GREEN SYNTHESIS OF SILVER NANOPARTICLES FROM URTICA DIOICA (STINGING NETTEL) LEAVES

Project Thesis submitted in fulfillment of major project of

BACHELORS OF TECHNOLOGY

IN

BIOTECHNOLOGY

By

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UNDER THE SUPERVISION OF

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MAY-2022

DEPARTMENT OF BIOTECHNOLOGY AND BIOINFORMATICS

JAYPEE UNIVERSITY OF INFORMATION TECHNOLOGY WAKNAGHAT,

SOLAN, HIMACHAL PRADESH - 173234

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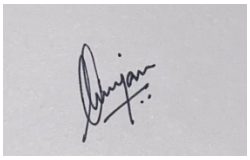
DECLARATION

We hereby declare that the major project work entitled "**Green synthesis of silver nanoparticles from urtica dioica leaves**" has been solely submitted to the Department of Biotechnology and Bioinformatics, Jaypee University of Information Technology, Waknaghat have carried out under guidance of our supervisor **Dr. Abhishek Chaudhary.**



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SUPERVISOR'S CERTIFICATE

This is to certify that the major project work titled "**Green synthesis of silver nanoparticles from urtica dioica leaves**" by **Gunjan** during their 8th semester in May 2022in fulfillment for the project thesis in Biotechnology of Jaypee University of Information Technology, Solan has been carried out under my supervision. This work has not been submitted partially to any other University or Institute for the award of any degree or appreciation.

Signature of Supervisor

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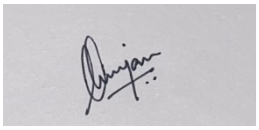
ACKNOWLEDGEMENT

I take this opportunity to express our first and foremost gratitude to our "DEPARTMENT OF BIOTECHNOLOGY AND BIOINFORMATICS" for the confidence bestowed upon me and entrusting my project titled "**Green synthesis of silver nanoparticles from urtica dioica leaves**"

At this juncture, with proud privilege and profound sense of gratitude, I feel honored in expressing my deepest appreciation to **Dr. Abhishek Chaudhary,** for being a lot more than just a supervisor and going beyond the call of duty in my guidance, support, advice, and motivation throughout. He has been the source of inspiration of come what may; these issues cannot bring you down. Sincere thanks for his insightful advice, motivating suggestions, invaluable guidance, help and support in successful completion of this major project and also for his constant encouragement and advice throughout my project work.

Special thanks to my parents for their infinite patience and understanding for the constant support and most importantly God, who in his mysterious ways, always made things work out in the end.

In gratitude,



GUNJAN (181835)

LIST OF FIGURES:

ABSTRACT

Nanotechnology is the science and study of nanoparticles and material on which we are studying at nano scale say about 10^{-9} m. Nano biotechnology or nano medicine is vast field and is used in almost every aspect of life. Green synthesis of nanoparticles simply means that the extraction or formation of nanoparticles without any harmful or any toxic gases releasing into the atmosphere or harming any water bodies or environment.

In biomedical field, mostly used nanoparticles are silver (AgNPs), copper ($CuSO_4$), palladium (Pd- NPs) and gold (Au-NPs). These are widely used among all others. Nanoparticles can be extracted of desired shape and size that is required for our experiment and for applications like optical or medicinal etc.

Nanoparticles have got wide attention now a days because of its properties. They are small in size but have large surface area to volume ratio which makes it open to facilitate chemical processes and create new materials. Because of this property they act as best catalyst.

Nowadays, Nanotechnology plus green synthesis is so much flourishing and has gained a lot more attention from scientists and researchers because for their non-toxic behavior, clean and easy processing, cost effective, simple technique and ecofriendly. There have been many research articles focusing on the use of many kinds of plant extracts as a reducing agent for synthesis of nanoparticles; especially silver nanoparticles (AgNPs).

This green synthesis approach requires biological entities like plants (their leaves or barks), algae, fungi etc are used as reducing and capping agents. Metal nanoparticles are used because of their exceptional physical and chemical properties for these biological entities such as optical behavior, antibacterial activity, electrical conductivity and biocompatibility.

Keyword: Urtica Dioica, Green Synthesis, Silver Nano-particles, Nanotechnology.

CHAPTER 1

INTRODUCTION

[1.1]

NANOPARTICLES:

Nanoparticles are the particles which are 10^{-9} m small. These particles are so small that they cannot be seen with naked eyes. They can be seen and characterized through transmission electron microscopy. A nanometer (nm) is an International System of Units (System worldwide unites, SI) unit that addresses 10−9 meter in length. On a basic level, NMs are portrayed as materials with length of 1-1000 nm in no less than one aspect; in any case, they are commonly characterized to be of distance across in the scope of 1 to 100 nm. Today, there are a few bits of regulation in the European Association (EU) and USA with explicit references to NMs. However, a solitary globally acknowledged definition for NMs doesn't exist. Various associations have a distinction in assessment in characterizing NMs [1]. As per the Environmental Protection Office (EPA), "NMs can show remarkable properties divergent than the same substance compound in a bigger aspect". The US Food and Drug Administration (USFDA) moreover alludes to NMs as "materials that have somewhere around one aspect in the scope of around 1 to 100 nm and display aspect subordinate peculiarities".

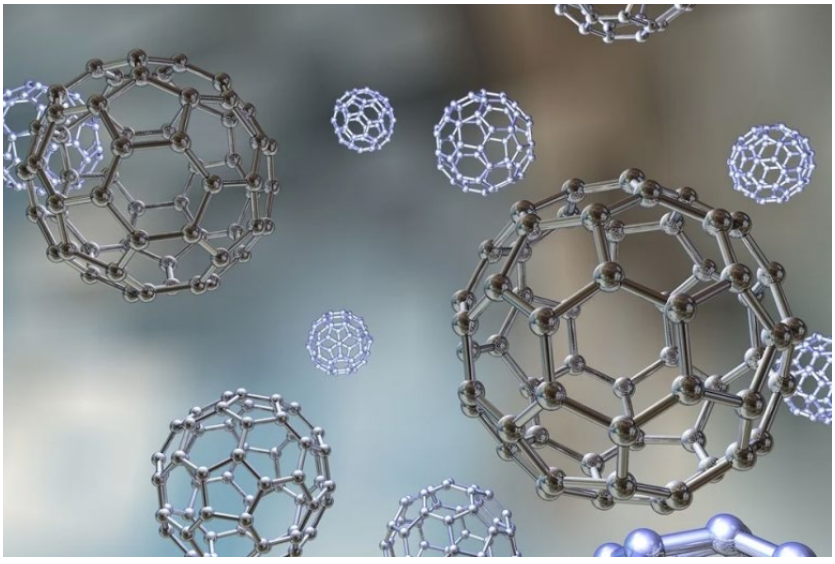


Fig 1: Nanoparticle [36]

[1.2]

SILVER NANOPARTICLES:

Now coming to silver nanoparticles, they are known for their antibacterial and antimicrobial activity over a wide spectrum. They are also known for their adjustable size and shape, high – density surface ligand. They have many advantages such as they are used as drug carriers; they have adjustable shape and size, and protection from attached therapeutics etc. Besides the advantages the disadvantage of silver is its toxicity. Many researches are going on to decrease the toxicity of silver nanoparticles and their adverse effects like discoloration of the skin but here we are not facing any such issue because we are working with the bacteria and their antibacterial activity of silver and not with drugs or not with the drugs for human consumption.

However, in this research we aimed to study the synthesis of AgNPs using a new kind of herbal medicinal plant extract called *Urtica Dioica* as a reducing and stabilizing agent to observe the stability of synthesized AgNPs. Further, antibacterial activity has been studied to observe toward bacerias of these stabilized AgNPs.

[1.3]

URTICA DIOICA:

Urtica Dioica which is also known as stinging nettle/common nettle or in laymen language we call it 'Bicchu butti'. It is basically an herb (medicinal herb) since long time and are found among worldwide especially in Europe, North America, North Africa and parts of Asia such as Himachal Pradesh. It is a kind of medicinal plant and has so many medicinal properties and in Uttrakhand people eat this. They cook the leaves and eat as potherb. People of Uttrakhand eat this in the form of 'saag'. This is called nettle because of the thorns present on it, the leaves and stems; the whole plant is covered with thorns.

The edible portion of this plant is full of phytochemicals (sterols, glycosides, alkaloids, phenolic acids etc.) and antioxidants.

As we know that humans have developed resistance against so many bacteria so it is quite tough to analysesdifferent bacteria for this. Therefore, this topic was taken so that the nanoparticle that was extracted was through green synthesis method and their antibacterial activity and synergistic effects with antibiotics against a wide group of pathogenic bacteria.

Fig 2: Urtica Dioica Plant [37]

CHAPTER 2

REVIEW OF LITERATURE

[2.1]

NANOPARTICLES:

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[2.2]

TYPES OF NANOPARTICLES:

Nanoparticles can be classified on the basis of their size, shape, physical and chemical properties and morphology etc. They are divided into 2 types- Organic and Inorganic nanoparticles.

[2.2.1] Organic Nanoparticles:

Dendrimers, micelles, liposomes and ferritin, and so on are generally knows the natural nanoparticles or polymers. These nanoparticles are biodegradable, non-poisonous, and a few particles like micelles and deeply , otherwise called Nano capsules and are delicate to warm and electromagnetic radiation, for example, intensity and light [5]. These interesting qualities make them an ideal decision for drug conveyance. The medication conveying limit, its steadiness and

conveyance frameworks, by the same token entangled drug or adsorbed drug framework decides their field of utilizations and their effectiveness aside from their ordinary attributes like the size, organization, surface morphology, and so forth. The natural nanoparticles are most broadly utilized in the biomedical field for instance drug conveyance framework as they are effective and furthermore can be infused on unambiguous pieces of the body that is otherwise called designated drug conveyance.

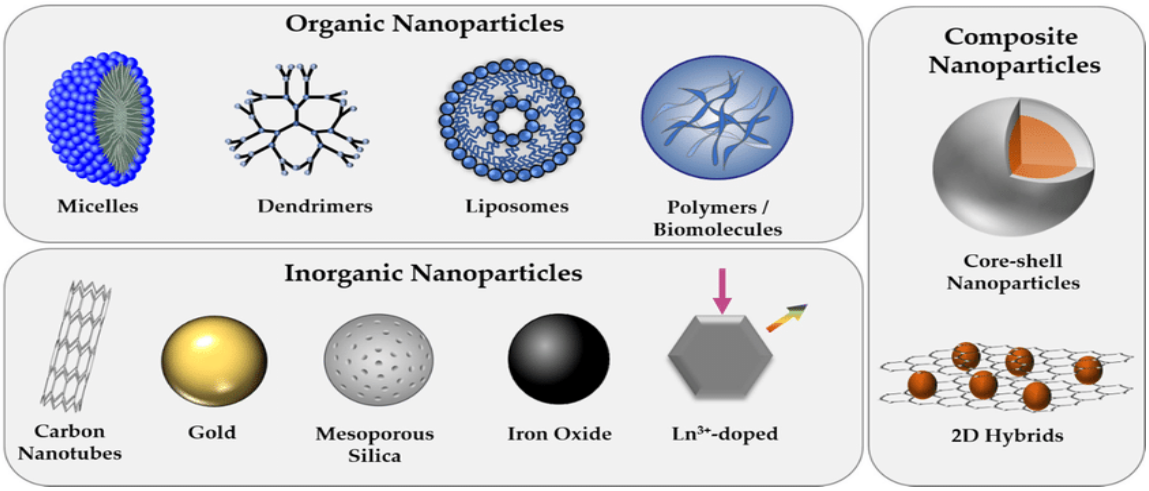

Fig 3: Organic and inorganic nanoparticles [38]

[2.2.2]Inorganic Nanoparticles:

In inorganic nanoparticles the classification is on the basis of their metal based, metal oxides based and carbon based.

- **Metal based nanoparticles:**

  Metal based nanoparticles are those nanoparticles that can be synthesized from metal such as silver, copper, palladium etc. Almost every metal can be synthesized to nanoparticles with different methods. All silver, palladium, gold, silver, copper can be produced by reduction and in that process they are called reducing or capping agents. They large surface area to volume ratio which makes them the ideal choice for selection. They have size that ranges from 10-100nm. They have crystalline, amorphous, cylindrical properties.

- **Metal oxide based nanoparticles:**

  Metal oxide nanoparticles are actually the advanced form of nanoparticles that are synthesized from the metal only. These are synthesized because they have high reactivity than normal nanoparticles. They are generally made from metal by oxidation in the presence of oxygen which increases its reactivity and efficacy. For example: aluminum is oxidized and formed as Al_2O_3 and iron is oxidized and formed as Fe_2O_3.

- **Carbon based nanoparticles:**

  These nanoparticles are made completely from carbon that's why they are carbon based. For example: fullerene, carbon black, carbon nanotubes, grapheme and carbon Nano fiber etc.

- **Fullerenes:**

  A fullerene (C60) is a carbon particle that is circular in shape and comprised of carbon molecules kept intact by sp2 hybridization. Around 28 to 1500 carbon particles shapes the round structure with breadths up to 8.2 nm for a solitary layer and 4 to 36 nm for complex fullerenes.

- **Carbon Nano tubes (CNT):**

  Carbon Nano Tubes (CNT), a graphene Nano foil with a honeycomb cross section of carbon iotas is twisted into empty chambers to frame nanotubes of breadths as low as 0.7 nm for a solitary layered and 100 nm for multifaceted CNT and length changing from a couple of micrometers to a few millimeters. The closures can either be empty or shut by a half fullerene atom.

- **Graphene:**

  Graphene is an allotrope of carbon. Graphene is a hexagonal organization of honeycomb cross section comprised of carbon molecules in a two layered planar surface. For the most part the thickness of the graphene sheet is around 1 nm.

- **Carbon black:**

  Carbon Black Nanoparticles is a conductive, non-contaminating powder created through high temperature (1300 °C) carbonization in a painstakingly controlled ignition process. Applications for carbon dark Nano powder incorporate gadgets, plastics, coatings, inks, and green innovation.

- **Carbon Nano fiber:**

  Graphene foil is used in the production of carbon Nano fiber just like carbon Nano tubes. It's just they are formed in the form of cone shape or cup shape and not in cylindrical shape.

[2.3]

SYNTHESIS OF NANOPARTICLES:

Nanoparticles can be synthesized by following two methods:

- **Top down and Bottom up method.**

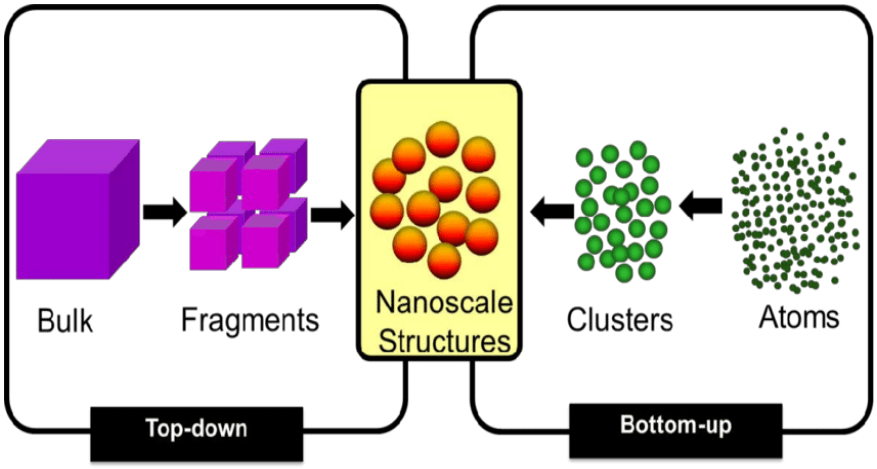

Fig 4: Top down and bottom up approach [39]

[2.3.1] Bottom Up Approach (Constructive Method):

Bottom up method is an approach by which nanoparticles are synthesized from an atom to nanoparticles. This method includes pyrolysis, chemical vapour deposition, spinning, sol-gel etc.

- **Spinning:**

  The combination of nanoparticles by turning is completed by a turning plate reactor (SDR). It contains a turning circle inside a chamber/reactor where the actual boundaries, for example, temperature can be controlled. The reactor is for the most part loaded up with nitrogen or other inactive gases to eliminate oxygen inside and stay away from synthetic responses [7]. The plate is pivoted at various speeds where the fluid for example antecedent and water is siphoned in. The turning makes the iotas or particles meld together and are encouraged, gathered and dried [11]. The different working boundaries, for example, the fluid stream rate,

plate pivot speed, fluid/antecedent proportion, area of feed, circle surface, and so on. Decides the qualities nanoparticles orchestrated from SDR.

- **Pyrolysis:**

  Pyrolysis is the most normally involved process in enterprises for large-scale creation of nanoparticle. It includes consuming a forerunner with fire. The forerunner is either fluid or fume that is taken care of into the heater at high tension through a little opening where it consumes [13]. The burning or side-effect gases are then air characterized to recuperate the nanoparticles. A portion of the heaters use laser and plasma rather than fire to create high temperature for simple vanishing [14]. The benefits of pyrolysis are straightforward, proficient, savvy and ceaseless interaction with high return.

- **Biosynthesis:**

  Biosynthesis is a green and ecological agreeable methodology for the combination of nanoparticles that are nontoxic and biodegradable [15]. Biosynthesis utilizes microscopic organisms, plant extricates, growths, and so on alongside the forerunners to create nanoparticle rather than show synthetic compounds for bio reduction and covering purposes. The biosynthesizednanoparticles havenovel and improved properties that find its direction in biomedical applications [1].

- **Chemical Vapor Deposition:**

  Compound fume testimony is the statement of a slender film of vaporous reactants onto a substrate. The statement is completed in a response chamber at surrounding temperature by consolidating gas atoms. A synthetic response happens when a warmed substrate interacts with the consolidated gas [8]. This response delivers a slim film of item on the substrate surface that is recuperated and utilized. Substrate temperature is the impacting factor in CVD. The benefits of CVD are exceptionally unadulterated, uniform, hard and solid nanoparticles. The detriments of CVD are the necessity of unique gear and the vaporous results are profoundly poisonous [12].

- **Sol- Gel:**

  The sol is a colloidal arrangement of solids suspended in a fluid stage. The gel is a strong macromolecule lowered in a dissolvable. Sol-gel is the most favored base up strategy because of its straightforwardness and as the majority of the nanoparticles can be integrated from this technique. It is a wet-compound interaction containing a substance arrangement going about as an antecedent for a coordinated arrangement of discrete particles. Metal oxides and chlorides are the ordinarily involved forerunners in sol-gel process [9]. The forerunner is then scattered in a host fluid either by shaking, blending or sonication and the resultant framework contains a fluid and a strong stage. A stage detachment is completed to recuperate the nanoparticles by different techniques, for example, sedimentation, filtration and centrifugation and the dampness is additionally eliminated by drying [10].

[2.3.2]Top – Down Approach (Destructive Method):

Top – down method is an approach where the bulk material is converted into Nano metric scale. This is a destructive method. This method includes mechanical milling, laser ablation, nanolithography, sputtering and thermal decomposition.

- **Nanolithography:**

  Nanolithography is the investigation of manufacturing Nano metric scale structures with at least one aspect in the size scope of 1 to 100 nm. There are different Nano lithographic processes for example optical, electron-pillar, multiphoton, Nano imprint and examining test lithography [17]. For the most part lithography is the method involved with printing a necessary shape or construction on a light touchy material that specifically eliminates a piece of material to make the ideal shape and design. The fundamental benefits of nanolithography are to create from a solitary nanoparticle to a bunch with wanted shape and size. The detriments are the necessity of intricate gear and the expense related [18].

- **Laser Ablation:**

  Laser Ablation Synthesis in Solution (LASiS) is a typical technique for nanoparticle creation from different solvents. The light of a metal lowered in a fluid arrangement by a laser

barconsolidates a plasma tuft that produces nanoparticles [19]. A dependable hierarchical strategy gives an elective answer for customary compound decrease of metals to amalgamation metal based nanoparticles. As LASiS gives a steady blend of nanoparticles in natural solvents and water that requires no settling specialist or synthetic compounds it is a 'green' process.

- **Sputtering:**

  Faltering is the statement of nanoparticles on a surface by launching particles from it by crashing into particles [20]. Faltering is typically a testimony of slight layer of nanoparticles followed by tempering. The thickness of the layer, temperature and length of strengthening, substrate type, and so forth decides the shape and size of the nanoparticles [21].

- **Mechanical Milling:**

  Among the different hierarchical strategies, mechanical processing is the most broadly used to create different nanoparticles. The mechanical processing is utilized for processing and post tempering of nanoparticles during combination where various components are processed in an inactive environment [16]. The affecting elements in mechanical processing are plastic distortion that prompts molecule shape, crack prompts decline in molecule size and cold-welding prompts expansion in molecule size.

- **Thermal Decomposition:**

  Thermal deterioration is an endothermic synthetic disintegration delivered by heat that breaks the substance bonds in the compound [6]. The particular temperature at which a component artificially deteriorates is the disintegration temperature. The nanoparticles are created by deteriorating the metal at explicit temperatures going through a substance response delivering optional items.

[2.4]

PROPERTIES:

Nanoparticles exhibit both physical and chemical properties.

[2.4.1] Physical Properties:

The physical properties incorporate optical, for example, the shade of the nanoparticle, its light infiltration, assimilation and reflection capacities, and UV ingestion and reflection capacities in an answer or when covered onto a surface. It likewise incorporates the mechanical properties like versatile, bendable, rigid qualities and adaptability that play a huge element in their application. Different properties like hydrophilicity, hydrophobicity, suspension, dissemination and settling attributes has tracked down its direction in numerous cutting edge ordinary things. Attractive and electrical properties, for example, conductivity, semi conductivity and resistivity have driven a way for the nanoparticles to be utilized in current gadgets warm conductivity in sustainable power applications.

[2.4.2] Chemical Properties:

The compound properties, for example, the reactivity of the nanoparticles with the objective and security and aversion to elements, for example, dampness, environment, intensity and light decide its applications. The counter bacterial, hostile to parasitic, sterilization, and harmfulness, properties of the nanoparticles are great for biomedical and ecological applications. Destructive, hostile to destructive, oxidation, decrease and combustibility attributes of the nanoparticles decide their separate utilization.

[2.5]

CHARACTERISATION:

Characterization of nanoparticles can be done on the basis of size, surface area, composition, surface charge, surface morphology, crystallography and concentration etc.

- **Size:**

  The molecule is one of the most essential and significant estimation for nanoparticle characterization. It decides the size and appropriation of the molecule and whether it falls under Nano or miniature size. The molecule size and dissemination is most generally estimated utilizing electron microscopy. The pictures of Scanning Electron Microscope (SEM) and Transmission Electron Microscope (TEM) are utilized for the estimation of particles and groups while laser diffraction strategies are utilized for estimating mass examples in strong stage [22]. The particles in fluid stage are estimated utilizing photon relationship spectroscopy and centrifugation. The particles in vaporous stage are troublesome and flippant to utilize the imaging strategies and subsequently a Scanning Mobility Particle Sizer (SMPS) is utilized which gives a quick and precise estimations contrasted with different techniques.

- **Composition:**

  The synthetic or essential synthesis decides the immaculateness and execution of the nanoparticle. Presence of higher optional or undesired components in the nanoparticle may diminish its proficiency and furthermore lead to auxiliary response and defilement simultaneously. The organization estimation is normally done by X-beam photoelectron spectroscopy (XPS) [23]. A few strategies include compound processing of the particles followed by wet substance investigation like mass spectrometry, nuclear discharge spectroscopy and particle chromatography. The particles in vaporous stage are gathered either by filtration or electrostatically and spectrometric or wet compound strategies are utilized for the investigation [24].

- **Surface Area:**

  The surface region is additionally a critical component in nanoparticle characterization. The surface region to volume proportion of a nanoparticle impacts its exhibition and properties. The surface region is most regularly estimated utilizing BET investigation. A basic titration is adequate for the surface region investigation of particles in fluid stage, yet it is a work concentrated process. Subsequently atomic attractive reverberation spectroscopy (NMR) is utilized. An altered SMPS and differential versatility analyzer (DMA) is utilized for the estimation of surface are of nanoparticles in vaporous stage.

- **Surface Morphology:**

  The nanoparticles have different shapes and surface designs that assume a critical part in taking advantage of its properties. A portion of the shapes incorporate circular, level, round and hollow, cylindrical, tapered and unpredictable shapes with surface like translucent or undefined with uniform or abnormalities on a superficial level. The surface is by not entirely settled by electron microscopy imaging strategies like SEM and TEM [25]. The particles in fluid stage are stored on a surface and examined while the particles in vaporous stage are catch electrostatically or by filtration for imaging utilizing electron microscopy.

- **Surface Charge:**

  The surface charge of a nanoparticle decides its associations with the objective. By and large a zeta potentiometer is utilized for the estimation of surface charges and its scattering strength in an answer [22]. A Differential Mobility Analyzer (DMA) is utilized for the charge assurance of nanoparticles in vaporous stage.

- **Crystallography:**

  It is the investigation of iotas and particles course of action in gem solids. The crystallography of nanoparticles is done by a powder X-beam, electron or neutron diffraction to decide the primary course of action.

[2.6]

APPLICATIONS

Nanoparticles have applications in almost every field such as cosmetics, food, medicine, electronics, catalysis, construction, renewable energy and environmental remediation etc.

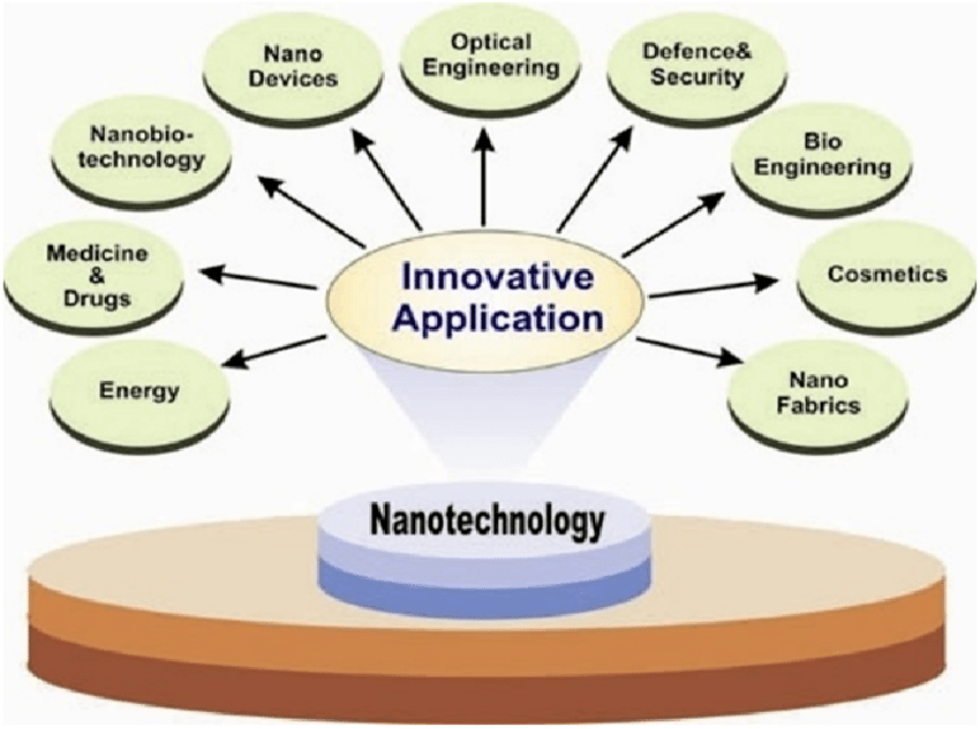

Fig 5: Applications of nanotechnology [40]

- **Cosmetics and sun screens:**

  Sunscreens lack the ability of long term usage that is its effect for long period and nanoparticles in sunscreen provides the same. Basically titanium dioxide is a metal oxide nanoparticle which has a lot of advantages in making sun screens. Zinc oxide is also used because both these play an important role in absorbing and reflecting light. Even some lipsticks are also made out of these such as iron oxide is used as a pigment.

- **Electronics:**

  The higher need for enormous size and high splendor shows as of late that are utilized in the PC screens and TV is empowering the utilization of nanoparticles in the presentation innovation. For instance Nano crystalline lead telluride, cadmium sulfide, zinc selenite and sulfide, are utilized in the light radiating diodes (LED) of present day shows [28]. The improvement in convenient customer hardware, for example, cell phones and PCs to the colossal interest for a conservative, lightweight and high limit batteries. Nanoparticles are the ideal decision for separator plates in batteries. An extensive more energy can be put away contrasted with customary batteries because of their froth like (aerogel) structure. Batteriesproduced using Nano crystalline nickel and metal hydrides, because of their huge surface region require less re-energizing and last longer [29]. The expansion in electrical conductivity of nanoparticles is utilized to distinguish gases like NO_2 and NH_3 [30]. This is because of expansion in the pores of nanoparticles because of charge move from nanoparticles to NO_2 as the gas particles tie them together making them a superior gas sensor.

- **Catalysis:**

  Nanoparticles play an important role in catalysis as they have high surface area to volume ratio. They can be used for catalysis. Platinum nanoparticles are used for catalysis. Some of the reduction experiments are also done using these.

- **Medicine:**

  Nanotechnology has worked on the clinical field by utilization of nanoparticles in drug conveyance. The medication can be conveyed to explicit cells utilizing nanoparticles [32]. The complete medication utilization and secondary effects are fundamentally brought down by setting the medication in the necessary region in required measurement. This technique diminishes the expense and secondary effects. The multiplication and fix of harmed tissue (Tissue designing) can be completed with the assistance nanotechnology. The conventional medicines like counterfeit inserts and organ transfers can be supplanted by tissue designing. One such model is the development of bones carbon nanotube platforms [33]. The utilization

of gold in medication isn't new. In Ayurveda an Indian clinical framework, gold is utilized in a few practices. One normal solution is the utilization of gold for memory upgrade. To upgrade the psychological wellness of child gold is remembered for specific clinical arrangements [34].

- **Food:**

  The improvement underway, handling, security and bundling of food is accomplished by integrating nanotechnology. For instance a Nano composite covering in a food bundling cycle can straightforwardly present the counter microbial substances on the covered film surface [35]. There was one example of canola oil that is used to transfer vitamins and minerals into the food.

- **Construction:**

  Nanotechnology has evolved in last few decades. They have been much of research for construction as well. Evolution is making it quicker, inexpensive, easy etc.For example if Nano silica is mixed with concrete, it can increase its mechanical properties and its durability.

[2.7]

URTICA DIOICA:

Urtica Dioica which is also known as stinging nettle/ common nettle or in layman language we call it 'Bicchu butti'. It is basically an herb (medicinal herb) since long time and are found among worldwide especially in Europe, North America, North Africa and parts of Asia such as Himachal Pradesh. It is a kind of medicinal plant and has so many medicinal properties and in Uttrakhand people eat this. They cook the leaves and eat as potherb. People of Uttrakhand eat this in the form of 'saag'. This is called nettle because of the thorns present on it, the leaves and stems; the whole plant is covered with thorns.

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[2.8]

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stability of synthesized AgNPs. Further, antibacterial activity has been studied to observe toward bacteria of these stabilized AgNPs.

[2.9]

SILVER NITRATE:

It is a chemical compound and has antiseptic properties. Its chemical formula is $AgNO_3$. It is extremely light sensitive and when exposed to light it gets reduced and its structure changes. It is also used to treat skin wounds and burns and acts as anti – infective agent.

The disadvantages of this are if someone is exposed more to it, it causes headache, nausea, dizziness, vomiting, lack of oxygen etc.As of now, there have been awesome endeavors to create clean, non-poisonous, solid and eco-harmless systems for thecombination and get together of nanoparticles with wanted sizes also, morphologies to grow their biomedical applications.

[2.10]

PHYTOSYNTHESIS OF AGNPS

The general response process was done in dim tokeep away from pointless photochemical responses. The variety changeof the AgNO3 arrangement from dry to dim brown was seen by unaided eye and phytodiminished example partwas affirmed by Ultraviolet-noticeable spectroscopy.

[2.11]

PHYTOCHEMICAL SCREENING

There are various methods for phytochemical screening and analysis.

Gas chromatography-mas spectroscopy is a method used for qualitative and quantitative analysis of phytochemicals.

[2.12]

CHARACTERIZATION OF AGNPS

Development of AgNPs was affirmed by Ultraviolet-apparent phantom examination. The absorbance spectra were recordedutilizing Ultraviolet-apparent spectroscopy (UV- 300-800).

The AgNPs were characterised by ultraviolet- visible spectroscopy exhibiting at 300-800nm which was consistant with spectra of AgNPs within the wavelength range mentioned above.

Microscopy methods make the images of nanoparticles individually to characterise their size, shape, morphology etc.

CHAPTER 3

OBJECTIVE

To green synthesis of the silver nanoparticles from Urtica Dioica i.e. Stinging nettle (Common nettle leaf), so that eco-friendly, cost effective nanoparticles are produced.

CHAPTER 4

RATIONALE AND RESEARCH GAP

In addition to all the methods of producing nanoparticles now, there is rapid growing interest in sonochemical and electrochemical methods for synthesizing AgNPs. These methods do not require chemical oxidants which makes them ideal for producing desired pure nanoparticles.

Many research is going on in biological synthesis of silver nanoparticles.

- Synthesis using plant extract
- Synthesis using microorganisms
- Synthesis using biodegradable polymers

Characterization of AgNPs is done using UV-Vis spectroscopy which is a primary and a crucial tool that elaborates the formation os silver nanoparticles at its initial stage. They show their maximum absorbance at 400nm.

Fourier Transform Infrared Spectroscopy (FTIR), it also characterizes nanoparticles and the presence of chemical residue if present on the surface of desired particle. This analysis gives only qualitative information that we need.

X-Ray diffraction is also a method that identifies and is used a fingerprint to identify the material and determine the crystallinity.

Atomic Force Microscopy, it is also an advanced method that is used to characterize nanoparticle topology at nanometer scale.

LATEST RESEARCH ON APPLICATIONS:

- Sensing and therapeutic applications
- Antimicrobial activity
- Sensors
- Pollution degradation
- Water treatment
- Food and agriculture

CHAPTER 5

MATERIAL AND METHODOLOGY

1. PREPARATION OF EXTRACT

(a) Ingredients and Equipment used

- Stinging nettle leaves
- Autoclaved water
- Pestle mortar
- Tray
- Scissors
- Falcon
- Measuring cylinder
- Glass rod
- Beaker
- Weighing balance
- gloves

(b) Procedure

- Fresh leaves were cut from stinging nettle plant very carefully as it has thorns all over it.
- Gloves were put on to avoid the thorns. All the leaves were put into the tray and washed properly so that all the dirt was washed away.
- Then it was once again washed with milli-Q water so that no dirt is remaining.
- It was allowed to dry in shade for three days. Not in sun because many substances will get destroyed. Shade dried for 3 days.
- Now after 3 days when they are completely dried that it can be crushed into fine powder we will cut the stem with scissors and will take the leaves in pestle mortar and crush it in a fine powder. Very fine powder was made so that extraction was easy to done.
- All the crushed powder was weighed and it came out to be around 8 grams.
- Now 2.5 grams was weighed and was taken into the beaker.
- 50ml of autoclaved distilled water was measured using measuring cylinder.
- This water was added to the beaker in which 2.5 grams of powder was taken.
- Mixed it well using glass rod.
- Now placed this mixture into the flask covered it with cotton plug and placed in the (shaker) incubator at 37 degrees for overnight.
- The solution was poured in two tarsons 25 ml each and centrifuged .

- It was centrifuged at 7000 rpm for about 10 minutes.

Fig 6: Dried and Powdered form of Urtica Dioica plant leaves [41]

Fig 7: Crushing dried leaves in pestel mortar [41]

Fig 8: Autoclaved water [41]

Fig 9: Extract was centrifuged [41]

- After taking the supernatant it was centrifuged again same at 7000rpm for 10 minutes.
- Supernatant was collected again.
- Still there were some particles of that powder so it was filtered using whatmann filter paper to remove thode particles and to get a clear extract.
- Finally the extract was made and then it was stored for further use at 4 degree celcius.

2. PREPARATION OF SILVER NITRATE SOLUTION (5Mm) IN 20ml

(a) Ingredients

- Silver nitrate powder
- Beaker
- Aluminum foil
- Tarson
- Weighing balance
- Spatula
- Distilled water

(b) Procedure

- To prepare 5Mm of silver nitrate solution 17 mg of silver nitrate was measured and mixed with 20 ml of distilled water in a beaker.
- Immediately it was transferred into the tarson and covered with aluminum foil because silver nitrate is light sensitive. It reacts with light and forms different compounds.

Fig 10: Silver nitrate (solid form) [41]

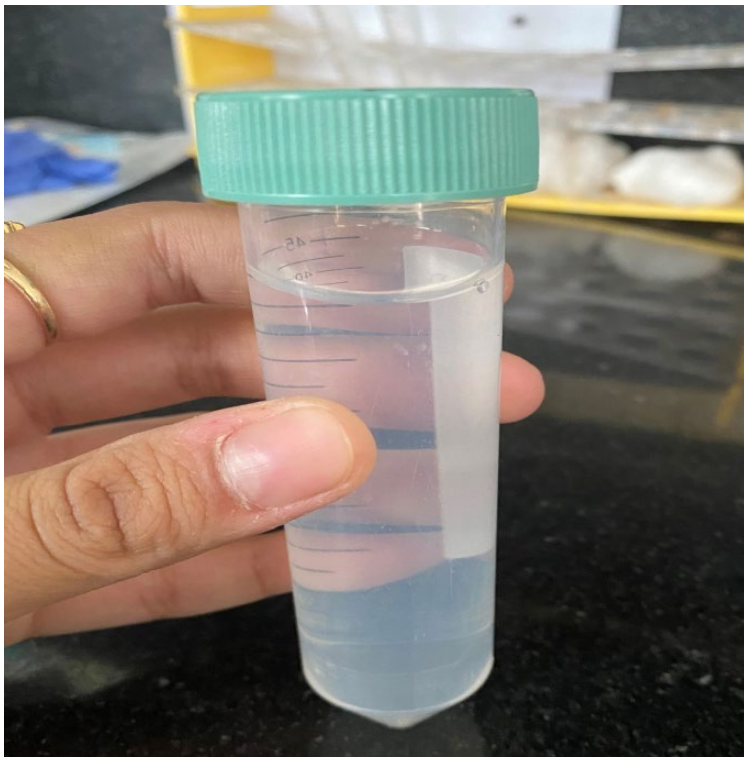

Fig 11: Solution of silver nitrate (5Mm) [41]

3. PREPARATION OF SILVER NANOPARTICLES

- To prepare silver nanoparticles the extract and silver nitrate solution was used.
- 5ml of extract and 5 ml of silver nitrate solution was added in a falcon and then covered with aluminum foil and place in an incubator at 37 degrees for one day. As this is a slow reaction it would take this much of time.
- Next day it was observed and spectra was taken around 300-700nm.
- The following graph was obtained.

Fig 12: Formation of silver nanoparticles (incubated at 37 degrees) [41]

Fig 13: Formation of silver nanoparticles (dark brown color indicates) [41]

CHAPTER 6

RESULT

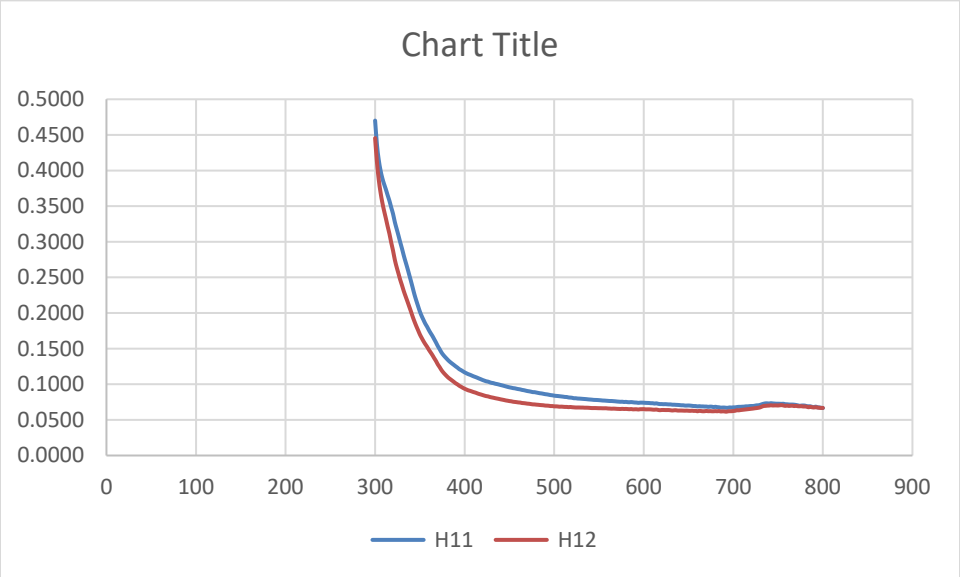

Fig 14: Spectra of theUrtica Dioica & silver nitrate sample (between 300-800nm) [41]

Dark brown colour indicates the presence of but after taking OD we got to know that the yield was very less. It was a slow reaction and took 2-3 days to show even slight brown colour. The yield was less so that indicates that the formation of nanoparticles was not accurate.

CHAPTER 7

CONCLUSION AND FUTURE PROSPECTS

The study shows the importance of nanoparticles and most importantly silver nanoparticles. They have adverse affects and shows a lot of properties that might come in use for medicine, drugs, antibacterial activity, antimicrobial activity, anti-inflammatory properties etc.

Not only stinging nettle silver nanoparticles can be extracted using various plants and fruits like orange peel potato peel etc. After extraction these are characterised on the basis of their shape , size etc by using TEM analysis and various other methods to know the quality of our extracted nanoparticles. The work will give good opportunity to shift towards nanotechnology and nanosciences. The formation of particles from green synthesis method was the best approach because by other methods harmful gases are released that harms us and the environment as well. Knowing that the cost of other methods aws high So in order to avoid that green synthesis was chosen. It was eco-friendly, cost-effective, simple, easy and highly approachable .

CHAPTER 8

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