GREEN SYNTHESIS OF MAGNESUIM OXIDE NANOPARTICLES USING BIOPHYTUM SENSITIVUM EXTRACT

Dissertation submitted in partial fulfillment of the requirements for the Degree of Bachelor of Science in Chemistry, Kerala University Trivandrum, Kerala

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DECLARATION

We hereby declare that this project work entitled “Green synthesis of Magnesuim Oxide nanoparticles using Biophytum Sensitivum extract” submitted to Kerala University in partial fulfillment of Bachelor's Degree in Chemistry is a bonafide record of the work carried out under the guidance of Dr. Bessy Mary Philip, Assistant Professor, Department of chemistry, Bishop Moore College during the year 2022-2023 and no part of it has been submitted for any other degree or diploma.

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CERTIFICATE

Certified that the dissertation entitled "Green synthesis of Magnesuim Oxide nanoparticles using Biophytum Sensitivum extract" is a bonafide record of the project work accomplished by K. Anagha, Keerthy. L and Lekshmi. A. B, in partial fulfillment of the requirement for Bachelor Degree in Chemistry of Kerala University is an authentic work carried out under my supervision and guidance during the period of 2022-2023.

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ABSTRACT

Magnesium oxide nanoparticles (MgO NPs) are of special interest in research activity because of their astonishing properties such as promising structural materials in biological implants due to their high strength to weight ratio, low density, good functionality, recycling activity, nontoxic, and hygroscopic nature. It is widely applicable for toxic waste remediation, antibacterial materials, removal of industrial pollutants and used in anti-arthritic and anti-cancer activities. In this study MgO nanoparticles were synthesized via a simple and ecofriendly green synthesis approach using the leaf extract of Biophytum Sensitivum. The characterization of the synthesized nanoparticles was done by Xray Diffraction and Transmission Electron Microscope. It was observed that use of Biophytum Sensitivum leaf extract makes a simple, eco-friendly, and cost-effective method for the preparation of the MgO NPs and can reduce Mg^{+2} ions to MgO NPs within 10 min of reaction time. MgO nanoparticles formation was recognized by color changes to dark brown and the XRD and TEM results confirm the presence of nanoparticles having polycrystalline cubic structure with an average size of 20nm.

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CHAPTER 1

INTRODUCTION

1.1Nanotechnology

Nanotechnology is the study of manipulating matter the atomic and molecular scale. It is the science that deals with matter at the scale of one billionth of a meter. A nanoparticle is the most fundamental component in the fabrication of a nanostructure, which is bigger than an atom or simple molecule that are governed by quantum mechanics. In general, size of a nanoparticle spans range between 1 and 100 nm. Nanotechnology is the study and use of materials, devices and systems on the scale of a nanometer. It is used in chemical processing techniques, such as filtration and catalysis. Nanotechnology is already with us, but it's impact on our daily lives is set to grow at a phenomenal rate. The ideas and concepts behind nanoscience and nanotechnology started with a talk entitled “There’s Plenty of Room at the Bottom" by physicist Richard Feynman at an American Physical Society meeting at the California Institute of Technology (Cal Tech) on December 29, 1959, long before the term nanotechnology was used. In his talk, Feynman described a process in which scientists would be able to manipulate and control individual atoms and molecules. After a decade, Professor Norio Taniguchi coined the term nanotechnology. After the development of scanning tunneling microscope that could find individual atoms, then the modern nanotechnology began [1]. Nanotechnology is the act of manipulating materials at very tiny scales. In considering materials under 100nm, the normal rules of Physics, Chemistry no longer can be applied. These materials start to display unique and sometimes surprising properties. They will become more conductive or reactive.

1.2 Applications of Nanotechnology

a) Medicines

Researches are developing nanoparticles in the size of molecule that deliver drugs directly to diseased cells in our body. When it is perfected, this method should greatly reduce the damage treatment such as chemotherapy does to a patient’s healthy cells. One of the applications of nanotechnology in medicine is currently being developed involves employing nanoparticles to deliver drugs, heat, light or other substances to specific types of cells (such as cancer cells). These particles are designed so that they are attracted to diseases cells, which allow direct treatment of those cells. It reduces damage to healthy cells. It helps for the early detection of diseases. Nanoparticles that deliver chemotherapy drugs directly to cancer cells are under development. These tests are in progress for the targeted delivery of chemotherapy drugs and their approval for their uses are in pending. Researches of University of Illinois have demonstrated that gelatin nanoparticles can be used to deliver drugs to damaged vaccine. Nanoparticles protect the vaccine by allowing the vaccine time to have a triggering effect to the immune response. Researches are in progress of developing a nanoparticle that can be consumed orally and that passes through the lining of intestine into the blood streams. This should allow drugs that must now be delivered with a shot to be taken in pill form. Researchers are also developing a nanoparticle to defeat viruses' molecules, but should be capable to deliver an enzyme that prevents the reproduction of viruses' molecules in the patient's bloodstream. Magnesium oxide nanoparticles also possess antibacterial activity against food borne pathogens such as E.coli and Salmonella entrididis by causing leakage in their cell membrane which leads to the death of the microorganism.

b) Nanoelectronics

- Single -walled carbon nanotubes have great ability for the application in the flexible electronics and cadmium selenide nanocrystals deposited on plastic sheets have been shown to form flexible electronic sheets [2].
- Magnesium oxide is being utilized for the electrical insulation. A low power method is used for the nanomagnets as switcher like transistors, in electrical circuits.
- Silver nanoparticles ink was used to form the conducting lines needed in circuit boards.

c) Food

Researches are using silicate nanoparticles to provide a barrier to gasses or moisture in a plastic film used for packaging. This could reduce the possibility of food spoiling or drying out [3]. Zinc oxide nanoparticles can be incorporated into plastic packaging to block UV rays and provide antibacterial protection, while improving the strength and stability of the plastic film. A method of spraying carbon nanotubes onto flexible plastic surfaces to produce sensors have been developed. The researchers believe that this method could produce low-cost sensors on surface such as the plastic film wrapping food, that the sensor could detect spoiled food.

d) Space

- Employing materials made from carbon nanoparticles to reduce the weight of spaceships while retaining or even increasing the structural strength

- Using carbon nanotubes is to make the cable needed for the space elevator, a system

- which could significantly reduce the cost of sending material into orbit.

- Including layers of bio-nano robots in spacesuits. The outer layer of bio-nano robots would respond to damages to the space suite,

- It also develops a network of Nano sensors to search large areas of planets such as mars for traces of water or other chemicals

e) Fuel Cells

Researchers found that the spacing between platinum nanoparticles affected the catalytic behavior, and that by controlling the packing density of the platinum nanoparticles they could reduce the amount of the platinum needed. Researches at Brown University are developing a catalyst that uses no platinum. The catalyst is made from a sheet of graphene coated with cobalt nanoparticles. If this catalyst works out for production use fuel cells it should be much less expensive than platinum-based [4] catalyst.

1.3 Magnesium Nanoparticles

In the last few decades, there has been a trend involving the use of nanoscale fillers in a variety of applications (eg: in industry, agriculture and medicine). One of these promising materials is Magnesium oxide (MgO), the unique properties of which make it a suitable candidate for use in a wide range of applications. Generally, MgO is a white, hygroscopic solid material and its lattice consists of Mg2+and O2- ions. Nanostructured MgO can be prepared through different chemical (bottom-top approach) or physical (top- down approach) routes the required resultant properties (eg: band gap, crystalline size, and shape) can be achieved depending on the reaction conditions, basic starting materials or their concentrations. In addition to its unique material properties, MgO is also potentially of interest due to its non-toxicity and environmental friendliness which allow it to be widely used in medicine and biotechnological applications.

1.3.1 Applications of Magnesium Nano particles

Magnesium oxide nanoparticles have emerged as a potential candidate for meeting ends of various problems due to its unique properties such as biodegradability, non-toxicity, inhibition of biofilm growth and degradation of harmful dyes such as methyl violet and many more. Along with its easy synthesis by methods such as sol-gel technique, precipitation method, and green synthesis, it is widely applicable for toxic waste remediation, antibacterial materials, removal of industrial pollutants and used in anti-arthritic and anti-cancer activities. Prior reviews have laid focus on singular domains whereas our review clubs three major domains i.e., clinical, agricultural, and environmental that are involved in the day-to-day life of plants as well as animals.

a) Agricultural

Magnesium oxide nanoparticles have also been known to comprise a number of advantages such as negligible phytotoxicity, thermal stability, non-genotoxicity as well as non-biotoxicity to humans, enabling brilliant application prospects for plant protection [5]. These nanoparticles apart from the above-mentioned properties have various other characteristics too which offers them prominent application in the various other agricultural prospects. Further these nanoparticles also help in increasing agricultural production of peanut by enhancing the development of seedling and plant growth as well as are being used as an approved food additive, food supplement, color retentate, etc.

b) Environmental

Environmental contamination is a serious problem which is faced by developed as well as developing nations around the globe. There are many ways to deal environmental contamination but the drawback which lies are the side effects of those cleansing agents when they themselves start behaving as a contaminant. Nanoparticles have evolved as an excellent alternative for various other ways of environmental cleanliness. Magnesium oxide nanoparticles, within a short span of time, has been successful in showing its presence in various environmental applications by the diverse range of properties it exhibits. These metal oxide nanoparticles possess high absorbing properties along with large surface area and high response capacity because of which they are being used as a potential absorbent of toxic gases such as nitrogen dioxide and sulphur dioxide [6-10].

c) Nanotoxicity

The varied range of Magnesium oxide nanoparticles has held it strong to accomplish applications in industries for cosmetics and skin care products as well as in biomedical for nanoelectronics biosensors, disease staging, etc. leaving behind some unwanted side effects, which are better pronounced as nanotoxicity. Nanoparticles, due to their small size and high reactivity can enter edible materials through their improper disposal into soil and water which can lead to adverse health effects. In couple of cases toxicity of magnesium oxide nanoparticles have been proven where V.B. Shah et al. reported the adverse effects of magnesium oxide nanoparticles leading to cellular apoptosis on the embryos of zebrafish [11,12] and it is also emphasized that magnesium oxide nanoparticles, in significant amount, when injected in Wistar rats resulted in alteration of biochemical pathways and DNA damage. Solely, not only magnesium oxide nanoparticles but TiO2 and cerium oxide nanoparticles along several others in the queue have been reported to cause toxicity in either plants or animals [13]. Although the toxicological effects of magnesium nanoparticles have been reported, but they are still widely used in various domains because the adverse effects of these nanoparticles are not proven to be lethal till date [14,12].

1.4 Synthesis of Magnesium Nano particles

1.4.1 Co-Precipitation

This method is widely used for the synthesis of nanoparticles. It is Based on the principle of precipitation and very often involves liquid-phase synthesis [15] and less often, vapor-phase synthesis [16]. Sodium hydroxide is commonly used as the Precipitating agent [17,18]. The basic principle is the homogenization of the precipitation. Reaction involves two processes—

i. nucleation and
ii. nuclei growth [19]

Generally, three principles are considered:

i. single nucleation and uniform growth by diffusion;

ii. nucleation, growth, and aggregation of smaller subunits;
iii. multiple nucleation and Ostwald ripening growth [20].

The critical solute concentration that initiates the process plays a major role in the classical process, with solute diffusion on the surface causing growth. Furthermore, it is necessary to separate these two processes. The resulting precipitate is then washed and dried.

1.4.2 Sol–Gel Method

The sol–gel method is one of the most fundamental approaches aimed at the formation of new material structures (primarily metal oxides and similar inorganic materials) in the presence of an inorganic precursor and an organic solvent [21]. The sol–gel approach was first introduced in the mid-19th century to produce silica gel [22]. Metal alkoxides together with suitable solvents and reactants can form homogeneous solutions, which can then form colloidal suspensions (sol) and eventually polycondense into integrated Networks (gel) [23], which are then transformed into xerogels or aerogels depending on the Drying method.

1.4.3 Solvo- Hydrothermal Method

The solvothermal method is another of the most widely used methods for controlled crystal growth of various materials [24]. When a precursor and suitable solvent are placed in an autoclave under simultaneous exposure to higher temperature and pressure, the desired products are formed [25]. It is the reaction conditions (temperature and pressure) that allow the formation of high crystallinity materials compared to the co-precipitation method [18]. In the case of the definition of the terminology of the “solvothermal” method a medium other than water (e.g., alcohols, or organic and inorganic solvents) is generally used as the solvent. In cases where water is used as the solvent, this method can be defined as “hydrothermal”.

1.4.4 Combustion Method

The combustion method is a method frequently used for the synthesis of metal oxide nanoparticles, mainly due to its efficiency and low-cost [26]. It can be based on two different approaches. The first is the so-called self-propagating synthesis and the second is the volume combustion synthesis [27]. In the case of self-propagating synthesis, it involves spontaneous redox reactions ignited by an external source that takes place between the precursor (oxidizer) and the reductant (fuel) mixed at the molecular level in solution, with the formation of solid products occurring without any further input energy [28]. In the Second case, the entire sample is heated until the reaction is initiated in its entire volume. This method of preparation is more difficult to control and is recommended especially for weak exothermic reactions that require preheating before ignition [29]

1.4.5 Green Synthesis

The so-called green synthesis is a modern approach to nanoparticle production, whereby there are no (or minimal) requirements for reaction conditions (high pressure, temperature, and energy) and no toxic chemicals are used [30]. The aim is to minimize the produced waste and to establish sustainable development in this field [31], as nontoxic reagents can be used in this production method. These can be various agents ranging from plant extracts [32] and bacterial strains [33] to enzymes and vitamins. Double distilled water is commonly used as the extraction medium in this process. In simplified terms, the process can be considered to comprise three fundamental stages: a) activation b) growth c) process termination.

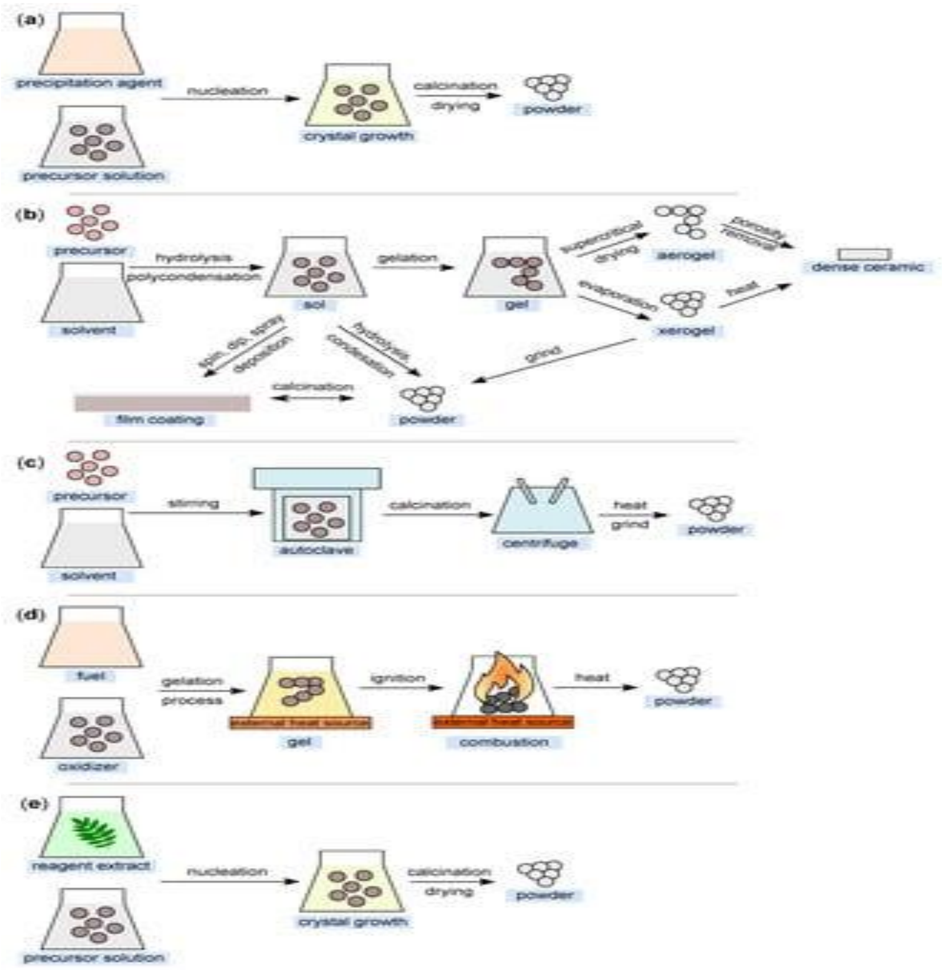

Fig 1.1 Illustration of synthesis of nanoparticles using different methods

1.5 Biophytum Sensitivum

Kingdom	:	Plantae
Family	:	Oxalidaceae
Genus	:	Biophytum
Species	:	B. sensitivum

Fig 1.2 Biophytum Sensitivium (Mukkutti)

Biophytum sensitivum, also known as little tree plant is a species of plant in the genus *Biophytum* of the family Oxalidaceae commonly called "Mukkutti". It is commonly found in Kerala, wet lands of Nepal, tropical India and in other Southeast Asian countries and is used for medicinal purposes in Nepal and India. The plant is also a common weed in tropical greenhouses, however, *Biophytum sensitivum* is particularly sensitive to spider mites. Investigations have been undertaken into the plant's chemistry, biological activities, and medicinal uses[41]. Similarly to *Mimosa pudica*, the leaflets of *Biophytum sensitivum* are able to move rapidly in response to mechanical stimulation such as touch. The little plant grows up to maximum of 20 cm and possess unbranched woody erect stem. Leaves: Leaves abruptly pinnate, leaflets opposite, 6 to 12 pairs, and each leaflet is up to 1.5 cm long, the terminal pair is the largest. Flower: The flowers are many and crowded at the apices of the numerous peduncles, normally yellow, white, or orange with red streak in the centre of each of the five petals. The sepals are subulate-lanceolate, striate, and about 7 mm long. Fruits: Fruits are ellipsoid capsules which are shorter than the persistent calyx. [42] Chemical analyses have shown that the plant parts are rich in compounds such as amentoflavone, cupressuflavone, and isoorientin. Its extracts are traditionally believed to be antibacterial, anti-inflammatory, antioxidant, antitumor, radioprotective, chemoprotective, antiangiogenetic, wound-healing, immunomodulatory, anti-diabetic, and cardioprotective in nature [40-42].

CHAPTER 2

REVIEW OF LITERATURE

Green synthesis approaches are gaining attention of researchers as they are less hazardous ways to obtain nanoparticles. Mg NPs have been effectively produced by green synthesis which is mainly very eco-friendly, nontoxic, great stability, also broad range of opportunities to produce materials in the nanoscale.

- Suresh et al. used an extract of Nephelium lappaceum L. and double distilled water for green synthesis, whereby magnesium nitrate was used as a precursor. They verified the cubic structure of MgO by the investigations that were carried out, and the average crystallite size was determined as 55 nm, which agreed very well with the SEM measurements (grain size 60–70nm)[34].
- Vargheese and Vishal [35] synthesized MgO by using Trigonella foenum-graecum extracted in double distilled water using magnesium nitrate as a precursor. The average crystalline size determined from XRD was around 14 nm. It is evident from the SEM measurements that the particles prepared in this way had a mixture of fine, spherical structures.
- Younis et al. used Rosa floribunda powder, which they dispersed in double distilled water, were magnesium nitrate was used as a precursor. The results show a cubic structure of high purity, and the nanoparticle size, as determined using high-resolution TEM, was around10nm [36].
- The synthesis of Rosmarinus officinalis L. with bulk MgO was studied by Abdallah et al. The resulting nanoparticles contained minimal impurities and had a hexagonal crystal structure, unlike in the previous cases. The average particle size described in this study was 8.8 nm [37].
- Khan et al. used Dalbergia sissoo extract and magnesium nitrate as a precursor. In their study, the authors focused on describing several aspects (extract concentration, precursor, and pH) affecting the bandgap size and photocatalytic activity. The cubic structure of MgO is evident from the results with a particle size of around 50 nm [38].

- The use of Saussurea costus biomass was the focus of the work by Amina et al. [39] in which magnesium nitrate was used as a precursor. The results show that the authors were able to produce particles with a cubic structure and a size of about 30 nm using this procedure.
- On the other hand, MgO particles with dimensions less than 20 nm were formed by phyto-assisted synthesis by Sharma et al. They used Swertia chirayaita as the reactant and magnesium nitrate as the precursor. The SEM results reported in their study show the spherical shape of the particles with slight variation in shape [40].
- Using magnesium chloride as a precursor and Moringa oleifera as a reagent, Fatiquin etal. attempted to synthesize MgO [41]. Their efforts resulted in particles with a crystalline size of around 21 nm. According to TEM, they exhibited a cubic structure and range in size between 2 and 50 nm.
- Nguyen et al. synthesized MgO particles from extracts of Tecoma stans L. (flower, bark, and leaf) and a magnesium nitrate precursor. The resulting particles had a spherical or hexagonal morphology depending on the used extract, with the average crystal size ranging from 20 to 50 nm. The authors determined that the flower extract was the most promising, mainly due to the high absorption capacity of the synthesized particles [42].

CHAPTER 3

OBJECTIVES

This project is an attempt to synthesize magnesium oxide nanoparticles, from the aqueous green leaf extract leaf extract of Biophytum Sensitivium and its characterisation. The primary objectives of the project are:

- The green synthesis of MgO nanoparticles from Biophytum Sensitivium leaf extract which can act as a reducing agent.

- This method can be used for the rapid and ecofriendly synthesis of stable magnesium oxide nanoparticle.

- To study the reduction of ionic Mg to MgO NPs.

- To characterize the presence of MgO nanoparticles using XRD and particle size determination and distribution using HR-TEM with SAED.

CHAPTER 4

MATERIALS AND METHODS

4.1. Materials

Biophytum Sensitivium leaf is used as the raw material. The chemical magnesium nitrate hexahydrate of 99 % purity (Merck) used in this experiment, is of analytical grade was dissolved in deionised water. The equipment that was used for the experiments are grinder, conical flask, magnetic stirrer and Whatman No.1 filter paper is used for filtering.

4.2 Methods

a) Collection of plant leaf

Fresh young Biophytum Sensitivium leaves with stem were collected in and around mavelikara. The collected leaf was tightly packed with polythene bag and then transfer to the laboratory. It is then washed in tap water to remove dust and then with distilled water twice.

b) Preparation of plant extract

Take 50gm of fresh plant leaves of (mukkutti) Biophytum Sensitivum and then transferred the leaves into a mortar and Pestle for crushing the leaves. It was transferred to a 250ml beaker and added 100ml of deionized water then it was subjected to boiling for 30 minutes then after cooling, muslin cloth was used to filter the extract followed by Whatman No.1 filter paper. Then the filtrate is stored in a tight seal pack under 4^0C for further use. This filtrate was used for the synthesis of magnesium oxide nanoparticles.

c) Preparation of 1M hydrated Magnesium Nitrate

Prepared 1 M of hydrated magnesium nitrate solution by taking 5.128 gm of powdered. Magnesium nitrate hexahydrate in a beaker and mixed with 100ml of water and is stirred for 30 minutes to form a uniform solution.

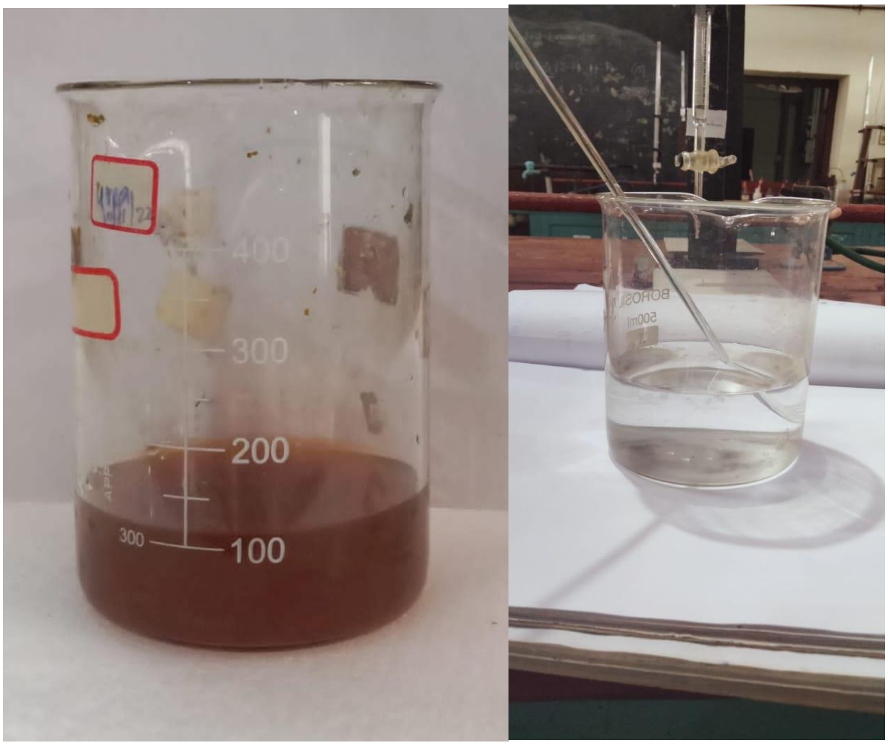

Fig 4.1 a) Plant leaves extract after filtration b) 1M hydrated magnesium nitrate

d) Synthesis of MgO nanoparticles

Exact amount (1M) of Mg $(NO_3)_2$ .6 H_2O was dissolved in 100 ml de-ionized water and under magnetic stirring at room temperature (1200 rpm). After obtaining a homogenous solution, 1-20 ml of an aqueous solution of Biophytum Sensitivium extract was added drop by drop in magnesium nitrate solution turned in white color to brown color which in turn changed to brown coloured colloidal solution. Leave-taking the mixture under stirring for 5-minute vigorous stirring. The suspended particles were purified by dispersing in sterilized distilled water and centrifuged for 15 minutes 4 times using deionized water and alcohol. After calcination at 1000 ^{0}c for five hours, MgO nanoparticles are formed.

Fig. 4.2 a) Formation of colloidal MgO b) After centrifugation

4.3 Characterization Technique

4.3.1 X-Ray Diffraction

X-ray diffraction (XRD) is a powerful nondestructive technique for characterizing crystalline materials. It provides information on structures, phases, preferred crystal orientations (texture), and other structural parameters, such as average grain size, crystallinity, strain, and crystal defects. X-ray diffraction peaks are formed by constructive interference of a X- ray beam at specific angles from each set of lattice planes in a sample. The peak intensities are determined by the distribution of atoms within the lattice. Therefore, the x-ray diffraction pattern is the fingerprint of periodic atomic arrangements in each material. A search of the International Centre for Diffraction Data (ICDD) standard database of x-ray diffraction patterns enables quick phase identification for a large variety of crystalline samples. XRD is carried out using Bruker, D8- Advance P-XRD-Analysis

4.3.2 Transmission Electron Microscope

Almost all the components in the TEM are the same. The Guns, lenses and detectors are combined to form the microscope and divide it into three component system: the illumination system, the objective lens/stages, and the imaging system.

1) The illumination system comprises the gun and the condenser lens. Its role is to take the electrons from the source and transfer them to the specimen.

2) The objective lens/stage system is the heart of TEM. The afflictive region was extended over less than 1cm along the length of the column. All the beam specimen interactions take place and create bright field, dark field images and selected area diffraction pattern (SAD) that are the fundamental TEM operations

3) The imaging system uses several lenses to magnify the image or the diffraction pattern produced by the objective lens and to focus these on the viewing screen.

TEM analysis is carried out using HR-TEM with SAED.

CHAPTER 5

RESULTS AND DISCUSSION

5.1 X-Ray Diffraction (XRD) analysis

Fig (5.1) shows the X-ray diffraction (XRD) pattern of MgO nanoparticles synthesized from magnesium nitrate hexahydrate in the presence of Biophytum Sensitivium extract at room temperature. In XRD pattern the peak position and peak intensity are in good agreement with already reported XRD pattern of MgO nanoparticles and hence can confirm the formation of polycrystalline cubic structure of MgO nanoparticles. The high intense peak at $2\Theta = 42$ degree reveals its high crystallinity nature.

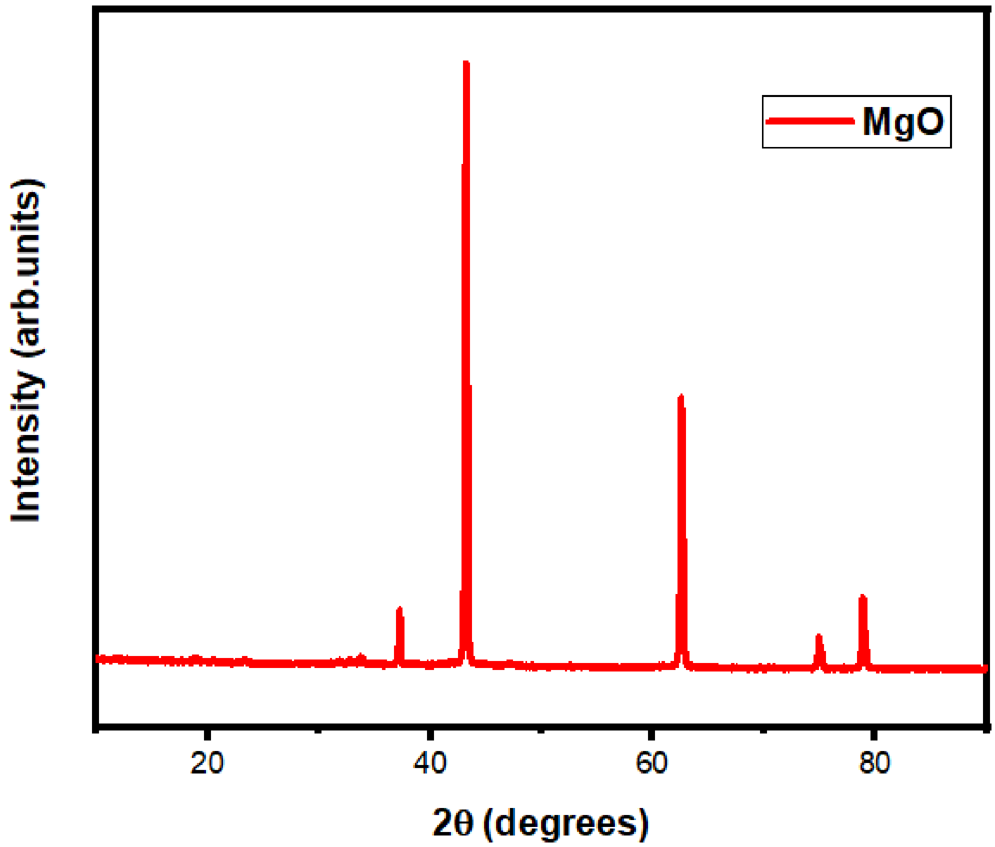

Fig 5.1 XRD pattern of synthesized MgO nanoparticles

5.2 Transmission Electron Microscopy (TEM) Analysis

HR-TEM images (Fig. 5.2) showed that MgO nanoparticles are pure, polycrystalline and the average particle size is 20nm.

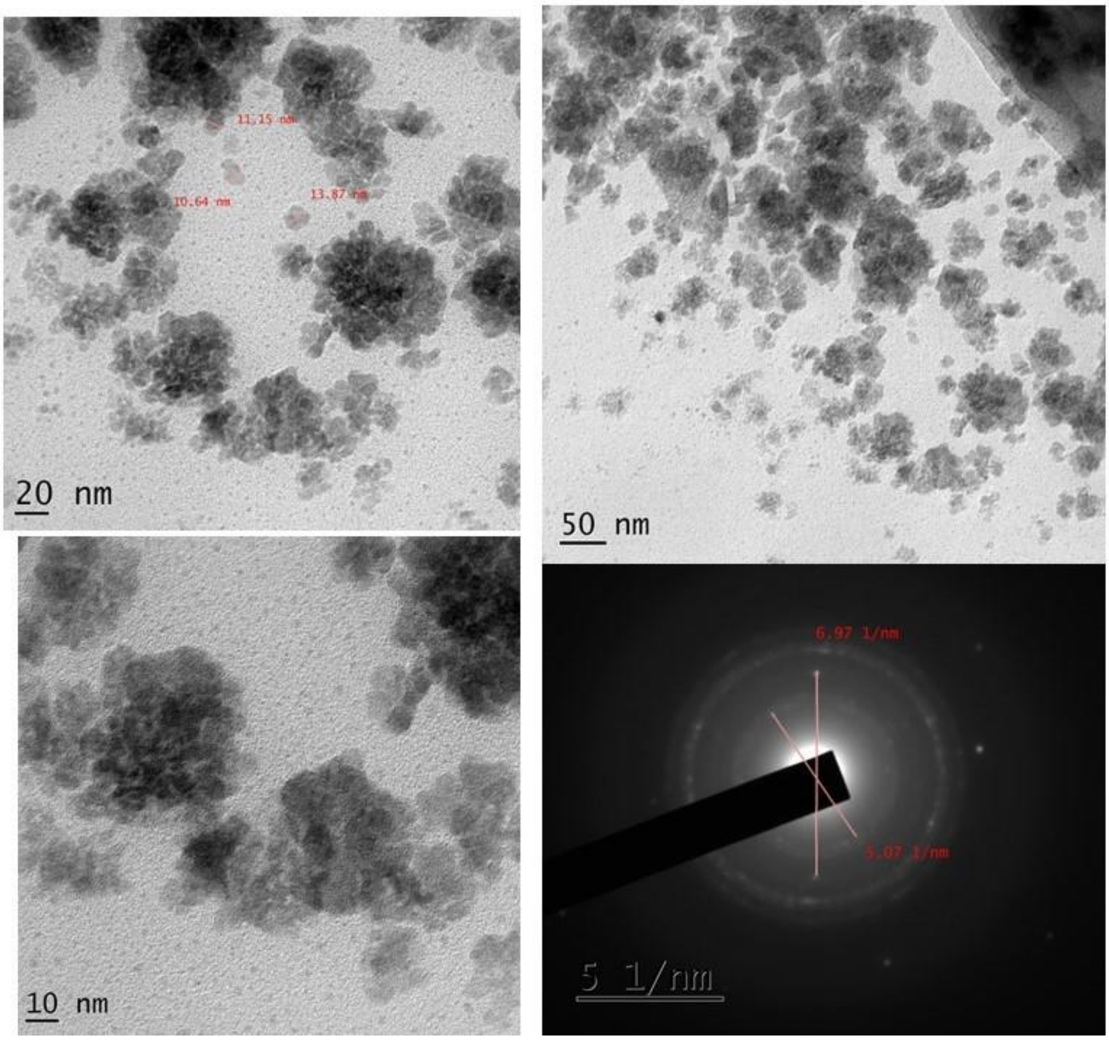

Fig 5.2 TEM images and SAED pattern of MgO nanoparticles

CHAPTER 6
CONCLUSION

In this study, reported a simple, green, and efficient route to synthesis Mg NPs by treating magnesium nitrate hexahydrate with Biophytum Sensitivum at room temperature. The leaf extract plays an important role as the bio friendly reducing and stabilizing agent reduces the cost of production and the environment impact. The characterization of the synthesized nanoparticles was done by Xray Diffraction and Transmission Electron Microscope. It was observed that use of Biophytum Sensitivum leaf extract makes a simple, eco-friendly, and cost-effective method for the preparation of the MgO NPs and can reduce Mg^{+2} ions to MgO NPs within 10 min of reaction time. MgO nanoparticles formation was recognized by color changes to dark brown and the XRD and TEM results confirm the presence of nanoparticles having polycrystalline cubic structure with an average size of 20nm.

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