# CHAPTER ONE

# INTRODUCTION AND LITERATURE REVIEW

## 1.0 INTRODUCTION

Nanoscience has been the subject of substantial research in recent years. It has been explored by researchers in various fields of science and technology (*Kholoud et al. 2010*). The novel properties of NPs have been exploited in a wide range of potential applications such as in medicine, cosmetics, renewable energies, environmental remediation, biomedical devices (*Quang Huy, 2013*), electronics, optics, organic catalysis, vector control, sensor, etc., have drawn extensive attention to this field of study (*Mousavand et al. 2007*). Among the metals, silver nanoparticles have shown potential applications in various fields such as the environment, bio-medicine, catalysis, optics and electronics (*Rao et al., 2000*). Silver nanoparticles are mostly smaller than 100 nm and consist about 20–15,000 silver atoms. In its nanoscale form, silver exhibits unique physicochemical and biological activities. This has made them useful as sensor, vector control, antimicrobial, anticancer, and antiplasmodial agents, catalysts, among others (*Elemike et al. 2014; Vinod et al. 2014; Kathiravan et al. 2014; Saraschandra and Sivakumar 2014; Namita and Soam 2014*).

Concerted effort has been made to synthesize diverse range of silver nanoparticles varying in size, geometry, and morphology because of their potential applications, particularly in electronics (*P. V. Kamat, 2002*), electrochemical sensing (*L. M. Liz-Marzán, 2006*), catalysis (*F. Zhang, Y. Pi et al., 2007*), and antimicrobial properties (*T. Sakai et al., 2006*). The size, geometry, dispersion and stability often determine the suitability of the nanoparticles for certain applications. Synthesis may involve physical means such as ultraviolet light, microwaves, photo-reduction, or chemical reduction using hydrazine, ascorbic acid, sodium borohydride, glucose, and organic stabilizers or biological means using plant extract, microorganism or plant sap. Several physical and chemical methods have been used to synthesize and stabilize silver nanoparticles (*Senapati et al., 2005, Klaus-Joerger et al., 2001*). The most popular chemical approaches, including chemical reduction using a variety of organic and inorganic reducing agents, electrochemical techniques, physicochemical reduction, and radiolysis are widely used for the synthesis of nanoparticles.

Although these means are fast and easy, they are either expensive or toxic particularly the chemical method and may lead to non eco-friendly byproducts thus the need for environmental, nontoxic synthetic protocols for nanoparticles synthesis. In the global efforts to reduce generated hazardous waste, “green” chemistry and chemical processes are progressively integrating with modern development in science and industry ( *Sharma et al., 2009*) leading to the developing interest in biological approaches which are free from the use of toxic chemicals as by products. Biological methods can be used to synthesize nanoparticles without the use of any harsh, toxic and expensive chemical substances. The bioreduction of metal ions by combinations of biomolecules found in the extracts of certain organisms (e.g., enzymes/proteins, yeast, fungi, bacteria and plants) is environmentally benign, yet chemically complex (*Ankamwar et al., 2005*). It has been elucidated that biomolecules with carbonyl, hydroxyl, and amine functional groups have the potential for metal ion reduction and capping of the newly formed particles during their growth processes (*Harekrishna et al., 2009, He et al., 2007*). Biomolecules in plants and spices extract are essential oils (terpenes, eugenols, e.t.c.), polyphenols, carbohydrates, e.t.c. and can reduce and stabilize $Ag^+$ to $Ag^0$. It provides advancement over chemical and physical methods as it is cost effective and environment friendly.

## 1.1 LITERATURE REVIEW

Disease-causing microbes are becoming resistant to drug therapy and therefore poses great public health problem. Many researchers are now engaged in developing new effective antimicrobial reagents with the emergence and increase of microbial organisms resistant to multiple antibiotics, which will increase the cost of health care. Colloidal silver has been known for a long time to possess antimicrobial properties and also to be non-toxic and environmentally friendly. It has been used for years in the medical field for antimicrobial applications such as burn treatment (*Parikh et al. 2005; Ulkur et al 2005*), elimination of microorganisms on textile fabrics (*Jeong et al. 2005; Lee et al. 2007; Yuranova et al. 2003*), disinfection in water treatment (*Russell and Hugo 1994; Chou et al. 2005*), prevention of bacteria colonization on catheters (*Samuel and Guggenbichler 2004; Alt et al. 2004; Rupp et al. 2004*), etc. It has also been found to prevent HIV from binding to host cells (*Sun et al. 2005*). The mechanism of the bacterial effect of AgNP as proposed is due to the attachment of AgNPs to the surface of the cell membrane, thus disrupting permeability and respiration functions of the cell (*Kevitec et al. 2008*). It is also proposed that AgNPs not only interact with the surface of a membrane but can also penetrate inside the bacteria (*Morones et al. 2005*), but the effects of silver nanoparticles (AgNP) on microorganisms have not been developed fully. Researchers believe that the potential of colloidal silver is just beginning to be discovered (*Dorjnamjin et al., 2008*).

## 1.2 Nanotechnology

Nanoparticles are viewed as the fundamental building blocks of nanotechnology (*Mansoori et al., 2005*). They are the starting points for preparing many nanostructured materials and devices and their synthesis is an important component of the rapidly growing research efforts in nanoscience and nanoengineering (*Mansoori et al., 2007*).

In nanotechnology, a nanoparticle is defined as a small object that behaves as a whole unit in terms of its transport and properties. Nanoparticles can equally be called ultrafine particles since their sizes range from 1 to 100 nm. Fine particles ranges from 100 to 2,500 nm, while coarse particles are sized between 2,500 and 10,000 nm (*Williams, 2008*). A nanometer is one billionth of a meter ($10^{-9}$ m), roughly the width of three or four atoms, smaller than the wavelength of visible light and a hundred-thousand the width of human hair.

Nanoparticles can be made of materials of diverse chemical nature, the most common being metals, metal oxides, silicates, non-oxide ceramics, polymers, organics, carbon and biomolecules. Nanoparticles exist in several different morphologies such as spheres, cylinders, platelets, tubes, flowers, cubes etc. They possess unique physiochemical, optical and biological properties which can be manipulated to suit a desired application. Nanoparticles are of great interest due to their externally small size, and large surface to volume ratio. They exihibit utterly novel characteristics compared to the large particles of the bulk material and have been included in fields of science as diverse as surface science, organic chemistry molecular biology, semi conductor physics, microfabrication, material science, inorganic chemistry and so on.

The concepts that seeded nanotechnology were first discussed in 1959 by renowned physicist Richard Feynman in his talk “There's Plenty of Room at the Bottom”, in which he described the possibility of synthesis via direct manipulation of atoms. In 1974, “Norio Taniguchi now used the word nanotechnology to describe precision manufacturing materials at the nanometer level which refers to the synthesis, manipulation, and control of matter at nano dimensions that will make most products lighter, stronger, cleaner, less expensive and more precise.

## 1.3 Physiochemical Properties of Nanoparticles

Nanoparticles also often possess unexpected optical properties as they are small enough confine their electrons and produce quantum effects e.g. gold nanoparticles appear deep red in dark solutions.

A unique property among nanoparticles is quantum confinement in semiconductor particles, surface plasmon resonance in some metal particles and super paramagnetism in magnetic materials. For example, ferroelectric materials smaller than 10 nm can switch their magnetization direction using room temperature thermal energy. Thus this property is not always desired in nanoparticles thus making them unsuitable for memory storage.

Suspensions of nanoparticles are possible since the interaction of the particle surface with the solvent is strong enough to overcome density differences, which otherwise usually result in a material either sinking or floating in a liquid.

The high surface area to volume ratio of nanoparticles provides a tremendous driving force for diffusion, especially at elevated temperatures. Sintering can take place at lower temperatures, over shorter time scales than for larger particles.

## 1.4 Methods of Nanoparticles Synthesis

Currently, many methods have been reported for the synthesis of nanoparticles which include chemical, physical, biological and photo-induced approach.

### 1.4.1 Chemical Approach:

The chemical approach is the most used method since it for provides an easy way to synthesize nanoparticles in solution. This consists of the chemical reduction of a metal salt in solution followed by the crystallization of zero-valence metal particles. The particle synthesis is usually conducted in the presence of a stabilizing agent that prevents excessive molecular growth and/or aggregation of the metal nanoparticles. Hence when nanoparticles are produced by chemical synthesis, three main components are needed: a salt (e.g. $AgNO_3$), a reducing agent (e.g. ethylene glycol) and a stabilizer agent (e.g. PVP) to control the growth of the nanoparticles and prevent them from aggregating.

In one study, Oliveira and coworkers (2005) prepared dodecanethiol-capped silver NPs, according to Brust procedure (Brust et al., 2002) based on a phase transfer of an $Au^{3+}$ complex from aqueous to organic phase in a two-phase liquid-liquid system, which was followed by a reduction with sodium borohydride in the presence of dodecanethiol as stabilizing agent, binding onto the NPs surfaces, avoiding their aggregation and making them soluble in certain solvents. They reported that small changes in synthetic factors lead to dramatic modifications in nanoparticle structure, average size, size distribution width, stability and self-assembly patterns.

### 1.4.2 Physical Approach:

In physical processes, nanoparticles are synthesized by evaporation-condensation, exploding wire technique, chemical vapour deposition, microwave irradiation, pulsed laser ablation, supercritical fluids, sonochemical reduction, and gamma radiation with evaporation-condensation and laser ablation being the most important physical approaches. The absence of solvent contamination in the prepared thin films and the uniformity of NPs distribution are the advantages of physical synthesis methods in comparison with chemical processes.

Siegel and colleagues (2012) demonstrated the synthesis of AgNPs by direct metal sputtering into the liquid medium. The method, combining physical deposition of metal into propane-1, 2, 3-triol (glycerol), provides an interesting alternative to time-consuming, wet-based chemical synthesis techniques. Silver NPs possess round shape with average diameter of about 3.5 nm with standard deviation 2.4 nm. It was observed that the NPs size distribution and uniform particle dispersion remains unchanged for diluted aqueous solutions up to glycerol-to-water ratio 1 : 20.

### 1.4.3 Biological Approach:

As stated earlier in the chemical method of synthesis, three main components are needed: a salt (e.g. $AgNO_3$), a reducing agent (e.g. ethylene glycol) and a stabilizer agent (e.g. PVP) to control the growth of the nanoparticles and prevent them from aggregating. In biological synthesis of nanoparticles, the reducing agent and the stabilizer are replaced by molecules produced by living organisms. These reducing and/or stabilizing compounds can be utilized from bacteria, fungi, yeasts, algae or plants.

The development of efficient green chemistry methods for synthesis of nanoparticles has become a major focus of researchers. In the global effort to reduce generated waste and toxic materials, “green” chemistry and chemical processes are progressively integrating with modern developments in science and industry. They have investigated in order to find an eco-friendly technique for production of well-characterized nanoparticles. Various approaches using plant extracts have been used for the synthesis of nanoparticles. These approaches have many advantages over chemical, physical, and microbial synthesis because there is no need of the elaborate process of culturing and maintaining the cell, using hazardous chemicals, high-energy and wasteful purifications.

The first successfully reported synthesis of nanoparticles assisted by living plants appeared in 2002 when it was shown that gold nanoparticles, ranging in size from 2-20 nm, could form inside alfalfa seedlings. Subsequently it was shown that alfalfa could form silver nanoparticles when exposed to a silver rich medium. Other works on plants and plant parts that have been used for the synthesis of silver nanoparticles are *Thevetia peruviana* latex (*Rupiasih et al. 2013*), *Wrightia tinctoria* (*Bharani et al. 2011*), *Solanum xanthocarpum* (*Muhammad et al. 2012*), *Opuntia ficus* (*Silva-de-Hoyos et al. 2012*), *Sphaeranthus amaranthoides* (*Swarnalatha et al. 2012*), *Punica granatum* (*Naheed et al. 2012*) *Citrullus colocynthis* ( *Satyavani et al. 2011*), *Eucalyptus chapmaniana* (*Ghassan et al. 2013*), *Acacia auriculiformis* (*Nalawade et al. 2014*), *Ficus benghalensis*, *Azadirachta indica* (*Debasis et al. 2015*), e.t.c.

The biomolecules present in these plants are responsible for the formation and stabilization of silver nanoparticles (*Iravani et al. 2014*). Nanoparticles produced by plants are more stable and the rate of synthesis is faster than in the case of microorganisms. Moreover, this method is simple, cost effective, energy-saving and reproducible. The nanoparticles are more various in shape and size in comparison with those produced by other organisms. The advantages of using plant and plant-derived materials for biosynthesis of metal nanoparticles have interested researchers to investigate mechanisms of metal ions uptake and bioreduction by plants, and to understand the possible mechanism of metal nanoparticle formation in plants.

### 1.4.4 Photo-induced Approach:

The photo-induced synthetic strategies can be categorized into two distinct approaches, that is the photo-physical (top down) and photochemical (bottom up) ones. The former could prepare the NPs via the subdivision of bulk metals and the latter generates the NPs from ionic precursors. The NPs are formed by the direct photo-reduction of a metal source or reduction of metal ions using photo-chemically generated intermediates, such as excited molecules and radicals which are known as photosensitization in the synthesis of NPs.

Huang and coworkers (2008) reported the synthesis of silver NPs in an alkaline aqueous solution of AgNO3/carboxymethylated chitosan (CMCTS) using UV light irradiation. CMCTS, a watersoluble and biocompatible chitosan derivative, served simultaneously as a reducing agent for silver cation and a stabilizing agent for the silver NPs. The diameter range of produced silver NPs was 2–8 nm, and they can be dispersed stably in the alkaline CMCTS solution for more than 6 months

The main advantages of the photochemical synthesis are;

- It is a clean process, with high spatial resolution, and convenience of use.
- It has great versatility; the photochemical synthesis enables one to fabricate the NPs in various mediums including emulsion, surfactant micelles, polymer films, glasses, cells, etc.

## 1.5 Applications of Nanoparticles

There is wide applicability of nanoparticles due to their interesting optical, conductive, physio-chemical, electronic, antimicrobial properties.

### 1.5.1 Medical and Pharmaceutical Applications

Nanoparticles can be made to control and sustain release of the drug during the transportation and as well as the location of the release since the distribution and subsequent clearance of the drug from the body can be altered. An increase in the therapeutic efficacy and reduction in the side effects can also be achieved. Targeted drugs may be developed.

The surface change of protein filled nanoparticles has been shown to affect the ability of the nanoparticles to stimulate immune responses. Researchers are thinking that these nanoparticles may be used in inhalable vaccines. Researchers are developing ways to use carbon nanoparticles called nanodiamonds in medical applications. For example, nanodiamonds with protein molecules attached can be used to increase bone growth around dental or joint implants.

Other medical and pharmaceutical applications include; tissue engineering, bio detection of pathogens, tumour destruction via heating (hyperthermia), drug and gene delivery, separation and purification of biological molecules and cells.

### 1.5.2 Biosensing

A biosensor is an analytical device used for the detection of an analyte, which combines a biological component with a physicochemical detector (*Florinel-Gabriel, 2012*). The sensitive biological element can be tissues, microorganisms, organelles, cell receptors, enzymes, antibodies, nucleic acids, e.t.c. or a biologically derived material or biomimetic component that interacts, binds or recognizes the analyte under study. Nanomaterials are exquisitely sensitive chemical and biological sensors. Their large surface area to volume ratio can achieve rapid and low cost reactions, using a variety of designs (*Gerald, 2009*).

Biosensing can have the following applications;

- Environmental applications e.g. the detection of pesticides and river water contaminants such as heavy metal ions.
- Determining the presence of pathogen and food toxins in food analysis.
- Determining levels of toxic substances before and after bioremediation.
- Determination of drug residues in food, such as antibiotics and growth promoters.
- Drug discovery and evaluation of biological activity of new compounds.

### 1.5.3 Optical Applications

The optical properties of noble metals nanoparticles have been of great interest because of many applications in optical devices (optical limiters, solar cells, medicals imaging, surface enhanced spectroscopy, surface plasmonic devices) and bio-applications (*Haglund et al. 1993*).

### 1.5.4 Optoelectronics

Optoelectronics is the study and application of electronic devices that source, detect and control light. The light includes invisible forms of radiation such as gamma rays, X-rays, ultraviolet and infrared, in addition to visible light. Optoelectronic devices are electrical-to-optical or optical-to-electrical transducers, or instruments that use such devices in their operation. It can function as an emitter of optical radiation, such as a light-emitting diode (LED), or as a photovoltaic (PV) device that can be used to convert optical radiation into electrical current, such as a photovoltaic solar cell.

In optoelectronics;

Nanoparticles can be applied in the production of optocouplers, a component that transfers electrical signals between two isolated circuits by using light. They prevent high voltages from affecting the system receiving the signal.

Nanoparticles are also applied in optical fibers which are used most to transmit light between the two ends of the fiber and find wide usage in fiber-optic communications. They are also used for illumination, and are wrapped in bundles so that they may be used to carry images, thus allowing viewing in confined spaces e.g. fiberscope.

### 1.5.5 Energy and Electronic Applications

**Quantum Dots;**

A quantum dot (QD) is a nanocrystal made of semiconductor materials that is small enough to exhibit quantum mechanical properties. Specifically, its excitons are confined in all three spatial dimensions. The electronic properties of these materials are intermediate between those of bulk semiconductors and of discrete molecules (*Brus, 2007, Norris, 1995, Murray et al., 2000*).

Quantum dots are applied in;

In textile technology, various kinds of organic dyes are used but more flexibility is being required of these dyes, and the traditional dyes are often unable to meet the expectations (*Walling et al., 2009*). To this end, Quantum dots have quickly filled in the role, found to be superior to traditional organic dyes on several counts. One of the most immediately obvious being brightness (owing to the high extinction co-efficient combined with a comparable quantum yield to fluorescent dyes (*Michalet et al., 2005*) as well as their stability (allowing much less photobleaching).

Also in biology, the usage of quantum dots for highly sensitive cellular imaging has seen major advances over the past decade (*Spie., 2014*). Another application that takes advantage of the extraordinary photostability of quantum dot probes is the real-time tracking of molecules and cells over extended periods of time (*Dahan et al., 2003*).

In light emitting devices, because Quantum dots naturally produce monochromatic light; they can be more efficient than light sources which must be color filtered. They are used to improve existing light-emitting diode (LED) design.

### 1.5.6 Antibacterial Applications

Silver nanoparticles serves as an inorganic antibacterial powder and play a critical role in the suppression and killing of pathogenic microorganisms such as *S. aureus*, *E. coli*, etc. This innovative anti-infective products has broad spectrum, non-resistance, durable, has a non-oxidized appearance and is unaffected by pH effects. Ag-Nps are incorporated in apparel, foot wears, paints, wound dressings, appliances, cosmetics and plastics for their antibacterial properties. The colloidal silver is capable of disinfecting water through sterilization.

### 1.5.7 Other Applications of Nanoparticles

Generally, nanoparticles are used or being evaluated for use, in many fields. The list below introduces several of the other uses under development. They include;

**Applications in Manufacturing and Materials:** Titanium dioxide and zinc oxide nanoparticles are commonly used in sunscreen, cosmetics and some food products while silver nanoparticles are used in food packaging, clothing, disinfectants and household appliances. Nano silver and carbon nanotubes are used for stain-resistant textiles; and cerium oxide as a fuel catalyst. Zinc oxides nanoparticles can be dispersed in industrial coating to prevent wood, plastic and textile from exposure to UV rays.

**Applications in Water Purification:** Nanotechnology is also being applied to or developed for application to a variety of industrial purification processes. Purification and environmental cleanup applications include the desalination of water, water filtration, wastewater treatment, groundwater treatment, and other nanoremediation.

**Applications in the Environment:** Researchers are using photocatalytic copper tungsten oxides nanoparticles to break down oil into biodegradable compounds. The nanoparticles are in a grid that provides high surface area for the reaction. It is activated by sunlight and can work in water, making them useful for cleaning up oil spills.

## 1.6 Silver Metal

Silver is a chemical element with symbol Ag (Latin name; argentum). It has its electronic configuration as [Kr] $4d^{10}$ $5s^{1}$ (no. of electron per shell; 2, 8, 18, 18, 1) and has an atomic number 47. It is very ductile, malleable metal (slightly less so than gold), with a brilliant white metallic luster that can take a high degree of polish. The electrical conductivity of silver is the highest of all metals, even higher than copper. Pure silver has the highest thermal conductivity of any metal.

The most common oxidation state of silver is +1 (e.g. silver nitrate, $AgNO_3$), other oxidation states include; +2 compounds (e.g. silver (II) fluoride, $AgF_2$), +3 (e.g. potassium tetrafluoroargentate (III), $KAgF_4$) and even +4 compounds (e.g. potassium hexafluoroargentate (IV), $K_2AgF_6$) (*Riedel et al., 2009*).

Silver is found in a native form as an alloy with gold (electrum), and in ores containing sulphur, arsenic, antimony or chlorine. Some ores include; argentite ($Ag_2S$), chlorargyrite (AgCl) and pyrargyrite ($Ag_3SbS_3$). The metal is primarily produced as a byproduct of electrolytic copper refining, gold, nickel, and zinc refining. Naturally occurring silver is composed of two stable isotopes, $^{107}$ Ag and $^{109}$ Ag, with $^{107}$ Ag being slightly more abundant (51.8% natural abundance).

### 1.6.1 Some Uses of Silver

Silver is used to make solder and brazing alloys, and as a thin layer on bearing surfaces can provide a significant increase in resistance and reduce wear under heavy load, particularly against steel. It is used in photography, in the form of silver nitrate and silver halides, for the development of coloured films. Some electrical and electronic products use silver for its superior conductivity, even when tarnished. Small devices, such as hearing aids and watches, commonly use silver oxide batteries due to their long life and high energy-to-weight ratio. Silver, in the form of electrum (a gold–silver alloy), was coined to produce money. Silver coins and bullion are also used as an investment to guard against inflation and devaluation. Silver salts have been used since the middle ages to produce a yellow or orange color to stain glass. Using a process called sputtering, silver, along with other optically transparent layers, is applied to glass, creating low emissivity coatings used in high-performance insulated glazing. Silver can be alloyed with mercury at room temperature to make amalgams that are widely used for dental fillings. Silver and silver alloys are used in the construction of high-quality musical wind instruments of many types. Flutes, in particular, are commonly constructed of silver alloy or silver plated both for appearance and for the frictional surface properties of silver. Brass instruments, such as trumpets and baritones, are also commonly plated in silver.

## ANTIBACTERIAL EVALUATION OF AFANG LEAF EXTRACT AND ITS SYNTHESIZED SILVER NANOPARTICLES