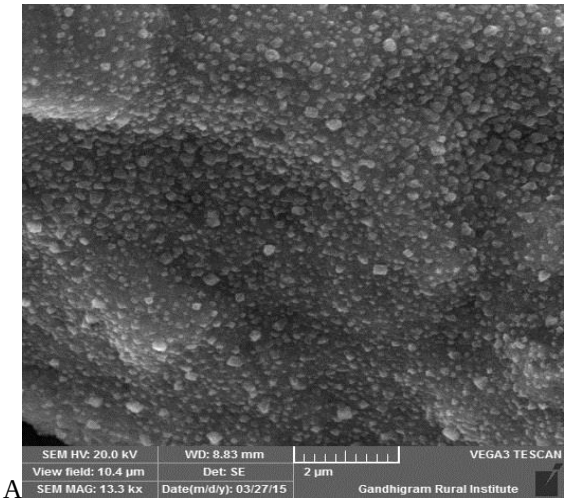
Scanning Electron Microscopy

SEM was used to view the morphologhy and size of silver nanoparticle. SEM image showed the high density nanoparticle synthesized by fresh cauliflower and cabbage extract were respectively flakes and spherical in shape. This confirmed the development of silver nanostructures. The fig.5 showed the nanoparticle in the range of 30 -50 nm (for both). This size of particle confirms that the presence of nanoparticle.

Fig.5 : SEM images of Ag nanoparticle



SEM image of sample



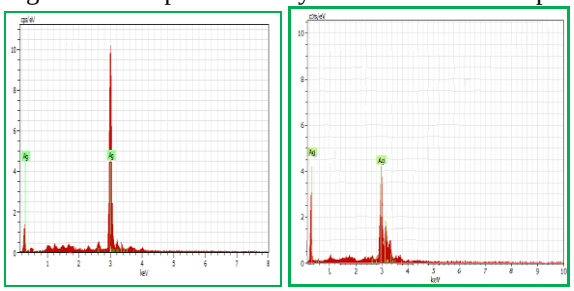
A

SEM image of sample B

Energy Dispersive X-Ray Spectroscopy (EDAX) measurement

The EDAX analysis obtained in the present study also confirmed the presence of silver nanoparticles synthesized from Cauliflower and Cabbage extract. Metallic silver nanoparticles generally show typical optical absorption peak approximately at 3 keV due to surface Plasmon resonance. The fig.6 clearly showed the presence of silver nanoparticles.

Fig.6: EDAX spectrum of synthesized silver nanoparticles



EDAX spectrum of sample Aand B

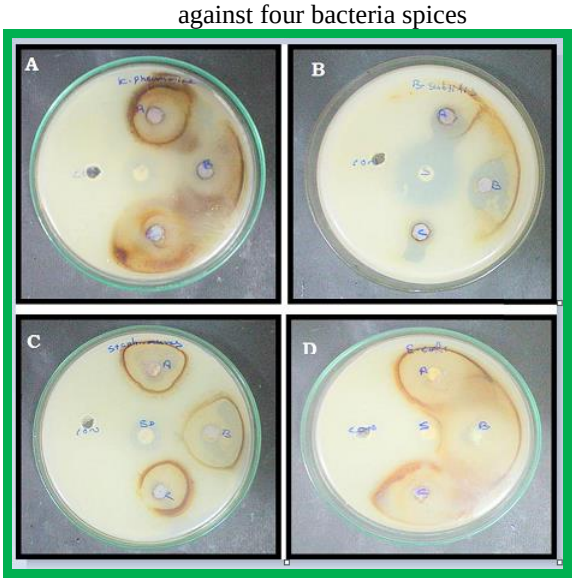
Antibacterial Activity by Disc Diffusion Technique

The antibacterial activity was assessed against four bacteria species. The results indicated that silver nanoparticles synthesized from the two vegetables showed effective antibacterial activity both in Gram negative and Gram positive bacteria which is compared with amikacin. Among the four Bacillus Subtills have the maximum inhibition. Its zone of inhibition was 16mm. The four imges showed the bacterial activity of four bacteria spices A-*Klebsiella pneumoniae*, B-*Bacillus subtilis*, C-*Staphylococcus aureus and* D-*Escherichia coli*. In each image A and B indicated the activity by our sample A and sample B. Ampicilin was taken as the positive control for the measurement of zone of inhibition (in mm).

Table 1: The antibacterial activity of silver nanoparticles synthesized using cauliflower and cabbage

Sample Code	Klepshiella pneumonia	Bacillus Subtilis	Staph. aureus	E.coli
A	R	16	14	10
B	R	16	R	10
CONTROL	R	R	R	R
STANDARD (AMIKACIN)	16MM	20	16	16

Fig.7: Bacterial activities of synthesized Ag nanoparticles against four bacteria spices



CONCLUSION

Silver nanoparticles were successfully synthesized from silver nitrate solution using cauliflower and cabbage extract was characterized using UV-Vis spectroscopy, X-ray diffraction, Scanning Electron Microscope and EDAX. And these silver nanoparticles showed good antibacterial activity against *Klebsiella pneumoniae*, *Bacillus subtilis*, *Staphylococcus aureus and Escherichia coli*. In the present study we found that cauliflower and cabbage can also be a good source for synthesis of silver nanoparticles and also it is environmental friendly and free from organic solvents and toxic chemicals. Further research will include the anti-inflammatory effect and anti-cancerous activity by silver nanoparticles synthesized from these vegetables.

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