Green synthesis of bio-inspired silver nanoparticles using plants extract and UV characterization

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Abstract

Nanotechnology deals with the Nanoparticles having a size of 1-100 nm in one dimension used significantly concerning medical chemistry, atomic physics, and all other known fields. Nanoparticles are used immensely due to its small size, orientation, physical properties, which are reportedly shown to change the performance of any other material which is in contact with these tiny particles. It is gaining attention due to its cost effective, ecofriendly and large scale production possibilities. In this present study four plants *Cissus quadrangularis*, *Adhatoda vasica*, *Gymnema sylvestre* and *Tinospora cordifolia* were taken to investigate their potential for synthesizing silver nanoparticles. The silver nanoparticles synthesized were confirmed by their change of colour to dark brown due to the phenomenon of surface plasmon resonance. The characterization studied was done by UV-Vis spectroscopy. By UV- characterization method, we confirmed that *Cissus quadrangularis* and *Adhatoda vasica* have their bands near 420nm so that these two plants extract have more potential to reduce Ag ions into Ag nanoparticles than *Gymnema sylvestre* and *Tinospora cordifolia*., which lead us for further research on synthesis of silver nanoparticles from these two plants.

Keywords: Nanotechnology, Nanoparticles, Green Synthesis, UV-Vis spectroscopy.

1. INTRODUCTION

1.1 Overview:

Nanotechnology can be termed as the fabrication, characterization, exploration and application of nanosized (1-100 nm) materials for the development of science. It deals with the study of extremely minute structures and the prefix “nano” is a Greek word which means “dwarf or miniature”. Nanotechnology has been spread to number of areas including biomedical services, cosmetics, drug gene delivery, environmental health, food, health care, catalysis, mechanics, nonlinear optical devices, optics, photo-electrochemical application, single electron transistors, space industries (Wang, 1991).

In recent days nanotechnology has induced great scientific advancement in the field of research and technology. Nanotechnology is the study and application of small object which can be used across all fields such as chemistry, physics, biology, material science and engineering. Nanotechnology is a fast growing area in the field on science which is an interdisciplinary field of both science and technology that increase the scope of investing and regulating at cell level between synthetic material and biological system (Du.*et al.,* 2006 & Sinha *et al.,* 2009). Nanotechnology proceeds by three processes - separation, consolidation, deformation of material by one atom or molecule (Taniguchi 1974). It is divided into three types- Wet nanotechnology which deals with the biological system such as enzymes, membrane, cellular components. Dry nanotechnology deals with the surface science, physical chemistry & gives importance on fabrication of structure in carbon, silicon, inorganic materials. Computational nanotechnology which deals with modeling & stimulating the complex nanometer scale structure (Sinha *et al.,* 2009), these three fields are interdependent to each other.

Nanoparticles can be synthesized using various approaches including chemical, physical, and biological. Although chemical method of synthesis requires short period of time for synthesis of large quantity of nanoparticles, this method requires capping agents for size stabilization of the nanoparticles. Chemicals used for nanoparticles synthesis and stabilization are toxic and lead to non-ecofriendly byproducts. The need for environmental non-toxic synthetic protocols for nanoparticles synthesis leads to the developing interest in biological approaches which are free from the use of toxic chemicals as byproducts. Thus, there is an increasing demand for "green nanotechnology" (Singhal *et al.,* 2011). Many biological approaches for both extracellular and intracellular nanoparticles synthesis have been reported till date using microorganisms including bacteria, fungi and plants (Mukherjee *et al.,* 2001 and Spring, 1995). The field of nanotechnology is one of the upcoming areas of research in the modern field of material science. Nanoparticle show completely new or improved properties, such as size, distribution and morphology of the particles etc. Novel applications of nanoparticles and nanomaterials are emerging rapidly on various fields (Kaviya and Viswanathan, 2011). Metal nanoparticles have a high specific surface area and a high fraction of surface atoms (Catauro *et al.*, 1995) because of the unique physicochemical characteristics of nanoparticles (Crabtree *et al.*, 2003), including catalytic activity, optical properties, electronic properties, antibacterial properties, and magnetic properties (Krolikowska *et al.,* 2003). They are gaining the interest of scientist for their novel methods of synthesis (Zhao, 1991). The biological synthesis of nanoparticle is a challenging concept which is very well known as green synthesis. The biological synthesis of nano material can solve the environmental challenges like solar energy conservation, agricultural production, catalysis (Kumar *et al.,* 2011), electronic, optics (Evanoff and chumanov, 2005), and biotechnological area (Soloviev, 2007).

Nanoparticle of gold, silver, copper, silicon, zinc, titanium, magnetite, palladium formation by plants has been reported. Colloid silver nanoparticle had exhibited distinct properties such as catalytic, antibacterial (Sharma *et al.*, 2009), good conductivity, and chemical stability. Silver nanoparticles have its application in the field of bio labeling, sensor, antimicrobial, catalysis, electronic and other medical application such as drug delivery (Jong *et al.,* 2008) and disease diagnosis.

Sometimes the synthesis of nanoparticles using various plants and their extracts can be advantageous over other biological synthesis processes which involve the very complex procedures of maintaining microbial cultures (Sastry *et al.*, 2003 and Khan *et al.*, 2003). Many such experiments have already been started such as the synthesis of various metal nanoparticles using fungi like *Fusarium oxysporum* (Nelson *et al.*, 2005), *Penicillium* sp. (Hemanth *et al.*, 2010) and using some bacteria such as *Bacillus subtilis* etc. (Natarajan and Ramchandra, 2010). But, synthesis of nanoparticles using plant extracts is the most adopted method of green, eco-friendly production of nanoparticles and also has a special advantage that the plants are widely distributed (Elumalai *et al.*, 2010), easily available, much safer to handle and act as a source of several metabolites (Ankamwar *et al.*, 2005). There have also been several experiments performed on the synthesis of silver nanoparticles using medicinal plants such as *Oryza sativa*, *Helianthus annus*, *Saccharum officinarum*, *Zea mays*, *Basella alba*, *Aloe vera*, *Capsicum annuum*, *Magnolia kobus*, *Medicago sativa* (Alfalfa), *Cinamomum camphora* and *Geranium sp*. in the field of pharmaceutical applications and biological industries. Besides, green synthesis of silver nanoparticles using a methanolic extract of *Eucalyptus hybrida* was also investigated (Kasthuri and Rajendran, 2009).

In the recent days, silver nanoparticles have been synthesized from the naturally occurring sources and their products like green tea (*Camellia sinensis*), Neem (*Azadirachta indica*) , leguminous shrub (*Sesbania drummondii*) , various leaf broth, natural rubber, starch , Aloe vera plant extract , lemongrass leaves extract, etc. (Vijayraghavan *et al.*, 2012). With respect to the microbes, the silver nanoparticles get attached to the cell wall, thereby disturbing the permeability of cell wall and cellular respiration. The nanoparticles may also penetrate deep inside the cell wall, thus causing cellular damage by interacting with phosphorus and sulfur containing compounds, such as DNA and protein, present inside the cell. The bacteriocidal properties of silver nanoparticles are due to the release of silver ions from the particles, which confers the antimicrobial activity (Amarendra, 2010).

Plant mediated synthesis is purely a green synthetic route and are considered better candidates among the different biological entities as they provide clean, ecofriendly, cost effective, safe, conveniently utilizable and beneficial way to the synthesis of metal Nps for the large scale production. Many plants are reported to facilitate the formation of AgNPs and their potential applications (Chandran *et al.*, 2006; Saxena *et al.*, 2010; Waidha *et al.*, 2015). The amount of accumulation of NPs varies with reduction potential of ions and the reducing capacity of plant depends on the presence of various polyphenols and other heterocyclic compounds (Nair *et al.*, 2010; Vedpriya, 2010). Plants are known to harbor a big range of metabolites that are most likely to be responsible in the green synthesis of metal nanoparticles. Biotechnological approaches, exclusively plant tissue cultures provide a better platform in producing specific phytoconstituents at a rate similar or superior to that of intact plants. Explants are cultured under appropriate physiological conditions and the desired product is extracted from the cultured cells/tissue by means of plant tissue culture. However, the biosynthetic capacity of cultured plant tissue can be enhanced by regulating environment factors, which are effective for the high production of NPs.

1.2 Properties of Nanoparticles

1. They can be built by assembling individual atoms or subdividing bulk materials (Charles *et al.*, 2003).

2. Size of nanoparticles is less than wavelength of light (Khanna, 2008).

3. Physically, materials can be characterized by some critical length, a thermal diffusion length or a scattering length (Charles *et al.*, 2003).

4. Vander wall forces or magnetic forces plays more important role than gravitational forces (Khanna, 2008).

5. Critical characteristics are their very high surface-to-volume ratio (Khanna, 2008).

1.3 UV-Vis Spectroscopy:

The optical properties of a solution are determined by Absorbance spectroscopy. A light beam is passed through the sample solution and the amount of absorbed light is measured. At each different wavelength, the absorbance is measured. By using Beer-Lamberts Law, the concentration of the solution is measured by absorbance. This technique is limited to sample identification but is very useful for quantitative measurements. For instance, the measured absorbance spectrum does not necessarily show the actual absorbance but the extinction of the light is both the absorbed and the scattered light from the particles. These wave lengths arise due to the surface Plasmon resonance of the particle.

UV-Vis spectroscopy is useful to characterize the transmission, adsorption and reflectivity of a variety of technologically important materials, such as coatings, pigments etc. In single-beam and dual-beam spectrometers, the light from a lamp is dispersed before reaching the sample cell. In an array detector instrument, all wavelengths pass through the sample and the dispersing element is between the sample and the array detector. The dual beam spectrophotometer was used to obtain the absorbance vs. wavelength curve of silver nanoparticles in this research.

2. OBJECTIVES

Therefore, keeping in mind the above considerations, the experiments were conducted with the following objectives-

1. Identification of plants (*Cissus quadrangularis, Adhatoda vasica, Gymnema sylvestre* and *Tinospora cordifolia*).
2. Preparation of plants extract.
3. Synthesis of silver nanoparticles from plants extract. Characterization of silver nanoparticles by UV-Vis spectroscopy.
4. Evaluation of antimicrobial activity of silver nanoparticles

3. MATERIALS REQUIRED

Chemicals-

1. Silver Nitrate – Purchased from local chemical suppliers
2. Ethanol
3. Acetone

Other Materials-

1. leaves of four plants- *Cissus quadrangularis, Adhatoda vasica*, *Gymnema sylvestre* and *Tinospora cordifolia*
2. Distilled water– Collected from DI plant

Other Appartus-

- Weight Machine
- Centrifuge machine
- Beaker
- Magnetic Stirrer
- Ultracentrifuge machine
- Autoclave
- Hot Plate
- Inoculating needle
- Funnel
- Refrigerator
- Flask
- Parafilm
- Filter paper
- Tissue paper
- Sieve

4. METHODOLOGY

4.1. Preparation of Plants extract:

- Plants parts are collected (stem, leaves).
- Washed several times to remove dust particles.
- Air dried or shade dried for 10 days.
- The fine powder obtained from stem and leaves.
- 5g of powdered sample and 100 ml of distilled water were added and boiled to 60-70°C for about 10 minutes.
- Then the resulting crude extracts filtered by standard filtration method.

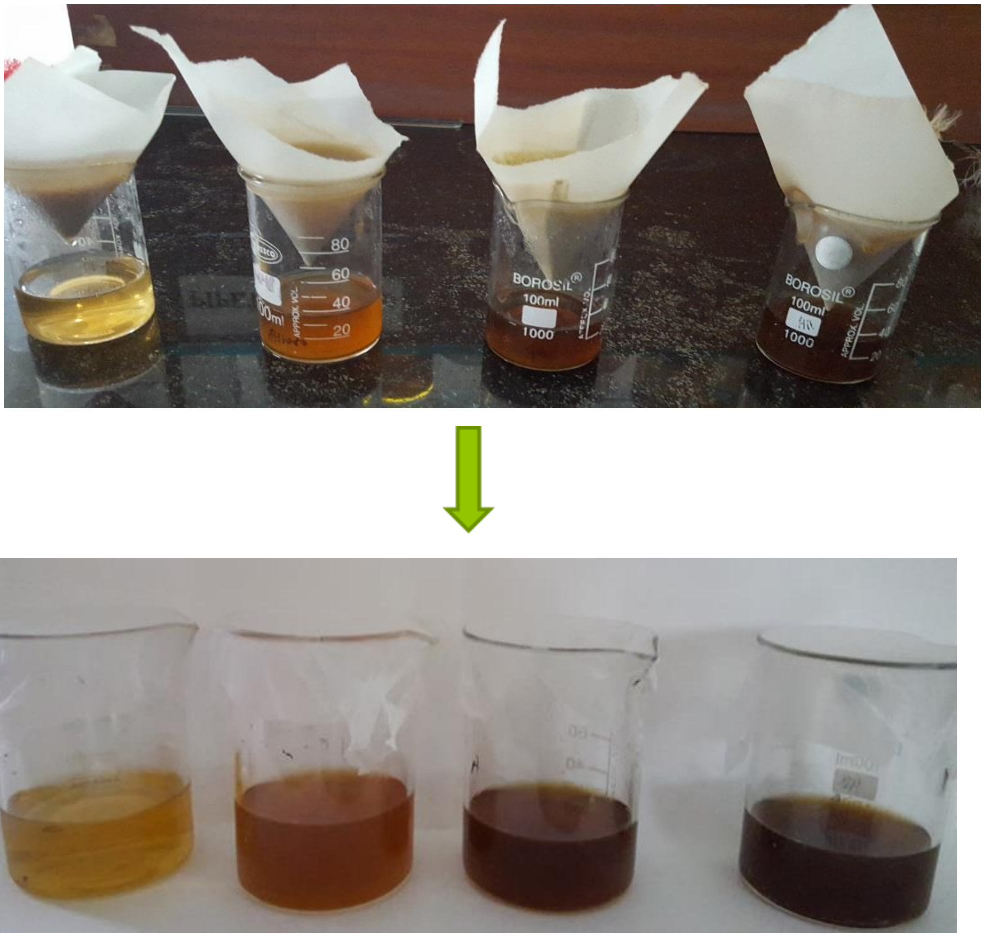
4.2 Synthesis of silver nanoparticles:

- From all the plants extract, each 10 ml of plant extract was added into 90 ml of prepared aqueous solution of 1 mm silver nitrate for reduction into Ag+ ions.

4.3 Characterization of silver nanoparticles by UV- Vis spectroscopy

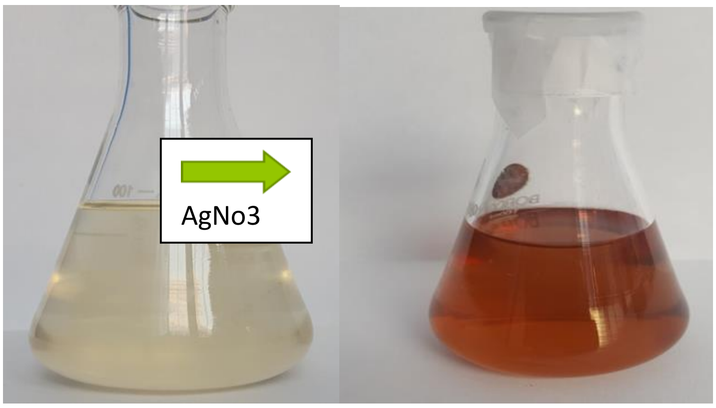
5. RESULTS

5.1 Preparation of plants extract:

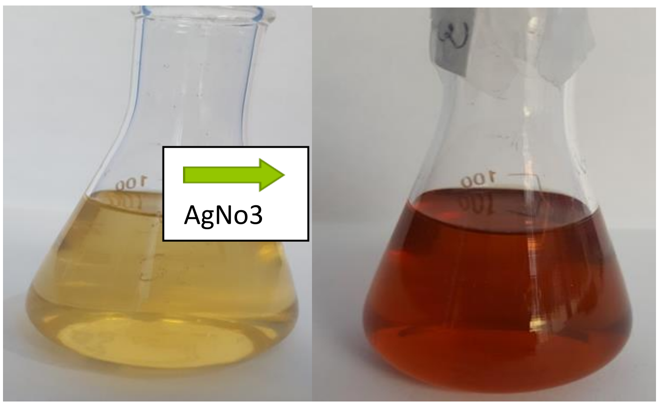


Preparation of plants extract: *cissus quadrangularis* (a), *Adhatoda vasica* (b), *gymnema sylvestre* (c), *Tinospoora cordifolia* (d).

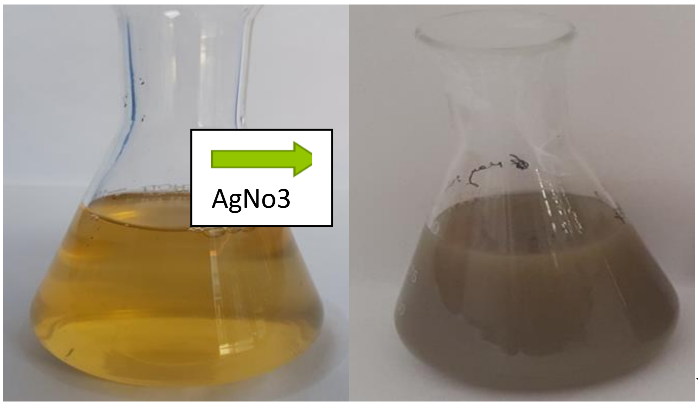
5.2 Synthesis of silver nanoparticles:



Before (a) After

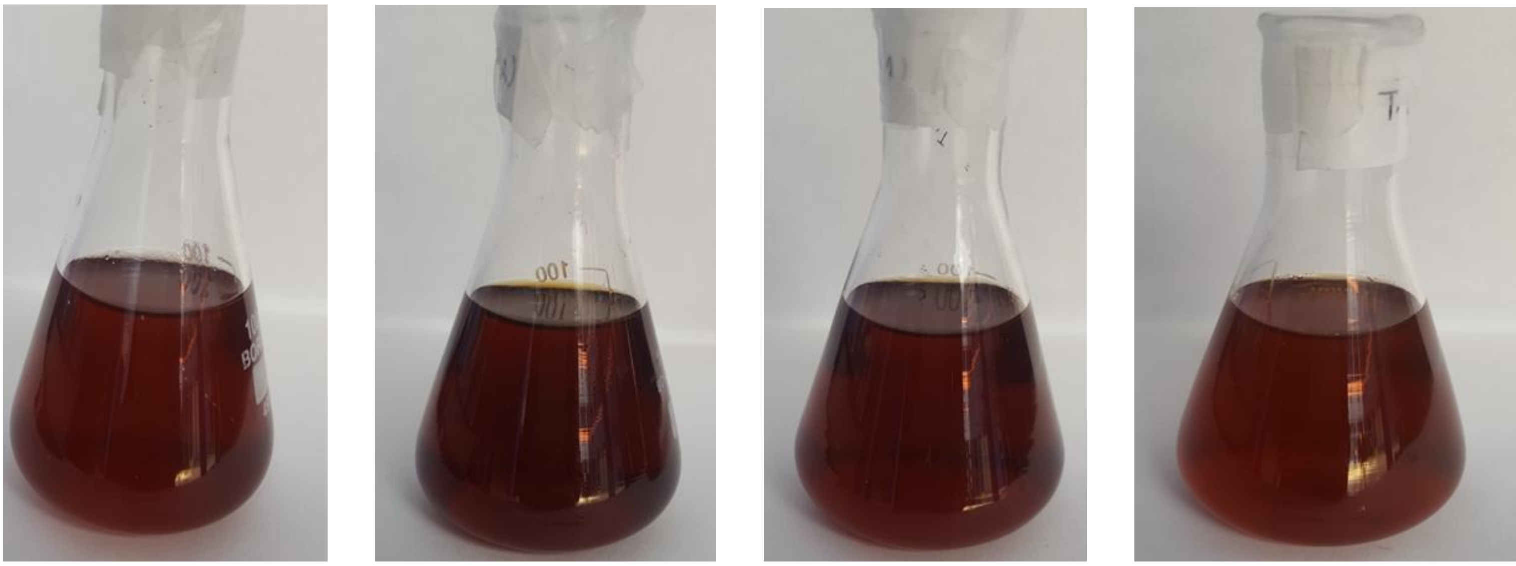


Before (b) After



Before (c) After Before (d) After

Colour change of plant extract before after addition of AgNO3 (a) *Cissus quadrangularis*, (b) *Adhatoda vasica*, (c) *Gymnema sylvestre*, (d)*Tinospora cordifolia.*

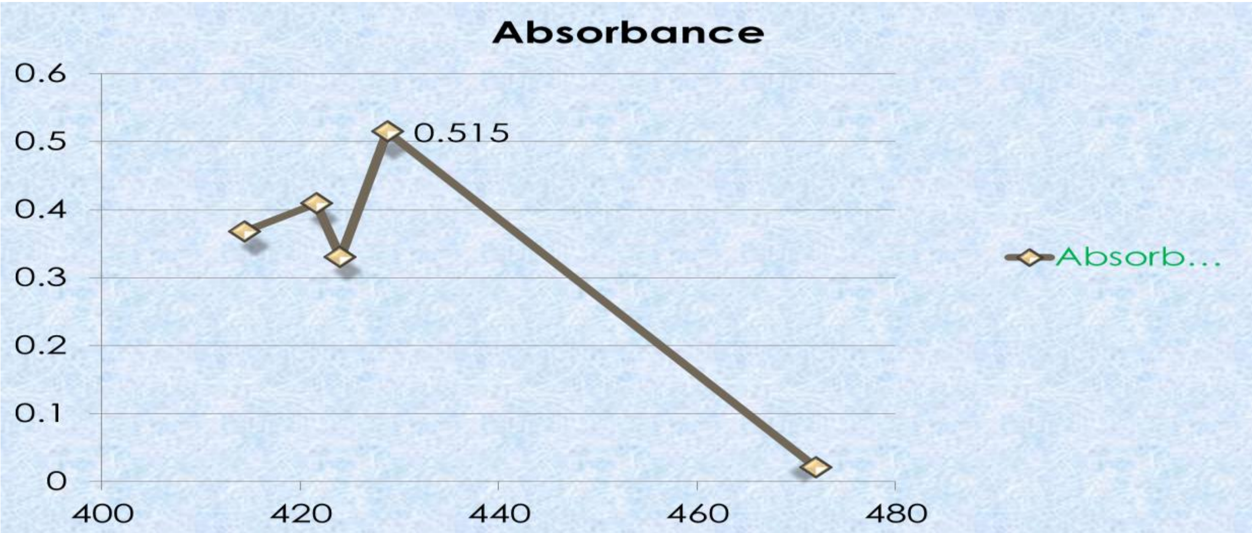


(a) (b) (c) (d)

After 24 hours in room temperature (a) *Cissus quadrangularis*, (b) *Adhatoda vasica*, (c) *Gymnema sylvestre* , (d) *Tinospora cordifolia*.

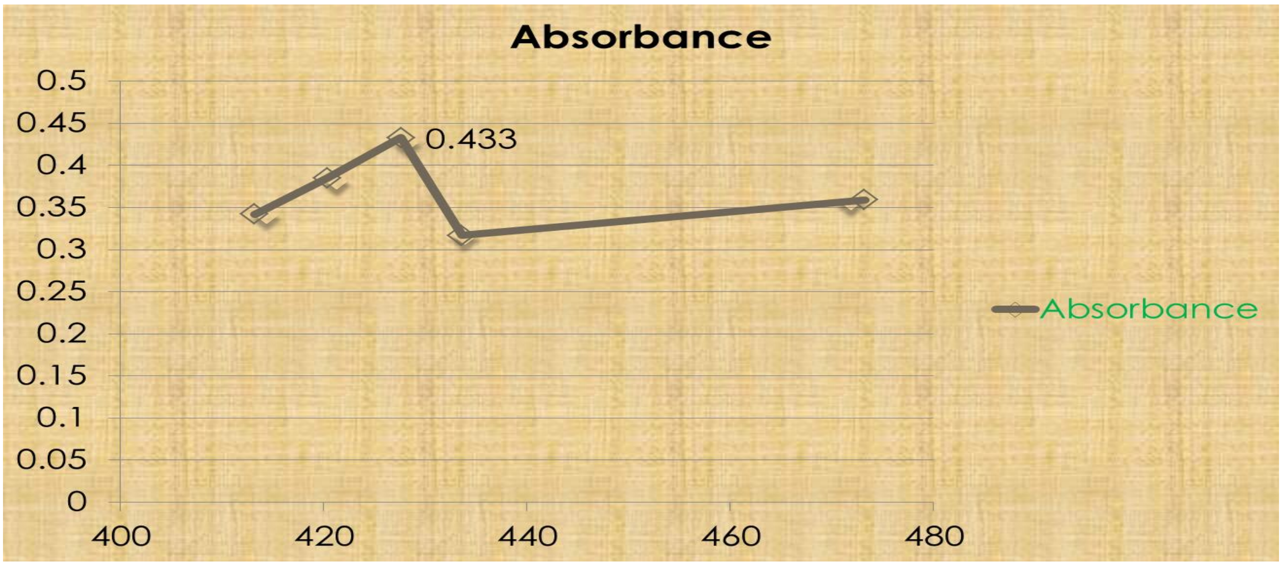
At different concentration of AgNo3 colour differntiation of nanoparticles in solution at room temperature temperature (a) *Cissus quadrangularis*, (b) *Adhatoda vasica*, (c) *Gymnema sylvestre* , (d) *Tinospora cordifolia*

5.3 UV-Vis Characterization:



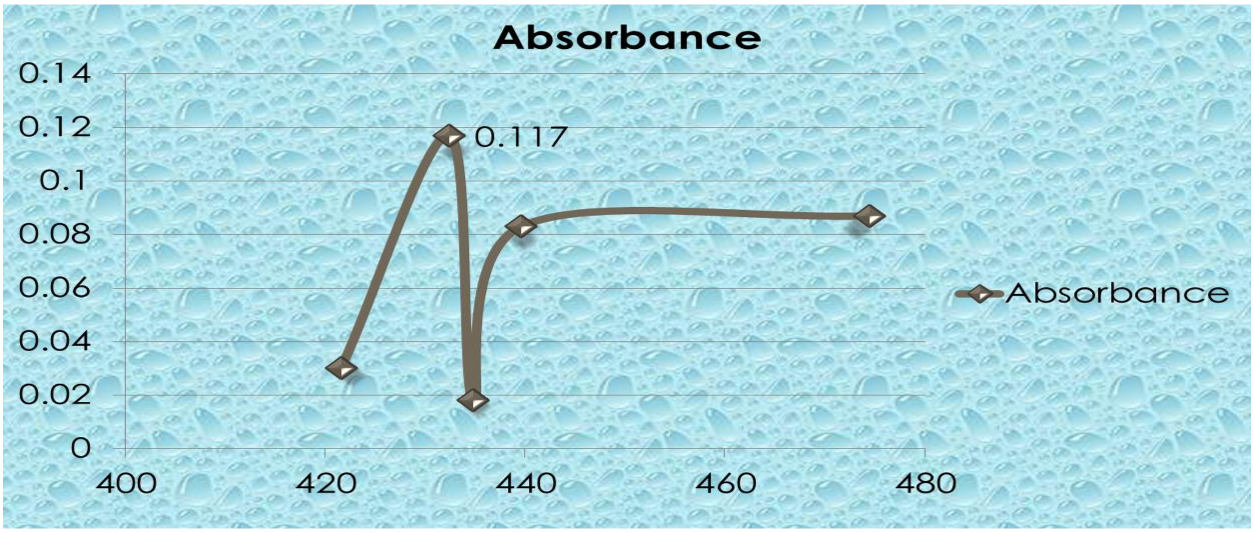
Wavelengths (nm)

The absorbance spectrum of silver nanoparticles showing maximum absorbance near 428.8 nm in *Cissus quadrangularis*



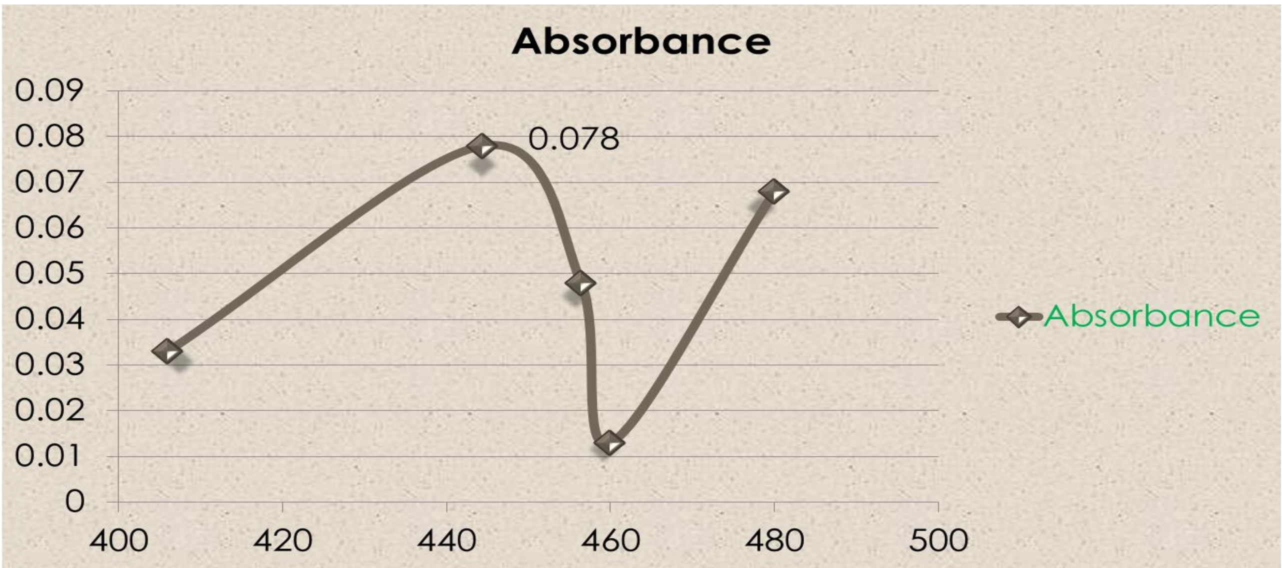
Wavelength (nm)

The absorbance spectrum of silver nanoparticles showing maximum absorbance near 427.6 nm in *Adhatoda vasica*



Wavelength (nm)

The absorbance spectrum of silver nanoparticles showing maximum absorbance near 432.4 nm in *Gymnema sylvestre*



Wavelength (nm)

The absorbance spectrum of silver nanoparticles showing maximum absorbance near 444.4 nm in *Tinospora cordifolia*

6. RESULTS AND DISCUSSION

According to literature studies silver nanoparticle solution has dark brown or dark reddish in colour. In *Cissus quadrangularis*, before addition of AgNo3, its colour was pale yellow but after its treatment with AgNo3 its color changes to dark brown which indicated the formation of silver nanoparticles. Likewise all the other three plants extact (*A. vasica*, *G. sylvestre*, *T. cordifolia*) changed to dark brown after treatment with $AgNO_3$ and the colour intensified after 48 hours. The control sets showed no change in color under the same experimental conditions. The color is characteristic of the surface plasmon resonance (SPR) of silver nanoparticles. This color change is due to the property of quantum confinement which is a size dependent property of nanoparticles which affects the optical property of the nanoparticles.

The reduction of silver ion to silver nanoparticle was reflected in spectral data obtained by using a UV-Vis spectrophotometer. The UV absorption peak of silver nanoparticles range from 400 nm –480 nm (Ramteke *el al.*, 2013). The sharp bands of silver nanoparticles were observed around 428nm in case of *C. quadrangularis* whereas the bands for *A. vasica* observed around 427.6 nm. In case of *G. sylvestre* and *T. cordifolia* sharp bands are observed on 432.4 and 444.4 nm. From different literatures it was found that the silver nanoparticles show SPR peak at around 420 nm. From our studies we found the SPR peak for *C. quadragularis* and *A. vasica* have their bands on near about 420nm wavelength so we confirmed that these two plants extract have more potential to reduce Ag ions into Ag nanoparticles than *G. sylvestre* and *T.* cordifolia., which lead us for further research on synthesis of silver nanoparticles from these two plants. The intensity of absorption peak increases with increasing time period.

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