Biological synthesis and characterization of tri-metallic alloy (Au Ag, Sr) nanoparticles and its sensing studies

Arvind Binod¹, Sharanabasava V. Ganachari¹, *, Jayachandra S. Yaradoddi¹,², Rakesh P. Tapaskar⁶, Nagaraj R. Banapurmath¹, Ashok S. Shettar⁷

¹ Research Scientist and Assistant Professor, Centre for Material Science, Advanced research in Nanoscience & Nanotechnology, KLE Technological University, Vidyanagar, Hubballi-580-031, INDIA
² Research Scientist, Centre for Material Science, Advanced research in Nanoscience & Nanotechnology, KLE Technological University, Vidyanagar, Hubballi-580-031, INDIA
³ Professor & Head, Centre for Material Science, Advanced research in Nanoscience & Nanotechnology, KLE Technological University, Vidyanagar, Hubballi-580-031, INDIA
⁴ Undergraduate student, School of Mechanical Engineering, KLE Technological University, Vidyanagar, Hubballi-580-031, INDIA
⁵ Vice Chancellor, KLE Technological University, Vidyanagar, Hubballi-580-031, INDIA
⁶ Assistant Professor, Energy Cluster, Centre for research in Renewable & Energy Systems, KLE Technological University, Vidyanagar, Hubballi-580-031, INDIA
⁷ Director, Extremz Biosciences Pvt. Ltd., KLE Technological University, Vidyanagar, Hubballi-580-031, INDIA

* Corresponding Author: sharanu14@gmail.com

Abstract: Crystallized Gold (Au), Silver (Ag), Strontium (Sr) tri-metallic nanometal alloy have been prepared and stabilised using plant extracts of coriander root (Coriandrum sativum) in an aqueous system. Aqueous solutions of Au⁺, Ag⁺, Sr⁺ ions are in 1:1:1 ratio of Au–Ag–Sr alloy was treated with a filtered solution of Coriandrum sativum root extract for the formation of Au–Ag–Sr trimetallic alloy nano particles (Au-Ag-Sr NP). Analysis of the feasibility of the biologically synthesized bio-functional core–shell nanometal alloy from plant root extract is particularly noteworthy. The colloidal suspensions obtained were in highly stable condition for 6-8 weeks. The prepared Au-Ag-Sr nanometallic alloy was then checked for the surface Plasmon resonance. The composition, morphology, size and structure of the nanoparticles were determined by Fourier transform infrared spectroscopy (FTIR) and field emission scanning electron microscopy (FESEM). Possible sensing applications of these tri-metallic alloy (Au Ag, Sr) nanoparticles are envisaged.

Keywords: Coriandrum sativum root extracts, green nanotechnology, tri-metallic alloy nanoparticles, Fourier transform infrared spectroscopy, x-ray diffraction, scanning electron microscopy.
1. Introduction
C Nano metals or metal nano particles are particles of the size 1-100 nm (that is 10-9m). Nowadays nanotechnology is widely used in various fields including surface chemistry, organic chemistry, molecular engineering, semiconductor and nano fabrication. Due to the usage of nanotechnology in industries and military applications various governments are investing a lot on nanotech research. Nanometals are used in variety of applications because of their small size and properties depending on their shape. The synthesis of these metal nano particles are either by physical or by chemical processes which are pretty expensive techniques also it might be potentially hazardous to the environment and living organisms in the environment. Thus, in order to make a cost effective and safe synthesis technique, biological method was brought into use.

Various applications of the metal nano particles that are mentioned in various articles are anti-microbial, anti-cancer, medicinal applications, cosmetics, coating, bio catalysis, DNA labeling, drug delivery and much more. Some metal nano particles that were synthesized were gold (Aunp), silver (Agnp), uranium (Unp), platinum (Ptnp), zinc (Znp) and many more. The biological synthesis method is basically of two types bottom up approach and top down approach. In bottom up approach the bulk nano particles are built or made from the smaller entities bringing them together whereas in top down approach it is reversing the smaller nano particles are made by breakdown of bulky nano material. When it comes to nano scale the bottom up approach of synthesis is preferred over top down as they also induce internal stresses in addition to defects and contaminations. The biological method of synthesis is a simple and environmentally safer approach where the salts of the metals are mixed with plant extracts or microbial extracts, whole plant or microbes like fungi, yeast and more useful microbes to get metallic ions in nanometer size of 1 to 100nm. For visualizing the physical properties or the chemical composition of the nano particles synthesized various characterization techniques are used like UV vis. spectroscopy, SEM, TEM, FTIR, powder XRD and EDS.

A review article on plant extract mediated synthesis of silver nanoparticles for antimicrobial applications [1]. This synthesis involved reducing and stabilizing of silver ions by various biomolecules like alkalis and proteins which we obtain from plant extracts. The nanoparticles synthesized were spherical in shape and of size 50-100 nm one of the plant extract was of Alternanthera dentate. XRD and TEM image revealed silver nanoparticles of size 25nm and having spherical shape and face centered cubic structure. These nanoparticles so formed were tested for antibacterial activity against bacterial pathogens named Pseudomonas flourescens and Aeromonas hydrophilia.

An article telling us about Synthesis and characterization of Copper nanoparticles using the leaf extract of Capparis zeylanica [3] which acts as both reducing agent and also as a capping agent. The copper nanoparticles so synthesized acts as antimicrobial element since copper is highly toxic to microbes such as E coli, Pseudomonas aeruginosa it is not considered toxic for human beings so it is used for food packaging applications and also for water treatment. The plant extract of the Capparis zeylanica mainly consists of fatty acids, alkaloids and flavonoids XRD showed simple cubic copper nanoparticles SEM showed spherical uniform copper nanoparticles of size 60 – 100 nm. The antimicrobial study was done against gram positive (Staphylococcus aureus) as well as gram negative bacteria (Pseudomonas aeruginosa) by disk diffusion method.

A paper wherein Rapid biological synthesis of platinum nanoparticles using Ocimum sanctum for water electrolysis applications [2]. This article suggests green synthesizing platinum nanoparticles from leaf extracts of Ocimum sanctum and an aqueous solution of chloroplatinic acid (H2PtCl6.6H2O). More conversion of platinum ions to platinum nanoparticles takes place when a tulsi leaf extract is used at a reaction temperature of 100oC. Platinum nanoparticles were then dried and their structure and composition were analyzed by XRD with EDAX, SEM, FTIR, and LSV. The EDAX showed platinum of about 71.56%. Also, sodium, calcium, carbon elements were also seen. Scanning electron microscopy showed images of the platinum nanoparticles are irregularly shaped ones some were rectangular and some were triangular. Size was seen as 50 nm. The XRD analysis showed the average particle size as 23 nm. FTIR showed peaks assigned as OH stretching in alcohols, alkali, amine, carboxylic acid functional groups present Application of this reduced platinum is for the electrolysis of water.
A paper suggesting Bio synthesis of nickel oxide and nickel nanoparticles and their characterization [4]. Here article, the nickel oxide and nickel nanoparticles were synthesized by boiling method using leaves of Azadirachta indica and Psidium guajava. The size and morphology of the particles was to be 17-77 nm by transmission and scanning electron microscopy. XRD showed formation of pure Ni and NiO cubic phases with a crystalline size of 44 nm. There is a chance that Ni and NiO nanoparticles can be used for cancer therapy.

A paper telling about Nano particles which are biosynthesized by Fungi and Yeast [5]. The species used for the synthesis is Fusarium oxysporium. These species generate silver nanoparticles in the range of 20 to 50 nm. Mostly the nanoparticles so formed were of quasi spherical shape. The shape and size of the nanoparticles changes with different fungal species. Verticillium species of fungi give cubo-octahedral shaped 100 to 400 nm sized magnetite nanoparticles. The yeast helps generate hexagonal silver nanoparticles of size 2 – 5 nm. The applications include anti cancerous, anti-microbial, anti-fungal, bio sensor, for medical imaging.

A review article showing Gold nano particles synthesis, its properties and applications. [6]. There are many methods to synthesize gold nanoparticles. By chemical method gold nanoparticles are synthesized by reducing hydrochloroauric acid using some stabilizing agent. Turkevich method synthesizes gold nanoparticles of size 10 to 20 nm, spherical in shape. Brust method involves reduction of hydrochloroauric acid with tetra octylammonium Bromide it would act as anti-coagulating agent and also a reducing agent the size of nanoparticles would be 6 nm. Seeded growth method synthesizes nanoparticles in variety of shapes such as rods, cubes, tubes. Also, the gold nanoparticles can be generated using plant extracts as it would be mostly uniform shape and size of nanoparticles. Some microbes used for the synthesis includes Bacillus subtilis, Fusarium oxysporium and Candida albicans [7], detecting hydrogen sulfide (H2S) using bio-functionalized gold nanoparticles [8]. This objective of this research article is about the biosynthesis of tri-metallic alloy Au-Ag-Sr formed out of its salt solutions by mixing it with plant root extract of Coriander sativum. The metals were generated from their salt solutions of AgNO3 for Ag, AuCl2 for Au and SrCl2 for Sr. The root extract preparation was done and few ml of extract was added to a 1:1:1 mixture of its 1N salt solution. Then the sample was sent for characterizations which was SEM and FTIR. The synthesis method in detail and the results of these will be shown in subsequent stages below.

2. Materials and methods
2.1 Materials
The metal salt solutions used were 1N Silver nitrate – AgNO3, 1N Gold chloride – AuCl3, 1N Strontium chloride – SrCl2. The root extracts prepared were of three plants Coriandrum sativum – coriander, Aloe indica – aloe Vera, Plectranthus amboinics – ajwain.

2.2 Method
The bottom up bio synthesis method is used. The roots were cleaned and then mashed by using mortar and pestle then the squashed roots were boiled in distilled water. Then the root remains were filtered out and the extract solution was stored in refrigerator. The tri metal alloy of Ag-Au-Sr was prepared by mixing 1:1:1 ratio of their 1 Normality salt solutions which is of silver nitrate, gold chloride and strontium chloride. 16.5ml of each of these salts were mixed together. Then 2ml of extract was added and a colour change was observed from colourless to light blue and then turned darker when heated and kept at room temperature. Then the sample was sent for characterization tests which are UV visible spectroscopy, SEM, FTIR, XRD.

3. Characterizations techniques
3.1 Field emission scanning electron microscopy
To conduct this test, the sample was sent to Osmania University, Hyderabad. Where the test was done on ZEISS Sigma field emission scanning electron microscope to get to know the morphology of the synthesized nano metal alloy.

### 3.2 Fourier transform infrared spectroscopy

This test was done on Nicolet 6700 FTIR spectrometer, Karnataka University, Dharwad to know the functional groups present in the synthesized nanometal alloy and also the chemical composition of the sample.

### 4. Results and Discussion

The first observation from the work was change of color of solution when the coriander root extract was added from colorless to blue when later when heated for 10 seconds turned dark. This allowed us to know the presence of tri-metallic alloy (Au Ag, Sr) nanoparticles (Fig 1).

![Figure 1: formation of tri-metallic alloy (Au Ag, Sr) nanoparticles](image)

#### 4.1 Field emission scanning electron microscopy

From this picture, we get to know that the shape of the nano particle generated as well as the size of the particle. The shape as we can see is almost spherical and the size was found to be 70nm. Also, the particles are agglomerated. We can also visualize that the surface of the particle is wavy and some amount of roughness exist. We can observe from the spectral image that along with the three particles an amount of carbon is also visible.

For analysis Gwyddion software was used, from where value of roughness of the surface was obtained. Also, 3D visualization can be made out of the obtained 2D image from FESEM. The agglomeration of
the particles is due to its hydrophilic nature. The presence of carbon is due to the usage of mica sheet as a substrate while sending the sample for characterization.

4.2 Fourier transform infrared spectroscopy

Figure 3 FTIR spectra of tri-metallic alloy (Au Ag, Sr) nanoparticles

The first absorbance peak of 3345.07 shows stretching of second degree amines (NH). Whereas the next peak of 1633.85 showed 1o R- NH2 with δ NH in plane (scissoring) with stronger bond than in 2o amines. The peak valued 1112.36 indicates the presence of aliphatic –C-O-C-.

5 Application studies

Gas sensing application

A setup was made to generate vacuum in a closed chamber where a probe applied with the sample needs to be placed and the gas needs to be inserted into this vacuum chamber through a nozzle from another container containing the gas. The probe was connected to an arduino UNO. A program to read the voltage value was uploaded into the arduino. Now, as the gas is sensed by the probe there is a variation in the voltage which indicates that gas has been sensed. This variable voltage gives rise to variable resistance. The sensing was taken at room temperature. The gases that were used are acetone, ethyl alcohol, methanol and ammonia.

The change in resistance value can be found using formula

$$Rs = \frac{(Vs-Vr)}{Vr} \times Rr$$

where

- $Rs =$ required change in resistance
- $Rr =$ resistance across reference
- $Vs =$ supply voltage
- $Vr =$ drop in voltage

Acetone sensing

Ammonia sensing
The voltage values were found to be 3.27V for acetone, 4.69V for ammonia, 3.40V for ethanol and 4.64V for methanol. Thus resistance values of these gases are in order ammonia < methanol < ethanol < acetone. Then conductivity of these gases are in reverse order that is acetone < ethanol < methanol < ammonia.

5. Conclusion
The nano metal alloy of gold silver and strontium was successfully synthesized of the size 70nm and are found of spherical shape. Also sensing studies showed sensing of gases like ethanol, methanol, acetone and ammonia.

Acknowledgement
This work is fully supported by KLE Technological University (Formerly known as B. V. Bhoomaraddi College of Engineering & Technology) under “Capacity Building Projects” grant Ref 2014-15 Sl. No 14. REU student Arvind B. and corresponding author Sharanabasava V Ganachari thanks KLE Society and KLE Technological University (BVBCET), Hubli for providing funding and support.

References
[1]. Shakeel A, Mudasir A, Babulal S and Saiqa I 2016 New Delhi, India. Journal of advanced research Rev 7, 17-28
[2]. C Sundarrajan, A Sankari, P dhandapani, S Maruthamuthu, S Ravichandran, G Sozhan and N Palaniswamy 2012. Karkauki, India Bioprocess Biosyst Eng. 35:827-833
[3]. K Saranyadevi, V subha, Ernest R and S Renganathan 2014. Chennai, India JJ. ChemTech Research 6 4533-4541
[4]. Ayesha M, M Khasif, S Arokiyaraj, M Bououdina, M G V Sankaracharyulu, M Jayachandran and U Hashim 2014. Digest J. nanomaterials and biostructures 9 1007- 1019
[5]. Amin B M, Farideh M, Mona M, Paridah T, Susan A and Rosfarizan M 2015. Malaysia Molecules. Rev 20 16540- 16565
[6]. Naveen S, Ganesh B, Preethi K 2015. India Indian J.Pharm.Biol.Res. Rev 3 13-27
[7]. R U Mene, M P Mahabole, R C Aiyer, R S Khairnar 2010. India Applied Physics J. 3 10-16
[8]. Sharanabasava V Ganachari, R Deshpande, R Bhat, NV Rao, DS Huh… Journal of Bionanoscience, 5, 2, 2011, 107-112.