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Synthesis of silver nanoparticles from curry, neem leaves, Citrobacter and its antimicrobial characterization against Staph and Proteus

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Abstract

Nanotechnology is the study and application of very small particles and may be used in different opposite fields namely biology, physics, chemistry, engineering and materials science. The silver nanoparticles (Ag-NPs) have fascinated growing interest because of their different biological and physiochemical properties and in between 1-100 nm of size. The purpose of current study was to investigate the synthesis of silver nano particles from Curry, Neem leaves, Citrobacter and checks its antimicrobial activity against Staph and Proteus. We have taken 2 samples of different plants made extract and synthesized the silver nano particles from Curry, Neem extract and microorganism. The higher values were recorded for Curry, Neem leaves extract and Citrobacter from 350 to 450 wavelengths respectively by the graphs. The current findings were concluded that the Curry, Neem leaves extract and Citrobacter play an important role inside the reduction and stabilization of silver to silver nanoparticles. It is utilized in several medicines and cosmetics etc, because it has conductive and optical properties.

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Introduction

Nanotechnology is the study and application of very small particles and may be used in different opposite fields namely biology, physics, chemistry, engineering and materials science. Applied science is quickly rising by producing the nanoparticles (NPs) which will have new and different physical and chemical properties and size from higher matter. These new properties of NPs are exploited in a very large choice of probable uses in drugs, makeups, renewable energies, ecological modification and remedy devices. In between them, the silver nanoparticles (Ag-NPs) have fascinated growing interest because of their different biological and physiochemical properties. These are the silver nanoparticles of in between 1-100 nm of size. (Syafiuddin *et al.*, 21017). In command to exploit the possible uses of AgNPs, the correct information of their properties is vital. The merit and demerit of AgNPs, using must be obviously calculated and qualified to recognize their properties on the environment. Crystal-like shape of AgNPs may be resulting from X-ray visual phenomenon designs. Many studies indicate that the AgNPs have the cube-shaped structure. Several studies have indicated that AgNPs fascinate radiation within the visual region from 380 to 450 nm by recommends that of a development called the excitation of restricted surface (Lok *et al.*, 2007).

The Curry tree mostly concerned with hot to sub-tropical containing dicot family. It is cosmopolitan in India and Sri Lanka with little tree growing up to 4–6 m tall. Curry leaf has freshly been found to be a powerful inhibitor thanks to great absorptions of carbazoles, a water soluble heterocyclic compound. They are also used as herb in Ayurvedic remedy medication. They also possess the properties against diabetes mellitus. The Indian as well as the Sri Lankan used their leaves as spinach and normally cooked in combination with onion sliced within the early phase of the preparation (Sajeshkumar *et al.*, 2015).Neem plant is mostly grown with hot to semi-tropical regions of the world containing meliaceae family. It is cosmopolitan in different countries like India, Nepal, Pakistan, Bangladesh, Sri Lanka and Maldives. It is a medicative plant used for different treatment of microorganism including fungi and bacteria. (Ahmed *et al.*, 2016). The margosa like liquid is extract form the plant used as antipyretic properties, bactericide activity and also used as to lower the higher fever (Verma and Mehata, 2016). Citrobacter belong to a genus containing gram-negative coliform bacteria of Enterobacteriaceae family. The species containing C. amalonaticus, C. koseri, and C. freundii respectively. It is distinguished by their aptitude to change vital amino acid to indole and as a sole carbon supply. The Citrobacter microorganism is present almost every apartment in soil, water and wastewater etc. It is also present in human alimentary canal. (Saeb *et al.*, 2014).

Medicinal plants are plentiful cause of antimicrobial molecules. A widespread series of medicinal plants extracts are applied for the cure of different pathogenic diseases, as they have probable antimicrobial activity. Few of them bioactive particles are separated and operated in market as raw product for numerous herbal companies (Renisheya *et al.*, 2011). The purpose of current study was aimed to synthesis of silver nano particles from Curry, Neem leaves, Citrobacter and checks its antimicrobial activity against Staph and Proteus.

Materials and methods

We have taken leaves of Neem and Curry tree form Karachi, Pakistan. First washed clearly with distal water for the removal of dust and dirts materials. After put it in a hot air oven at 128 °C for 10 minutes to dry the leaves and made powder in grinder.

Aqueous extraction

First taken 10g of powder in 100ml of distal water in conical flask. Cover the mouth of conical flask with cotton pluck. Set the temperature of water bath at 80 °C and after temperature reached 80 °C, kept the conical flask in water bath for 10 minutes. Filter the extract through whatman 1 filter paper. Collect the extract in separate sterile conical flask and stored the conical flask in refrigerator at 4 °C respectively. The same procedure was also repeated for Curry and

Neem leaves (Figure 1&2).

Synthesis of silver nano particles

Taken 2.5ml of ammonium solution in separate sterile conical flask. Added 10ml plant extract and 32.5ml deionized water in conical flask. After looked the solution color who changed from yellowish to brown indicated the presence of silver nano particles. Kept all solution in Erlenmeyer flask and incubated at 37°Cunder agitation (200 rpm) for 24-48 hours.

Procedure of silver nano particles by using microorganism

First taken 5 ml extract in silver nanoparticles synthesize in sterile conical flask and added 5 ml nutrient broth in which culture of Citrobacter was present. After it incubated for 24 hours at 37 °C.Then inoculate it in already prepared extraction and kept in water bath at 37 °C for 48 hours under agitation 200rpm (Figure 3&4).

Antimicrobial activity of silver nano particles

Taken MHA plates, made stain of Staph and Proteus on it and made a holes with the help of sterile Bauer follow the Kirby Bauer method.

After pour the crude extract, where the silver nano particles is present and synthesized by Citrobacter at different concentrations , silver nitrate ($AgNO_3$) as a control is used and placed erythromycin antibiotic on plates . Incubated the plates at 37°C for 24 hours to checked antimicrobial activity of silver nano particles against Staph, Proteus and erythromycin antibiotic (Figure 5&6).

Results

We have taken 2 samples of different plants made extract and synthesizedthe silver nano particles from Curry, Neem extract and microorganism. It also checked its frequency at different wavelengths, presented by various abovegraphs.

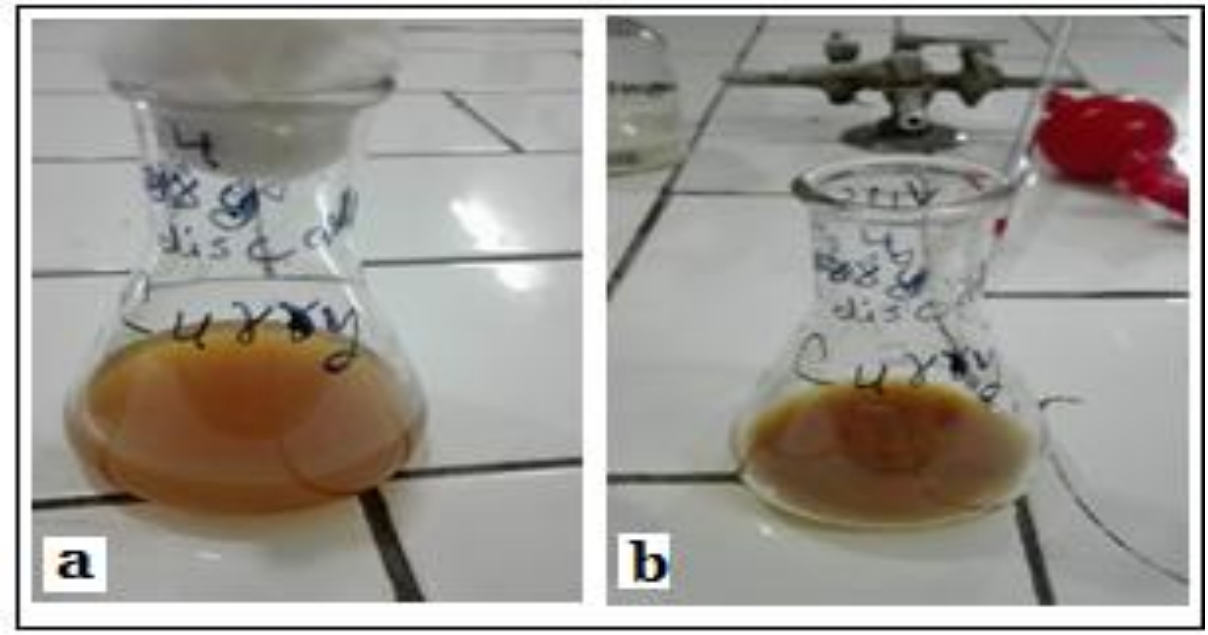

Fig. 1. (a) Crude extract of Curry; (b) Silver nano particles from Curry.

The graphs indicated different wavelength with different frequencies. We also made silver nano particles from Citrobacter. It gives oxidase negative and citrate positive with mucoid colonies onEosin-Methylene Blue. Brownish yellow color indicated the confirmationof silver nano particle production. We have shownto check the zone by yellow agar diffusion method and placed the erythromycin antibiotic which shows high antimicrobial activity against Proteus and small antimicrobial activity against Staph. Erythromycin is an antibiotic which inhibit protein synthesis and silver nano particles are also inhibit growth of Proteus which are synthesized from curry leave extract and microorganisms.

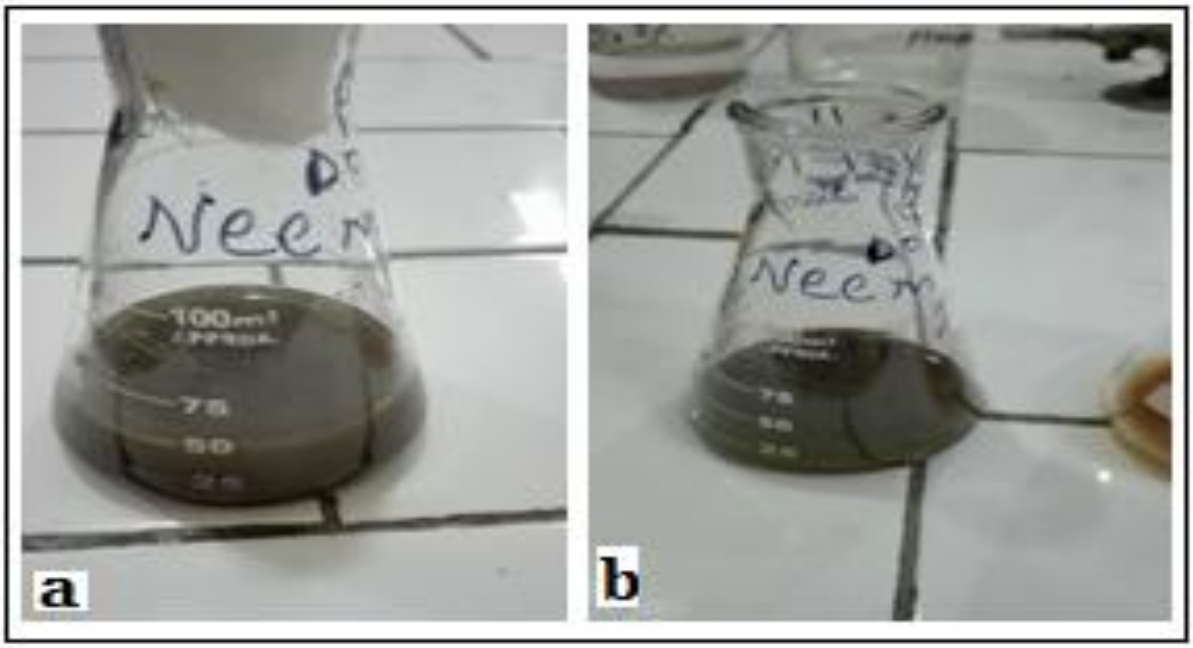

Fig. 2. (a) Crude extract of Neem; (b) Silver nano particles from Neem.

We also shown the zone of silver nano particles who synthesized by Citrobacter. It is indicated that the silver nanoparticles who synthesized from Curry, Neem leaves extract and Citrobacter recorded high values with range of 350 - 450 wavelengths respectively (Figure 7, 8&9).

Fig. 3. Mucoid colonies of Citrobacter on Eosin-Methylene Blue agar.

Discussion

In the present we have taken 2 samples of different plants made extract and synthesizedthe silver nano particles from Curry, Neem extract and microorganism. It also checked its frequency at different wavelengths, presented by various above graphs.

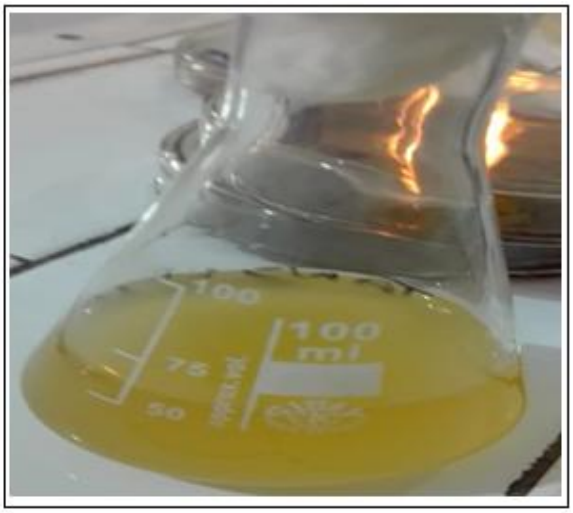

Fig. 4. Presence of Citrobacter in nutrient broth.

The graphs indicated different wavelength with different frequencies. We also made silver nano particles from Citrobacter.

It gives oxidase negative and citrate positive with mucoid colonies onEosin-Methylene Blue. Brownish yellow color indicated the confirmationof silver nano particle production. We have shownto check the zone by yellow agar diffusion method and placed the erythromycin antibiotic which shows high antimicrobial activity against Proteus and small antimicrobial activity against Staph.

Fig. 5. Zone of inhibition shows silver nano particles inhibit growth of Proteus.

Fig. 6. Biochemical test.

It is indicated that the silver nanoparticles who synthesized from Curry, Neem leaves extract and Citrobacter recorded high values with range of 350 - 450 wavelengths respectively.

Manufactured silver nanoparticles were considered by UV spectrometry. The highest follows at 435 nm which resembles to the transmittance of silver nanoparticles.

The concentration of the highest at 435 nm was improved with time until the decrease finishes. The synthesis of silver nanoparticles reduces after 120 minutes. The extreme highest was originate to be 435 nm for *Murraya koenigii*. Similarly the highest value for Curry plant was recorded at 420 nm respectively (Christensen *et al.,* 2011).

The peak absorptions of silver salts confine the usage of them in medicine now a day. Usage of nanoparticles metals, reductions the absorption of metal salts and silver. The bacteriological result of nano particles metals has been recognized to their minor size and peak surface to volume ratio which permits them to cooperate carefully with bacterial films and is not only due to relief of metal ions in clarification or in producing dishes.

The manner of exploit of both silver ions and nano particles were recorded to be a shame, while the nano particles were recorded to be active at suggestively lesser absorption than the ions (Morones *et al.,* 2005).

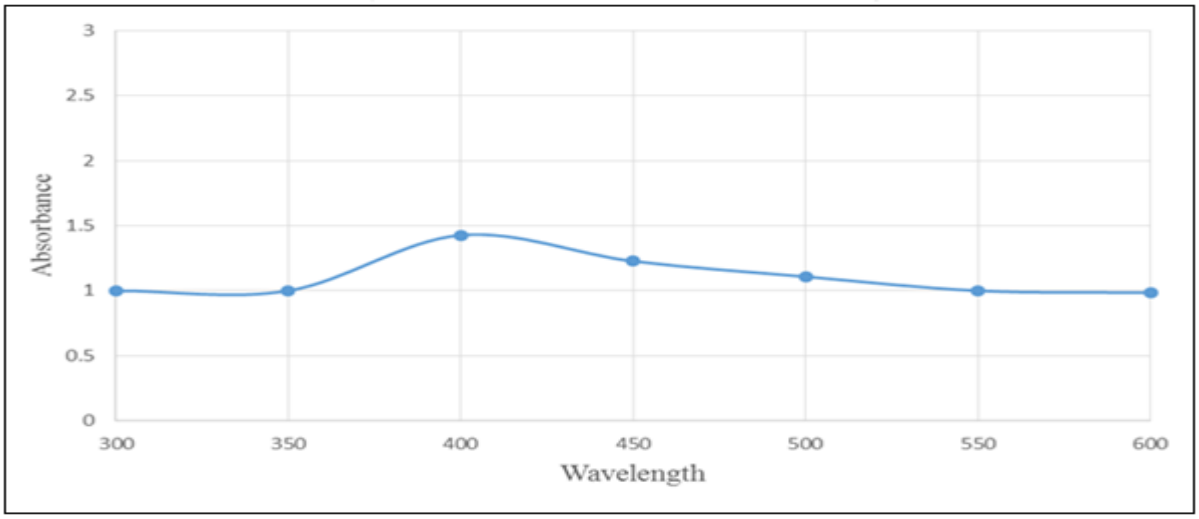

Fig. 7. Silver nano particles from Curry extract.

The part of both nano particles and silver ions to the cell film produced acclimization of covering protein parts producing degeneracy of the protein cause power (Lok *et al.*, 2007).To classify the probable biomolecules existing in Neem leaf broth which are liable for covering and soothing the nano particles metals, in which the fourier transform infrared (FTIR) spectra were restrained. The FTIR spectra were restrained for clean Neem leaf broth and sanitized AgNPs found after centrifugation of sample at 10,000 rpm. When AgNO3 is poured in to aqueous Neem leaf solution, resulting a color variation from pale yellow to yellow brown and lastly to dark brown color is appeared (Verma and Mehata, 2016).

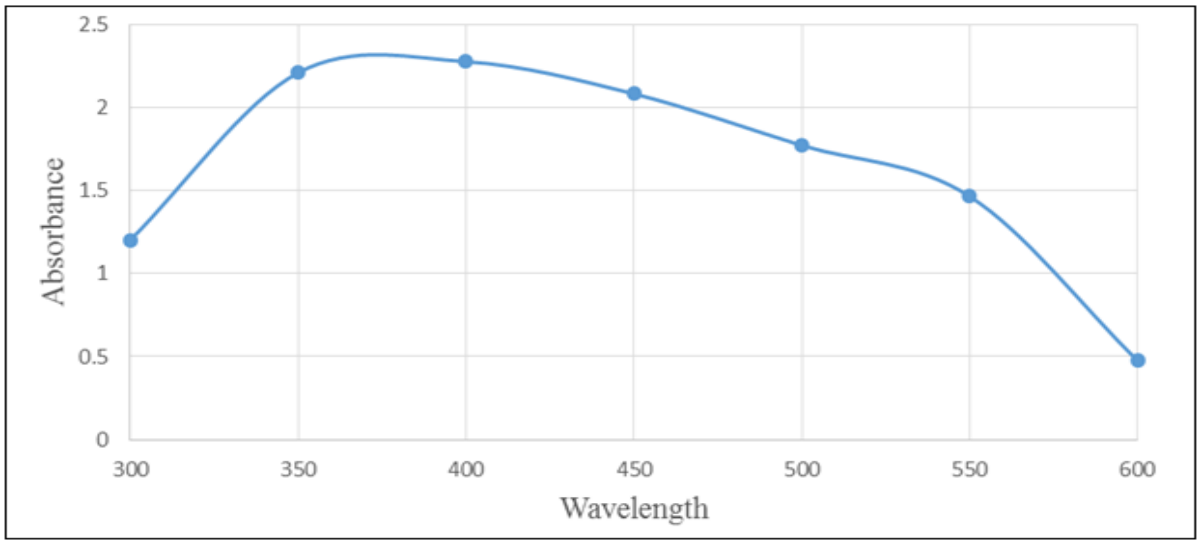

Fig. 8. Silver nano particles from Neem extract.

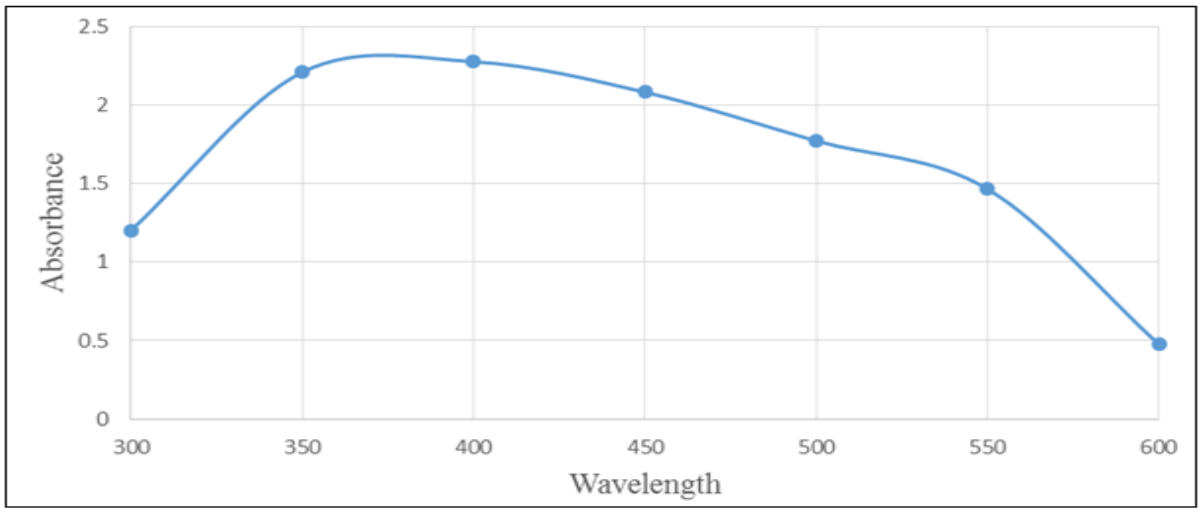

Fig. 9. Silver nano particles from Citrobacter.

The alteration in color of the extract is due to the occurrence of silver nano particles designed by the decrease of silver salt. The decrease of silver salt to silver ions is due to the occurrence of reducing agents. It was recommended that the molecules like theophylline and caffeine react as reducing agent when *Acalypha indica* leaf solution was added (Krishnaraj *et al.*, 2010).

Conclusion

The current findings were concluded that the Curry, Neem leaves extract and Citrobacter play an important role inside the reduction and stabilization of silver to silver nanoparticles. It also showed medicinal activity on every gram positive and gram negative microorganism. Furthermore, it is utilized in several medicines and cosmetics etc., because it has

conductive and optical properties. It has low priced, material free and eco-friendly.

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