BHARATA MATA COLLEGE

THRIKKAKARA

MAHATMA GANDHI UNIVERSITY

PROJECT REPORT ON

ZINC OXIDE (ZnO) NANOPARTICLE SYNTHESIS, CHARACTERIZATION, AND IT'S TOXICITY IN PLANTS

SUBMITTED BY

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B.Sc physics model 2(2021-2024)

DECLARATION

This dissertation “ZnO Nanoparticle Synthesis, Characterization and its Toxicity in Plants” is a bona fide work done by me at Bharata Mata College, Thrikkakara under the guidance of Dr. Sr. Rintu Varghese, Department of Physics. I further declare that this is my original work, as part of my academic course.

The work reported in this project is completely known to me and true.

Place: Thrikkakara

Name: Fathima saffrin V.A , Nadiya P.T

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ABSTRACT

Title: Zinc Oxide (ZnO) Nanomaterial Synthesis, Characterization, and Applications.

Zinc oxide (ZnO) has garnered significant attention in the scientific community due to its versatile properties and wide-ranging applications in various fields. This review provides an overview of the synthesis methods, characterization techniques, and diverse applications of ZnO nanomaterials. Characterization plays a pivotal role in understanding the structural, morphological, and optical properties of ZnO nanostructures. Techniques such as X-ray diffraction (XRD), scanning electron microscopy (SEM), Fourier-transform infrared spectroscopy (FTIR), and UV-visible spectroscopy provide insights into crystal structure, particle size, surface morphology, chemical composition, and optical bandgap of ZnO nanomaterials.

Therefore the objective of this study is to synthesize zinc oxide nanostructures with the most practical way precipitation method, characterize the nanostructures, and application to study their toxicity in plants.

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CHAPTER 1

Introduction On Nanoparticle

1.1 Nanoparticle

Nanoparticles are minuscule particles, usually measuring less than 100 nanometers in at least one dimension. Materials such as metals, metal oxides, polymers, and carbon-based materials like graphene can be used to create these particles. Nanoparticles frequently have distinct physical, chemical, and biological properties that set them apart from their bulk counterparts because of their minuscule size. Because of their high surface area to volume ratio and tunable qualities, they are widely used in many different fields, including environmental remediation, electronics, medicine, and catalysis.

Nanoparticles (NPs) are materials with a size range of 1 to 100 nm. Based on their characteristics, forms, or sizes, they can be divided into several classes. Fullerenes, metal NPs, ceramic NPs, and polymeric NPs are among the various groups. Because of their large surface area and nanoscale size, NPs have special physical and chemical characteristics. It is stated that their size affects their optical characteristics, resulting in varying colors because of absorption in the visible spectrum.

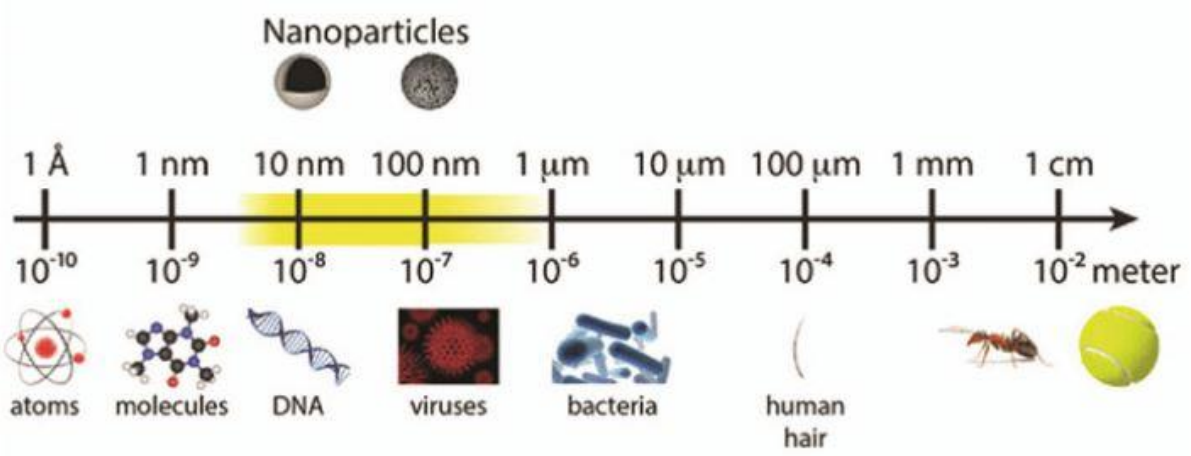
Top-down and bottom-up methods are frequently employed in nanoparticle research to create and modify nanoparticles with particular characteristics and functions.

Top-down approach: Using a variety of physical or chemical techniques, bigger structures or materials are reduced to smaller nanoparticles in the top-down approach. For instance, lithography techniques use techniques like electron beam lithography or photolithography to create nanoscale patterns on a substrate. Then, by using techniques like etching or deposition, these patterns can be utilized to produce nanoparticles. The mechanical milling of bulk materials to create nanoparticles is another illustration. Using grinding or milling techniques, large materials are physically reduced to smaller particles in this method.

Bottom-up method: Using atomic or molecular precursors, the bottom-up method builds nanoparticles and then progressively assembles them into larger structures. Common bottom-up techniques in chemical synthesis include sol-gel synthesis, chemical vapor deposition (CVD), and solution-phase synthesis. These techniques create nanoparticles by carefully regulating the reaction of precursor molecules. Bottom-up strategies also include

self-assembly methods, in which nanoparticles spontaneously assemble into ordered structures in response to external stimuli or their intrinsic characteristics. Lipid-based nanoparticle formation and DNA-based nanoparticle self-assembly are two examples.
[1]

1.2 Nanosize



fig(1.21)

Comparison of materials with nanoscale dimensions in varying sizes

Human hair typically has a diameter of 50–100 micrometers (µm), which is significantly larger than most nanoparticles. Although bacteria vary greatly in size, many of them are between 0.2 and 10 micrometers (µm) in size. Usually thousands of times smaller than bacteria, nanoparticles are much smaller. Typically, viruses range in size from approximately 20 to 300 nanometers, making them even smaller than bacteria. A red blood cell is much larger than most nanoparticles, with a diameter of about 6 to 8 micrometers (µm). A DNA double helix's width is roughly 2 nanometers (nm), or about the same size as some smaller nanoparticles. These contrasts demonstrate how tiny nanoparticles are about commonplace items and biological structures.

Example: Silver nanoparticles: These are utilized in textiles, medical device coatings, and wound dressings due to their antimicrobial qualities. The special optical and catalytic properties of gold nanoparticles make them useful in cancer therapy, diagnostics, and chemical reaction catalysis. Because of their special characteristics, nanoparticles have many uses in a variety of fields. Several noteworthy applications consist of:

Healthcare and Medicine:

Drug Delivery: By encapsulating drugs in nanoparticles, medications can be delivered to particular cells or tissues with greater precision, fewer side effects, and increased therapeutic efficacy. Medical Imaging: In imaging techniques like MRIs, CT scans, and fluorescence microscopy, nanoparticles like iron oxide and quantum dots are used as contrast agents. This allows for more precise diagnosis and the visualization of biological structures.

Remediation of the Environment:

Water Treatment: By using techniques like adsorption, photocatalysis, and filtration, nanoparticles are used to remove impurities from water.

Air purification: By assisting in the breakdown of air pollutants, nanoparticles coated with catalytic materials can help create cleaner indoor and outdoor environments.

1.3 Nanotoxicity

The term "nanotoxicity" describes how nanoparticles may hurt the environment and living things. Although nanoparticles have many uses and advantages, their small size and special characteristics can also interact with biological systems in ways that could have unexpected effects. Because of their large surface area to volume ratio, nanoparticles can interact and react with biological molecules more favorably. The stability, aggregation behavior, and interactions of surface coatings and functionalizations with cells and tissues can also have an impact on their toxicity. Reactive oxygen species (ROS) can be produced by nanoparticles via surface reactions, electron transfer, or photoexcitation. ROS can oxidatively damage biomolecules and cellular structures, which can result in inflammation, DNA damage, and cell death. After being exposed, nanoparticles can accumulate in particular organs and pose a risk to the liver, kidneys, lungs, brain, and other systemic organs. Size, shape, surface chemistry, and exposure route are some of the variables that affect a nanoparticle's biodistribution and clearance. When released into the environment by consumer goods, manufacturing processes, or waste disposal methods, nanoparticles can build up in ecosystems and interact with different trophic levels of organisms. Both aquatic and terrestrial species may be harmed by this, as well as ecological disturbances.

To tackle nanotoxicity, multidisciplinary endeavors are necessary to comprehend the fundamental mechanisms, establish dependable techniques for assessing toxicity, and execute plans for the secure creation and application of nanoparticles. Researchers and

regulatory bodies are attempting to set norms and guidelines for evaluating the safety of nanomaterials and reducing possible hazards to the environment and public health.

1.4 Methods Of Synthesis Of Nanoparticle

Synthesis techniques for nanostructured materials, which encompass nanoparticles, can be broadly classified into three categories: chemical, biological, and physical synthesis techniques.

The types of nanoparticles being produced, the desired size and shape, and the intended application are just a few examples of the many variables that affect nanoparticle synthesis techniques. Here are a few typical techniques:

Chemical Reduction: In this technique, metal ions in a solution are reduced to create nanoparticles. One method of creating gold nanoparticles is to reduce gold chloride using a reducing agent such as sodium borohydride or citrate.

The sol-gel method: involves hydrolyzing and condensing precursor molecules, then gelating the resulting three-dimensional network to create a sol—a stable colloidal suspension of nanoparticles. The nanoparticles are then created through drying and calcination.

Co-precipitation: In this technique, solutions containing metal ions and a precipitating agent are added simultaneously to precipitate nanoparticles from the solution. It is frequently employed in the synthesis of magnetic nanoparticles, such as iron oxide.

Microemulsion: These are thermodynamically stable mixtures of water, oil, and surfactant that can be used to create nanoparticles in their small spaces. The size and shape of nanoparticles can be precisely controlled using this technique.

Electrochemical Deposition: Using an electric current to a solution containing metal ions, nanoparticles are deposited onto an electrode surface in this technique. This technique is frequently applied to the synthesis of nanoparticle thin films.

Green Synthesis: To reduce metal ions and create nanoparticles, this eco-friendly method uses natural sources like plant extracts, bacteria, or fungi. Solvents and benign reducing agents are commonly used in green synthesis techniques.

Laser Ablation: In this process, a target that is submerged in a liquid is exposed to a high-energy laser beam, which causes the target to ablate and create nanoparticles in the surrounding solution.

Template-Assisted Synthesis: During synthesis, the size, shape, and structure of nanoparticles can be manipulated by using templates like porous membranes or self-assembling monolayers.

There are several ways to synthesize zinc oxide (ZnO) nanoparticles, and each one gives you a different level of control over the size, shape, and characteristics of the particles. The one we used is:

Chemical Precipitation: In this technique, a solvent, usually water or alcohol, is used to dissolve zinc salts like zinc acetate or zinc nitrate. Zinc hydroxide is then precipitated from the solution by adding a base, such as sodium hydroxide or ammonium hydroxide.

ZnO nanoparticles are created by heating or otherwise processing the resultant zinc hydroxide.

One frequently used technique in the laboratory for producing ZnO nanoparticles is the precipitation of zinc oxide (ZnO) using zinc acetate and sodium hydroxide (NaOH). This is the reaction's simplified equation:

Zn(OH)2 (s) + 2CH3COONa (aq) is the result of Zn(CH3COO)2 (aq) + 2NaOH (t)

ZnO (s) + H2O (l) → Zn(OH)2 (s)

Zinc acetate (Zn(CH3COO)2) and sodium hydroxide (NaOH) combine in this reaction to form zinc hydroxide, which is subsequently dehydrated to yield zinc oxide precipitate.

Zinc acetate is soluble in water and separates into acetate (CH3COO^-) and zinc ions (Zn^2+).

Strong base sodium hydroxide produces hydroxide ions (OH^-) in solution.

At first, zinc hydroxide forms as a white precipitate.

When heated or allowed to stand, the white zinc hydroxide precipitate dehydrates and releases water to form the white ZnO precipitate.

CHAPTER 2

ZnO Nanoparticle

2.1 Introduction

Because of their special qualities, zinc oxide (ZnO) nanoparticles have gained a lot of interest in a variety of industries, including electronics, optoelectronics, catalysis, biomedical applications, and environmental remediation.

Zinc and oxygen atoms arranged in a crystal lattice structure make up zinc oxide (ZnO) nanoparticles. Their size usually falls between 1 and 100 nanometers, and they have special optical, chemical, and physical characteristics. ZnO nanoparticles are helpful in medical applications like wound dressings, antimicrobial coatings, and disinfectants because it has been demonstrated that they have antibacterial qualities.

Numerous techniques, such as chemical precipitation, sol-gel synthesis, hydrothermal techniques, vapor-phase deposition, and template-assisted techniques, can be used to create ZnO nanoparticles.

Chemical properties: While pure zinc oxide (ZnO) is a white powder, zincite, a rare mineral found in nature, contains impurities such as manganese that give it a yellow to red color. Thermochronically, crystalline zinc oxide turns yellow when heated in air and back to white when cooled. An amphoteric oxide is zinc oxide. It dissolves in most acids, including hydrochloric acid, but is almost insoluble in water.

[2][3][4]

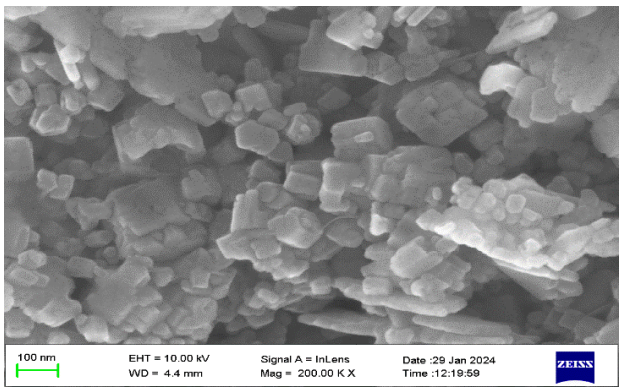


fig (2.11)

2.2 Instrumentation

X-RAY DIFFRACTION:

The experimental science of identifying a crystal's atomic and molecular structure is known as X-ray crystallography. The crystalline structure of a crystal causes an incident X-ray beam to diffract into numerous distinct directions. A three-dimensional image of the electron density inside the crystal can be created by a crystallographer by determining the angles and intensities of these diffracted beams. The locations of the atoms in the crystal, their chemical bonds, the degree of crystallographic disorder, and other details can all be inferred from this electron density.

FIELD EMISSION SCANNING ELECTRON MICROSCOPY (FESEM):

FESEM is a potent method for creating high-resolution surface images of a sample that is utilized in nanotechnology, materials science, and many other fields. At the nanoscale scale, it offers comprehensive details on the topography, morphology, and composition of materials. To create incredibly clear and detailed images, a focused electron beam is scanned over a specimen's surface by the FESEM. This allows it to detect a variety of signals, including X-rays, secondary electrons, and backscattered electrons. For scientists and engineers researching the composition and characteristics of materials, it is a priceless resource.

ULTRAVIOLET (UV) SPECTROSCOPY

It refers to the full, adjacent visible region of the electromagnetic spectrum as well as a portion of the ultraviolet or reflectance spectroscopy. This methodology is widely used in a variety of applied and fundamental applications due to its ease of implementation and relative affordability. All that is needed for the sample to be a chromophore is for it to absorb in the UV-Vis range. Fluorescence spectroscopy is enhanced by absorption spectroscopy. Other than the measurement wavelength, the parameters of interest are the absorbance (A), transmittance (%T), and reflectance (%R), as well as how they vary over time.

[5][6][7]

2.3 Application Of Nanoparticle

The applications of nanoparticles are of a wide range. here our application is to find its toxicity in plants. according to the data obtained, we find the optimum concentration in which its toxicity is maximum. further studies can be made by studying on its shape, structure, etc.

Among the noteworthy uses for nanoparticles are:
Personalized medicine and early detection can be achieved by using nanoparticles in diagnostic assays to identify pathogens, diseases, and biomarkers.
Medical imaging methods like fluorescence imaging, CT scans, and MRIs can have their sensitivity and resolution increased by functionalizing nanoparticles with imaging agents.
Zinc oxide (ZnO) nanoparticles are used in many different fields because of their special qualities and adaptability. Among the noteworthy applications are:

UV Protection: Because zinc oxide (ZnO) nanoparticles can both absorb and scatter ultraviolet (UV) radiation, they are frequently found in sunscreens, cosmetics, and textiles. They offer strong defense against UVA and UVB radiation, assisting in the prevention of skin damage and lowering the chance of developing skin cancer.
Photocatalysis: ZnO nanoparticles are helpful for environmental remediation applications because they show good photocatalytic activity when exposed to UV light. By aiding in the breakdown of toxic compounds into harmless substances, they can degrade organic pollutants.
Gas Sensors: Because of their large surface area, sensitivity to a wide range of gases, and capacity to promote the adsorption and desorption of gases, ZnO nanoparticles are used in gas sensing devices. They are used in healthcare, industry safety, and environmental monitoring to identify gases like carbon monoxide, nitrogen dioxide, and volatile organic compounds.

These are only a few instances of the many uses for ZnO nanoparticles, which emphasize how crucial they are for resolving a range of technical, environmental, and medical issues.

Research is still being done to find new uses for ZnO nanoparticles and improve their characteristics for better functionality and performance.

2.4 ZnO In Cancer Treatment

Many methods have been used to study the potential applications of zinc oxide (ZnO) nanoparticles in cancer treatment.

ZnO nanoparticles are being investigated for use in cancer therapy in several ways, including:

Drug Delivery: To enhance the delivery of anticancer medications to tumor cells, ZnO nanoparticles can be functionalized or loaded with drugs. By improving the drugs' solubility, stability, and targeting, the nanoparticles can boost their effectiveness and lessen systemic toxicity. ZnO nanoparticles can also be used to release medications under controlled conditions, resulting in long-lasting therapeutic benefits.

ZnO nanoparticles with photocatalytic properties, or the ability to produce reactive oxygen species (ROS) in response to light, are used in photodynamic therapy (PDT). ZnO nanoparticles are applied to the tumor site in photodynamic therapy, and they are subsequently activated by a particular wavelength of light.

Application: Because ZnO nanoparticles can both absorb and scatter ultraviolet (UV) radiation, they are frequently found in sunscreen lotions, makeup, and skincare products. Because they block UVA and UVB rays, they offer effective protection against sunburn and skin cancer. There is a wide range of applications of ZnO nanoparticles.

2.5 Zno Toxicity

Although ZnO nanoparticles are generally regarded as safe for topical application, some people may experience skin irritation or sensitization reactions at high concentrations or after extended exposure. ZnO nanoparticles, which are found in sunscreen powders and sprays, can irritate, redden, and discomfort the eyes. Applying should be done with caution to prevent direct eye contact and with the appropriate safety precautions in place.

Zinc oxide (ZnO) nanoparticles are being investigated for a range of therapeutic uses in cancer treatment, such as drug delivery, photodynamic therapy (PDT), and photothermal

therapy (PTT). According to certain research, ZnO nanoparticles may harm DNA and result in chromosomal abnormalities in cancerous cells. ZnO nanoparticles have great potential as a cancer treatment tool, but before they can be used in oncology, their safety profile and toxicity to living cells must be thoroughly assessed.

Chapter 3

Synthesis Of ZnO Nanoparticle

3.1 Introduction

Zinc oxide (ZnO) nanoparticles are usually synthesized or extracted using a variety of physical or chemical techniques designed to regulate the size, shape, and characteristics of the particles.

Chemical Precipitation: In this technique, a base (such as sodium hydroxide or ammonium hydroxide) is added dropwise while being stirred to dissolved zinc salts (such as zinc acetate and zinc nitrate). Zinc hydroxide precipitates as a result, and it then goes through thermal breakdown to produce ZnO nanoparticles. The temperature, pH, stirring rate, and other reaction parameters can be adjusted to fine-tune the size and shape of the nanoparticles.

A chemical precipitation process is used to extract or synthesize zinc oxide (ZnO) nanoparticles using zinc acetate and sodium hydroxide (NaOH). Here are the chemical equations involved in the process along with a general overview:

Zinc hydroxide is created when zinc acetate and sodium hydroxide react, and zinc oxide nanoparticles are the result of the thermal breakdown of zinc hydroxide.

Steps:

* To create a transparent solution, dissolve zinc acetate dihydrate

(Zn(CH3) COO) 2· 2 H 2 O Zn(CH3) COO) 2 · 2H 2 O) in distilled water. Using distilled water, dissolve sodium hydroxide (NaOH) to create a distinct solution.

*Continually stir the zinc acetate solution while gradually adding the sodium hydroxide solution. A white precipitate of zinc hydroxide will result from this.

*Keep churning the mixture for as long as necessary to guarantee full reaction and precipitation.

The precipitate can be separated from the reaction solution by filtering or centrifuging it.

*To get rid of any contaminants or unreacted reagents, wash the precipitate with distilled water.

* To obtain zinc hydroxide, dry the precipitate after washing it at a suitable temperature.

* Apply a high temperature to the zinc hydroxide to decompose it into zinc oxide nanoparticles.

* Use methods like transmission electron microscopy (TEM), X-ray diffraction (XRD), scanning electron microscopy (SEM), and UV-Vis spectroscopy to evaluate the size, shape, and optical characteristics of the synthesized ZnO nanoparticles.

By varying variables like reagent concentration, reaction temperature, and reaction duration, ZnO nanoparticles with precise size, shape, and characteristics can be synthesized via the chemical precipitation method. It is a reasonably easy and economical way to produce ZnO nanoparticles on a large scale for a variety of uses.

3.2 ZnO synthesis experiment

Aim: to obtain ZnO nanoparticles.

Apparatus used:500ml beaker(2),100 ml beaker (1), 2 Petri dishes, spatula, magnetic stirrer, chemicals: zinc acetate and sodium hydroxide, cylinders for precise liquid measurement, Centrifuge, Ultrasonicator, Weighing Balance, oven

Chemicals:

distilled water to make solution preparations, Precursor: zinc acetate dihydrate ($Zn(CH_3COO)_2 \cdot 2H_2O$), Reactant: sodium hydroxide (NaOH)

Procedure:

Amount of chemicals used-Zinc acetate molecular weight=219.50g

Sodium hydroxide molecular weight=39.9g/mol

$$\text{Zn acetate} = \frac{.2\text{molar} * \text{molecular weight of zn acetate} * \text{dist.water taken}}{1000}$$

$$\text{NaOH} = \frac{.4\text{ molar} * \text{molecular weight of NaOH} * \text{dist.water taken}}{1000}$$

Zn acetate= $\frac{.2*219.5*50}{1000}$ = 2.2g

NaOH= $\frac{.4*39.9*50}{1000}$ = 0.8g

weighing balance

fig(3.21)

As we got the ratio of concentrations, we are now taking the amount of zinc acetate and NaOH as mentioned above by using the Weighing balance.

Weighing balance: A precise weighing balance is essential for precisely measuring the amounts of reagents used in the extraction of ZnO nanoparticles. The process is simple, taking the desired amount of the above-mentioned ratio of chemicals by placing chemicals in a piece of paper(paper weight is removed by error-correcting in balance). the amount of chemicals needed is shown in balance.

50ml of dist. water is taken in 2 beakers and the chemicals of above mentioned gram are added in these 2 beakers. now we get 2 solutions one is sol of zinc acetate in 50ml dist. water and NaOH sol in 50ml dist. water. now take zinc acetate sol for magnetic stirring for about 45 minutes.

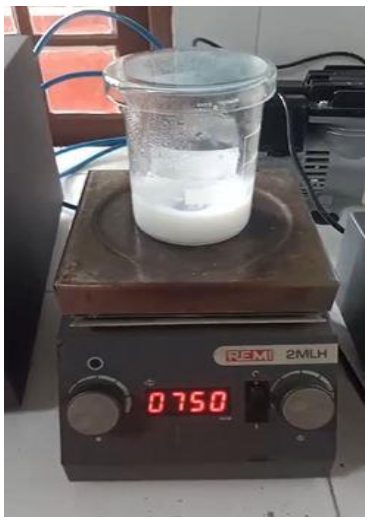
magnetic stirrer: One common lab tool for stirring or mixing liquids or solutions is a magnetic stirrer. It is composed of a magnetic stir plate that is positioned beneath the container holding the liquid to be stirred and a magnetic stir bar, sometimes referred to as a flea or stir disc, that is placed inside the container. The magnetic stir bar spins and agitates the liquid when the stir plate is activated, creating a revolving magnetic field. **Stir Plate**, A revolving magnetic field generator is located at the magnetic stirrer's base. Usually, it has controls to change the rotational speed. A tiny **magnetic bar** or disc inserted into the liquid to be stirred is called a magnetic stir bar. To stop reactions with the solution, it is typically coated with a chemically inert substance like PTFE (Teflon).

Beaker or Container:The container holding the liquid that needs to be agitated. Usually, glass or a plastic that resists chemicals are used to make it..

magnetic stirrer Take another beaker of NaOH sol in 50 ml dist.water and place it in another stirrer and stir it for 15 min.(both in 300-400 rpm)range. Finally add naoh sol to zn acetate that continues its stirring .then keep them both mixing for about half an hour(magnetic stirrer). The solution will turn to turn to white colour.

fig(3.22)

White color precipitate formed



fig(3.23)

Sonicator- An apparatus called a sonicator is used in labs to sonicate samples, which is the process of subjecting them to ultrasonic vibrations. Applications for this process include homogenizing, dispersing, emulsifying, mixing, and breaking up cells. A generator that generates high-frequency electrical signals, a transducer that translates the signals into mechanical vibrations, and a probe or horn that transfers the vibrations to the sample are the usual components of sonicators.

The solution that had changed its color is now placed in a sonicator for about half an hour to sonicate samples.

Note: According to the number of samples required these processes are continued 2,3 times. so we obtained 3 beakers of samples in low quantities for our use.

Centrifugate: A centrifuge is a device that uses the medium's viscosity, rotor speed, density, size, and shape to separate particles from a solution. It is frequently used in scientific and medical labs for a variety of tasks, including purifying samples, isolating DNA, and separating blood components.

Usually, the device is just a rotating rotor housed in a closed container. Depending on the particular application, lighter particles either stay suspended or rise to the top while heavier particles are pushed to the bottom by centrifugal force when the rotor spins at high speeds.

Regardless of the size of the centrifuge rotor, its RPM (revolutions per minute) indicates its speed of rotation. The force applied to the rotor's contents, on the other hand, is referred to as G-force (or RCF, or relative centrifugal force). It is a measurement of the acceleration that particles in the centrifuge experience and is based on the rotor's radius and revolution per minute.

We took out our sample and added it to the centrifuge tube filling about 13ml sol in each of 6 tubes and placed in the centrifuge apparatus. For 10 min we rotated at 3000-5000rpm and took out a sample, the sample as observed shows the sedimented particles at the bottom of the tube and clear water on top of the tube, repeat the centrifuge process by removing the water from the tube and refilling the same amount distilled water into the tube. We repeated the process of centrifugating and the total no of times centrifugated is 3 times. Each sol is done by this process and the complete solutions are centrifugated. the sedimented paste is collected using a spatula and the water is removed. It was a difficult process to take out the paste so we mixed drops of dist. water and then took them out. the paste is subjected to heating in high temperatures using an oven(laboratory).

Centrifuge apparatus

fig(3.24)

Oven: A piece of equipment used in scientific and industrial settings for material drying, sterilization, and curing is a laboratory oven. It offers a temperature-controlled, frequently humidity-controlled environment for sample or material processing.

These ovens are available in a variety of sizes, from large walk-in ovens for industrial use to benchtop models ideal for small-scale experiments. Usually, they consist of a chamber or cavity to hold the samples, heated components, and a temperature control system to manage the chamber's temperature.

Drying: Getting away moisture from samples. it's the use of oven heating in this process. We heat the paste and the moisture is removed leaving just a coat of white color that is thickened and sticks to the Petri dish.

The process is like this, we place the Petri dish containing the paste of the sample in an oven and dry it at 75 degrees Celsius for about 8 hours. Once the moisture is gone we get a white-colored hardened paste, we then scratch the surface of the Petri dish to remove the substance into powdered form. then we grind the powder to make it a fine powder without any hard substance.it is done using mortar and pestle.

Mortar and pestle- apparatus is used to grind the sample and make it fine powder and this is done manually. before inserting the sample into the mortar, the apparatus is cleaned using acetone.

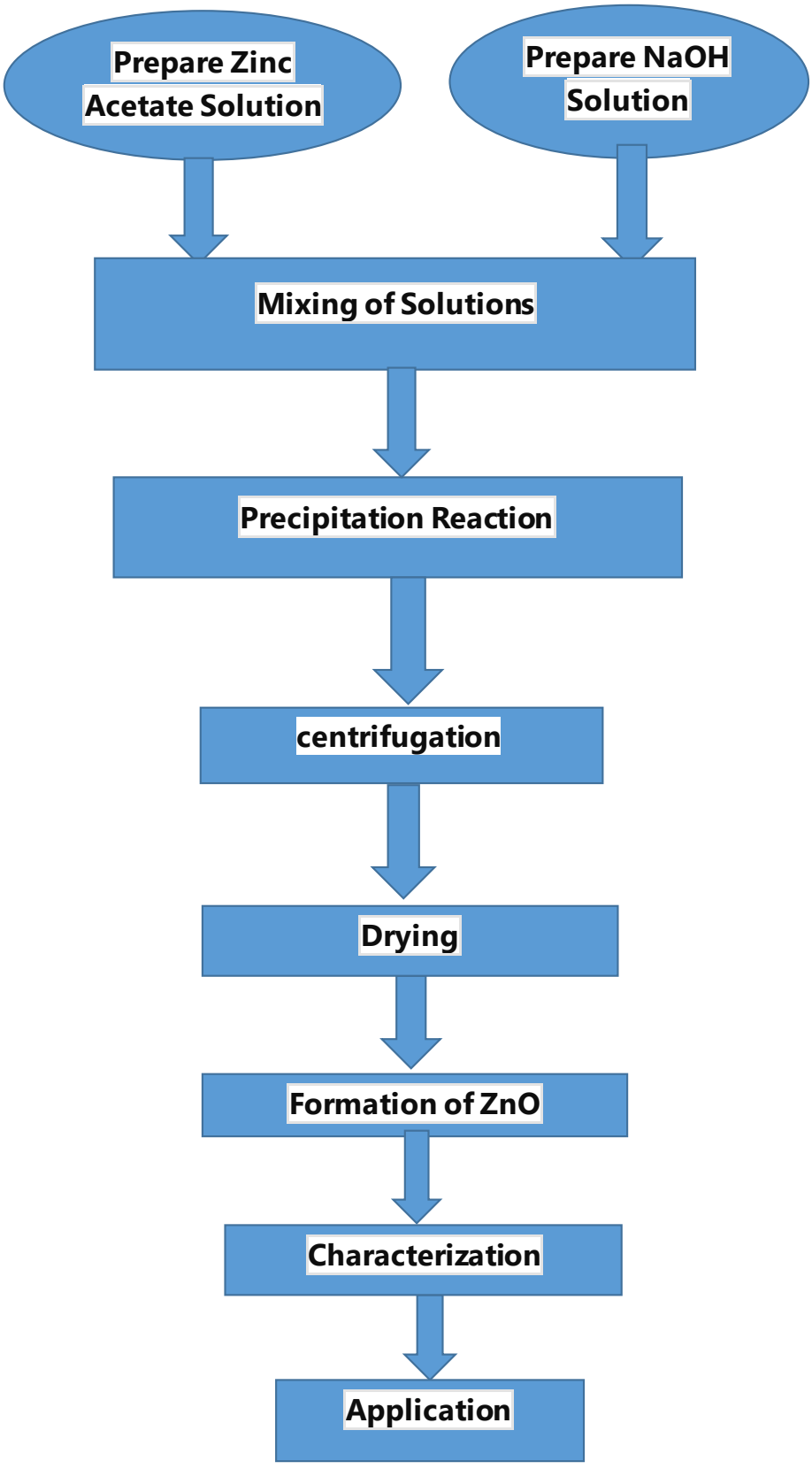
Propanone is another name for acetone, which is a colorless liquid. At certain concentrations, it is considered generally recognized as safe (GRAS) due to its low toxicity

and high flammability. This substance works well as a solvent in many home and industrial applications because it evaporates fast and mixes easily with water. Because of its high solvency, it is used as a cleaning agent to get rid of residue from glassware and other lab equipment.

Once the setup is completely cleaned using acetone, we put the sample into mortar by scratching them from the Petri dish. using a pestle manually we grind them making them into fine powder. now we store the powdered sample in an air-tight container. we then subject the sample to various tests like XRD(X-ray diffraction analysis), UV, Scanning electron microscopy (SEM) , and Fourier Transform Infrared Spectroscopy(FTIR).

*__The ZnO nanoparticle is extracted__.

Flowchart


Prepare Zinc Acetate Solution
Prepare NaOH Solution
Mixing of Solutions
Precipitation Reaction
centrifugation
Drying
Formation of ZnO
Characterization
Application

Chapter 4

Experiment on Nanoparticle Toxicity in plants

The possible negative effects of nanoparticles on biological systems, such as people, animals, and the environment, are investigated in research on nanoparticle toxicity. To experiment we use simple apparatus we use tubes, normal water, plants, and samples.

The sample is taken from low to high concentrations in different tubes and the solution is where the plant is grown as the medium. we use aquatic plants for this purpose. we observe the resultant growth or destruction of the plant. hence concluding the toxicity of nanoparticles in living cells. the concentrations of ZnO nanoparticles were taken in 6 different tubes with concentrations 0.019g,0.064g,0.22g,0.140g,0.26g, and (normal water) without nanoparticle.

Here we observe the effect in plants and find the optimum concentration where the effect is maximum. the plant used is the azola plant. The genus Azolla includes the small aquatic fern known as the "Azolla plant," or just Azolla. Because of its ability to quickly cover the surface of bodies of water, it is commonly referred to as "mosquito fern" or "duckweed fern." This is because it prevents mosquito breeding and provides habitat for waterfowl.

I took an azola plant for the test by placing healthy 1 plant in each tube (6). Take normal water in a tube of 13 ml and add the ZnO power to it. make the sol with different quantities of samples and test the growth or retardation rate in plants.

Toxicity testing

fig(4.1)

fig(4.2)

fig(4.3)





fig(4.4) **fig(4.5)** **fig(4.6)**

4.1 Result Of Toxicity Of Nanoparticle

Concentration(g)	Day			
	1	4	7	15
Normal water Without conc.	Plant is healhy	Plant is healthy	New growth	Slight brown color at the end of plant
.019	Healthy plant	Healthy plant	New growth,ends are slightly brown	Half healthy,rest are slightly brown
.026	Healthy plant	1/4th of the plant turned brown	New growth,but there is brown	Same as observed in day 7th
.064	Healthy plant	1/4th of the plant turned brown	½ of the plant is brown	3/4th of the plant turned brown
.140	Slightly brown at the ends	Half brown and half green	3/4th portion is brown	Full brown
.22	Slightly brown at corner	Half brown and half green	3/4th portion is brown	Full brown(dark color)

We conclude that the experimented concentrations in which the maximum potential of destruction in the plant is observed in .140g is a similar effect of .22 but is less concentrated compared to .22.

Few studies have examined the toxicity of biogenic metallic nanoparticles, even though a large variety of them have been studied, The synthesis and characterization of biogenically synthesized metal oxide nanoparticles and its thorough analysis of the toxicity of metal oxide nanoparticles to humans and the environment is necessary before developing applications utilizing these nanoparticles, which can be produced using classical or biogenic methods.

[8]

Chapter 5

Result of Synthesis

5.1 X-Ray Diffraction (XRD)

A potent analytical method for determining a material's crystal structure is X-ray diffraction (XRD). It is extensively utilized in many scientific fields, such as geology, chemistry, materials science, physics, and archaeology. This is how it operates:

Principle: X-ray diffraction, which happens when X-rays interact with a material's regular atomic arrangement (crystalline structure), is the basis for XRD. Atoms in a crystal lattice are scattered in various directions when X-rays strike them. A diffraction pattern containing details about the crystal structure is produced when the scattered X-rays interfere either constructively or destructively.

Experimental Setup: An X-ray tube is usually used to generate monochromatic X-rays, which are then used to irradiate a sample in an XRD experiment. Various angles are used to direct the X-rays onto the sample. The wavelength of the incident X-rays and the spacing between the lattice planes determine the specific angles at which the atoms in the sample diffract as a result of the X-rays' interaction with the crystal lattice.

Diffraction Pattern: A detector, such as a digital or photographic film, gathers and identifies the diffracted X-rays. The angles at which the crystal lattice diffracted X-rays are represented by a sequence of peaks in the resulting diffraction pattern. The position, width, and intensity of each peak reveal details about the crystal structure, such as phase composition, lattice parameters, and crystal symmetry.

All things considered, X-ray diffraction is a strong and adaptable method for examining the atomic and molecular structure of crystalline materials, offering insightful knowledge about their characteristics and behavior.

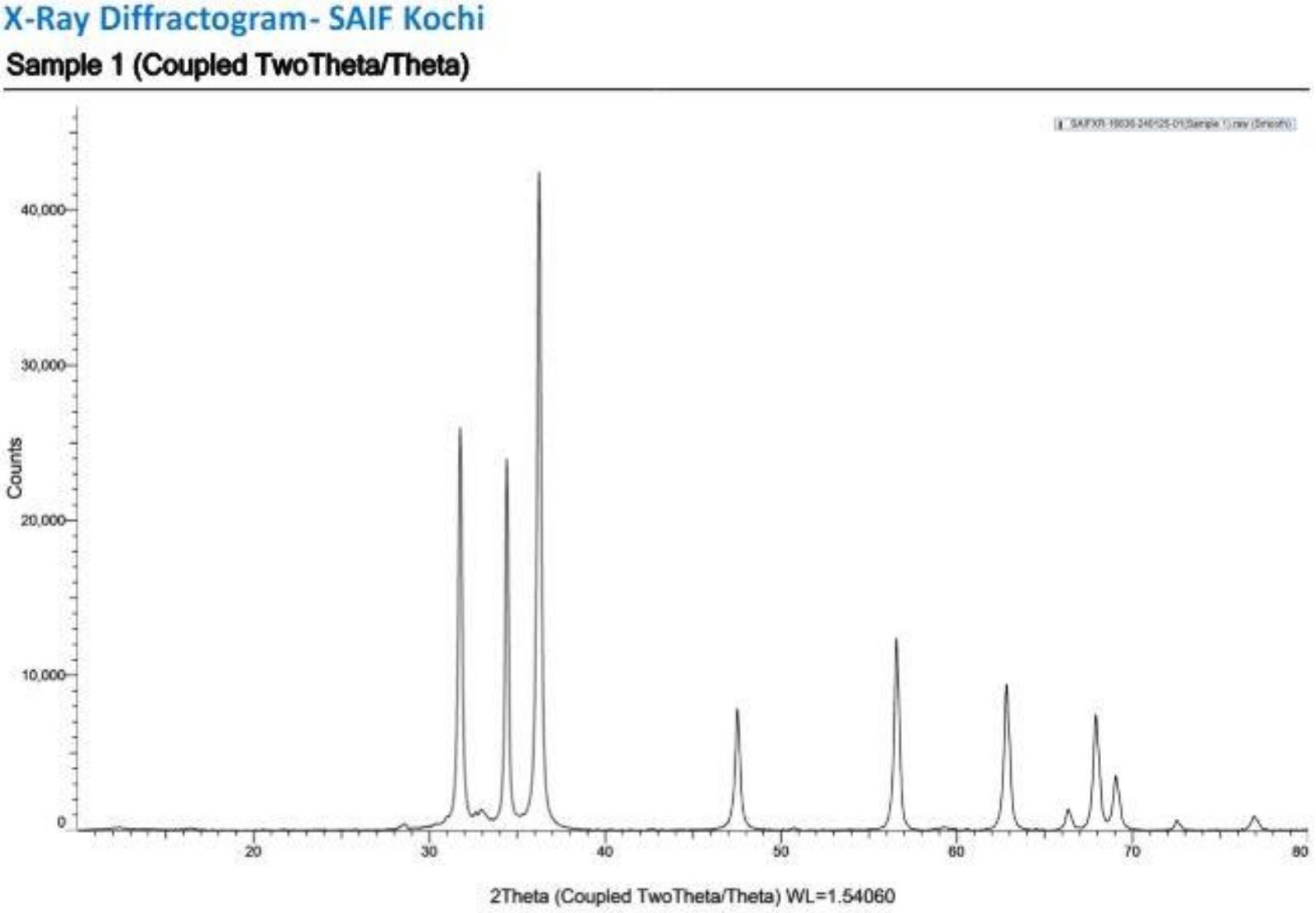
Data: The XRD plot of the pure-ZnO sample contained peaks at 2Θ of 31. 75, 34.35, 36.25, 47.50 and 56.60 in previous experiments. the obtained peak for ZnO particle was almost similar ,hence it Shows there exists impurity free nanoparticle . peaks obtained were 31.761, 34.414, 36.249, 47.535, 56.582.

Debye-Scherrer formula ,

$d = 0.89 \lambda / \beta \cos \theta$

Using the Debye-Scherrer formula, the diameter of the synthesized ZnO nanoparticle was determined. The sample's average particle size was determined to be 33.788 nm ((35.37+41.98+33.59+29.35+28.65)/5)using the Scherrer formula, which takes into account the FWHM of more intense peaks, λ (X-ray wavelength), θ (Bragg diffraction angle), and β (full width at half maximum) (FWHM) of the diffraction peak that corresponds to the plane.

The obtained size is like that of done in previous experimens .hence,synthesised particle is said to be nanoparticle.


X-Ray Diffractogram- SAIF Kochi
Sample 1 (Coupled TwoTheta/Theta)
Counts
40,000
30,000
20,000
10,000
0
20
30
40
50
60
70
80
2Theta (Coupled TwoTheta/Theta) WL=1.54060

fig(5.11)

2Theta

Sample ID	Sample 1
File Name	SAIFXR-16536-240125-01(Sample 1).raw (Smooth)
Scan Type	Coupled TwoTheta/Theta
Scan Mode	Continuousscan
Start	10.000 °
End	80.002 °
Step Size	0.020 °
Total Time/Step	57.60 s
Temperature	25 °C (Room)
Goniometer Radius	200.5 mm
Sample Rotation	15.000 1/min
Anode	Cu
ka2 Ratio	0.50000
Wavelength for display	1.54060 Å
Generator kV	40.0 kV
Generator mA	35.0 mA
Detector Opening	2.944 °
Primary Soller slit	2.500 °
Secondary Soller slit	
Air-Scatter Screen Mode	Automatic
Divergence Slit	15.000 mm
Antiscatter Slit	18.000
Slit Mode	Variable
Compute Crystallinity	No
%-Crystallinity	
%-Amorphous	
Global Area	

fig(5.12)

Peak List #1

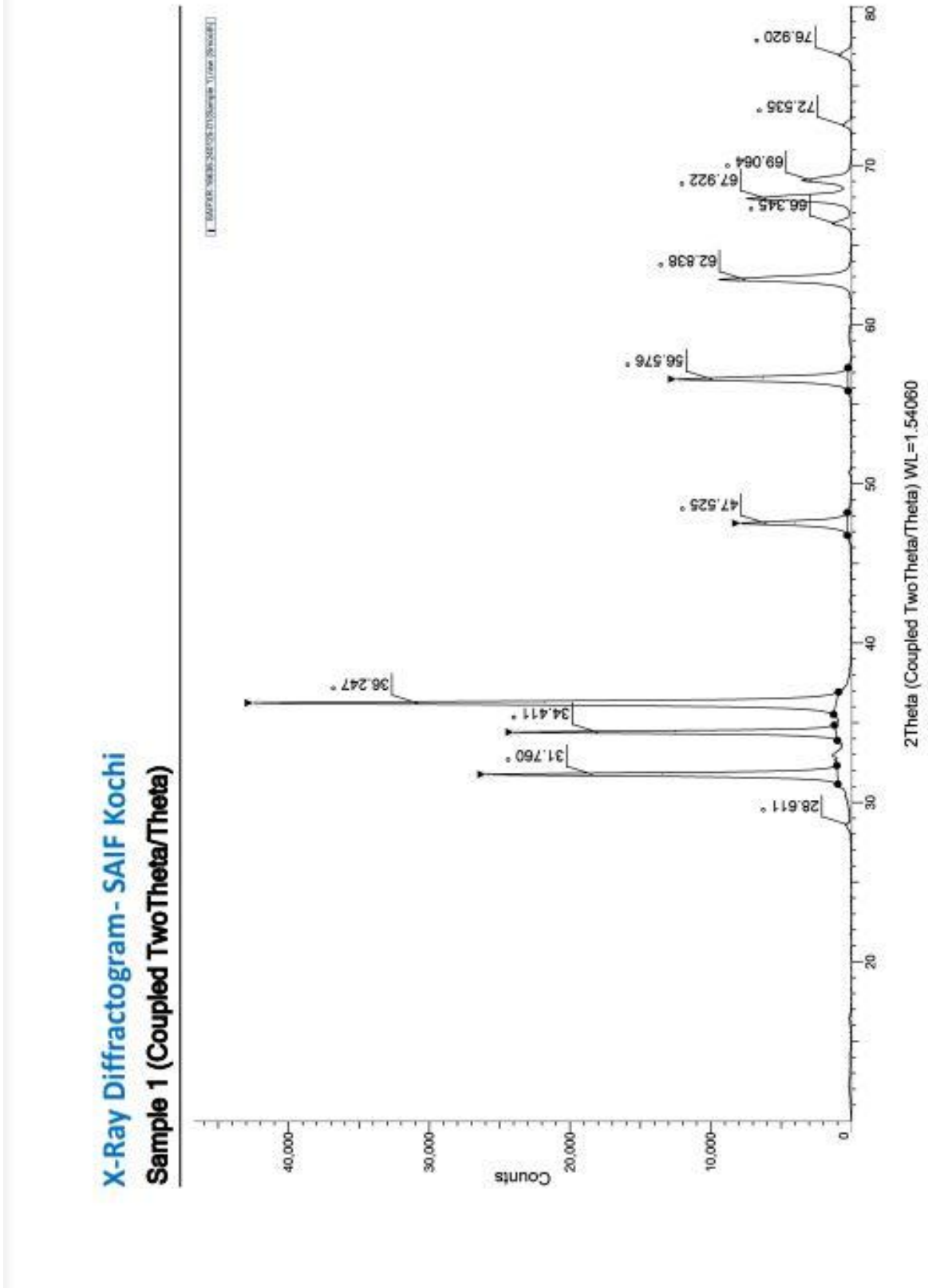
Angle	Net Intensity	d Value	Gross Intensity	Rel. Intensity
28.611 °	301.325	3.11746 Å	2020.30	1.0%
31.760 °	18424.7	2.81515 Å	20298.1	59.7%
34.411 °	18032.0	2.60409 Å	19965.7	58.4%
36.247 °	30854.0	2.47633 Å	32792.0	100.0%
47.525 °	6048.32	1.91166 Å	7856.62	19.6%
56.576 °	9926.37	1.62542 Å	11782.3	32.2%
62.838 °	7581.76	1.47768 Å	9413.66	24.6%
66.345 °	1164.12	1.40781 Å	2935.95	3.8%
67.922 °	6083.96	1.37891 Å	7820.19	19.7%
69.064 °	2836.52	1.35887 Å	4532.88	9.2%
72.535 °	578.131	1.30216 Å	2138.33	1.9%
76.920 °	763.524	1.23849 Å	2271.06	2.5%

Area List #1

Scan	Obs. Max	d (Obs. Max)	Gross Int.	FWHM	I. Breadth	Gravity C.	d (Gravity C.)
SAIFXR-16636-240125-01(Sample 1).raw (Smooth) #1	31.761 °	2.81510 Å	482.130	0.244	0.292	31.754 °	2.81570 Å
SAIFXR-16636-240125-01(Sample 1).raw (Smooth) #1	34.414 °	2.60389 Å	448.316	0.207	0.243	34.408 °	2.60435 Å
SAIFXR-16636-240125-01(Sample 1).raw (Smooth) #1	36.249 °	2.47616 Å	770.209	0.260	0.315	36.241 °	2.47672 Å
SAIFXR-16636-240125-01(Sample 1).raw (Smooth) #1	47.535 °	1.91129 Å	166.900	0.309	0.375	47.529 °	1.91152 Å
SAIFXR-16636-240125-01(Sample 1).raw (Smooth) #1	56.582 °	1.62528 Å	246.611	0.329	0.378	56.589 °	1.62509 Å

Raw Area	Net Area	C. Size	K	Instr. Width
184.03	126.04	375.5 Å	0.890	0.050
146.85	96.291	446.5 Å	0.890	0.050
300.30	226.24	357.0 Å	0.890	0.050
100.82	48.979	311.7 Å	0.890	0.050
132.09	79.398	305.0 Å	0.890	0.050

fig(5.13)


X-Ray Diffractogram - SAIF Kochi
Sample 1 (Coupled TwoTheta/Theta)
Counts
40,000
30,000
20,000
10,000
0
20
30
40
50
60
70
80
28.611°
31.760°
34.411°
36.247°
47.525°
56.576°
62.838°
66.345°
67.922°
69.064°
72.535°
76.920°
2Theta (Coupled TwoTheta/Theta) WL=1.54060

fig(5.14)

5.2 UV Testing

The term "UV testing" describes a variety of methods used to evaluate how ultraviolet (UV) radiation affects surfaces, materials, and products. UV testing is essential in many industries to assess the performance, longevity, and mechanisms of material degradation when exposed to artificial UV radiation sources or sunlight.Tests for UV absorption and transmission quantify a material's capacity to either absorb or transmit UV radiation. UV transmittance, UV absorbance, and optical characteristics are measured and the UV spectrum of materials is analyzed using spectrophotometers, also known as UV-Vis-NIR spectrometers. Sun protection products, UV-blocking coatings, and UV-resistant materials must be designed with this information in mind.

All things considered, UV testing is essential to comprehending how UV radiation affects materials and to creating products with improved UV resistance, longevity, and durability. It enhances product performance and customer satisfaction by reducing the negative effects of UV exposure, such as deterioration, discoloration, and loss of mechanical properties.

fig(5.21)

Experiment Information

Title: Untitled-1
Comment:
Instrument Serial No.: 365K21011906
Software Version: UV Express - Version 4.1.3

Experimental Date: Mar 11 2024 13:53:46 (GMT +5:30)
System Name: Undefined
Firmware Version: 160529

Method

Experiment Type : Scan Setup

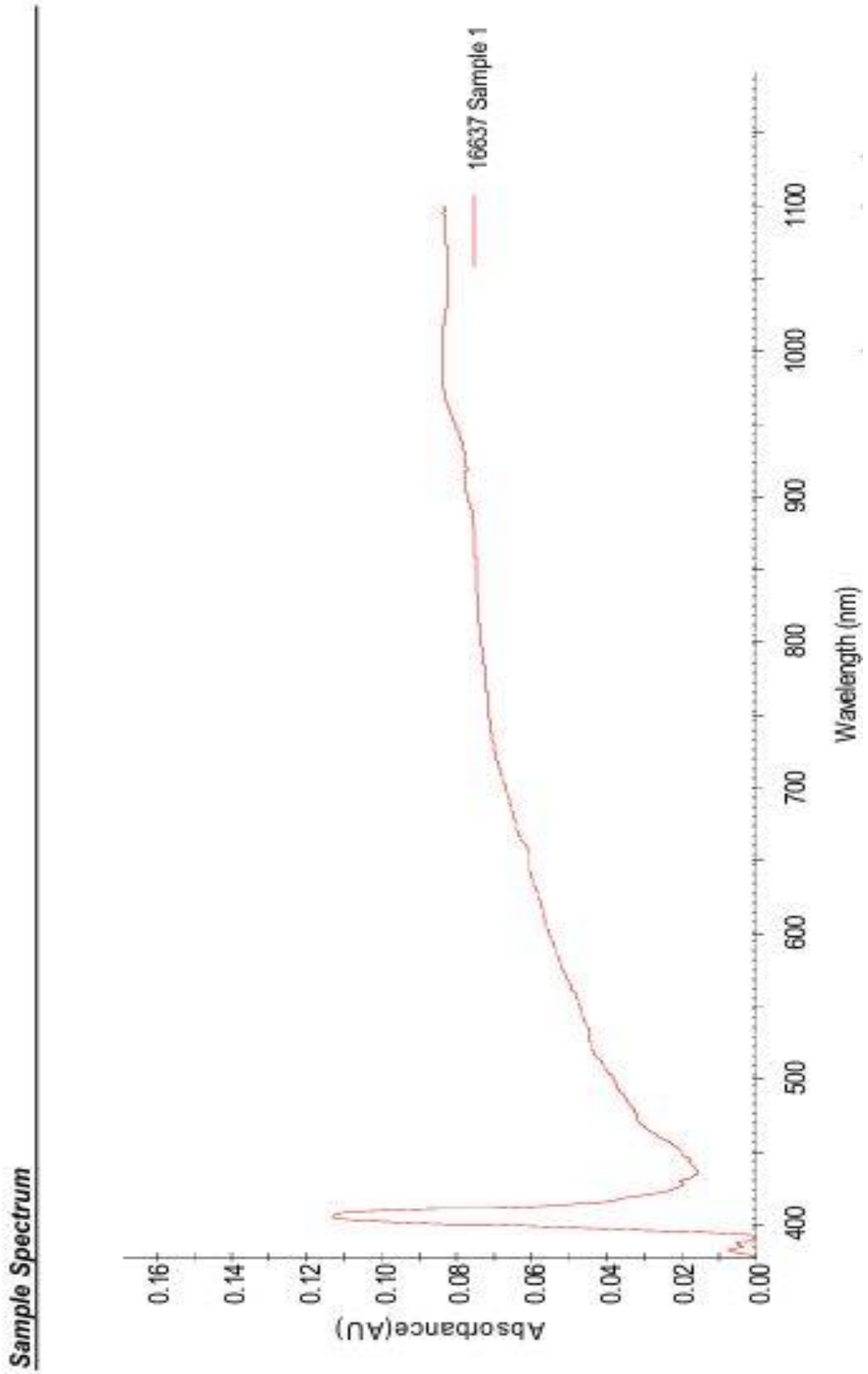
Experiment Setup	Internal Reference	Scan Setup
Data Type: Absorbance Spectra No.: 1 Measurement Range (nm): 1100~250 Data Interval (nm): 1.0 0%T / Blocked Beam Baseline : No SBW (nm): 5.0 Beam Type: Double Normal Lamp: UV+VIS Lamp Change (nm): 400 Accessory : Integrating Sphere	Use: No	Monitoring Wavelength (nm): 560, 550, 540, 430, 400

Result Data

Name	AU(560.00nm)	AU(550.00nm)	AU(540.00nm)	AU(430.00nm)	AU(400.00nm)
16637 Sample 1	0	0	0	0	0.1

Spectrum List

Name	Date
16637 Sample 1	Mar 11 2024 14:01:00 (GMT +5:30)


Sample Spectrum
16637 Sample 1
Absorbance(AU)
0.16
0.14
0.12
0.10
0.08
0.06
0.04
0.02
0.00
400
500
600
700
800
900
1000
1100
Wavelength (nm)

fig(5.22)

User Information

Name:

Experiment Information

Title: Untitled-1
Comment:
Instrument Serial No.: 365K21011906
Software Version: UV Express - Version 4.1.3

Experimental Date: Mar 11 2024 13:53:46 (GMT +5:30)
System Name: Undefined
Firmware Version: 160529

Method

Experiment Type : Scan Setup

Experiment Setup	Internal Reference	Scan Setup
Data Type: Reflectance Spectra No.: 1 Measurement Range (nm): 1100~250 Data Interval (nm): 1.0 0%T / Blocked Beam Baseline : No SBW (nm): 5.0 Beam Type: Double Normal Lamp: UV+VIS Lamp Change (nm): 400 Accessory : Integrating Sphere	Use: No	Monitoring Wavelength (nm): 560, 550, 540, 430, 400

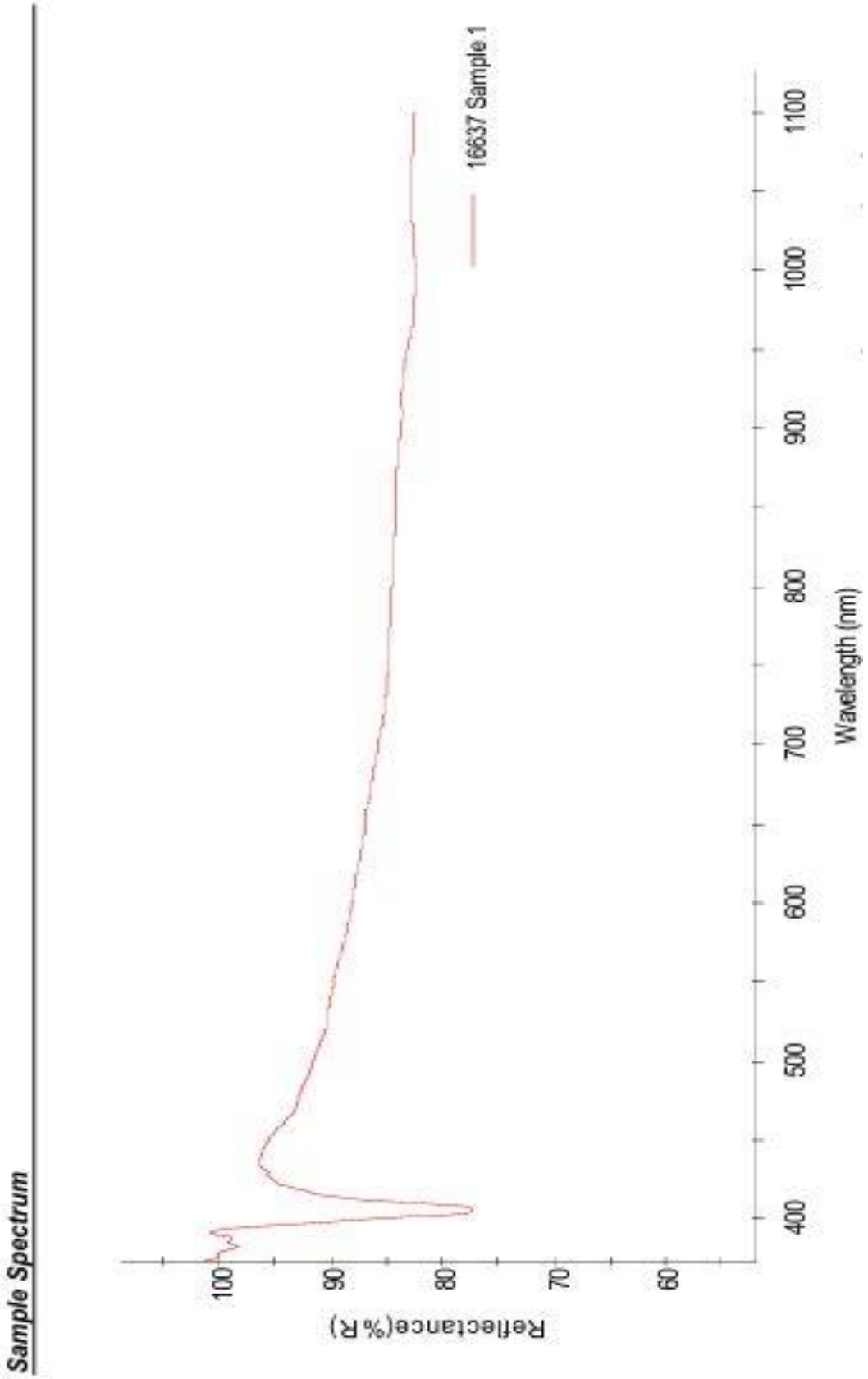
Result Data

Name	%R(560.00nm)	%R(550.00nm)	%R(540.00nm)	%R(430.00nm)	%R(400.00nm)
16637 Sample 1	89.4	89.7	90	95.5	85.9

Spectrum List

Name	Date
16637 Sample 1	Mar 11 2024 14:01:00 (GMT +5:30)

fig(5.23)


Sample Spectrum
16637 Sample 1
Reflectance(% R)
100
90
80
70
60
400
500
600
700
800
900
1000
1100
Wavelength (nm)

fig(5.24)

5.3 Field Emission Scanning Electron Microscopy(FESEM)

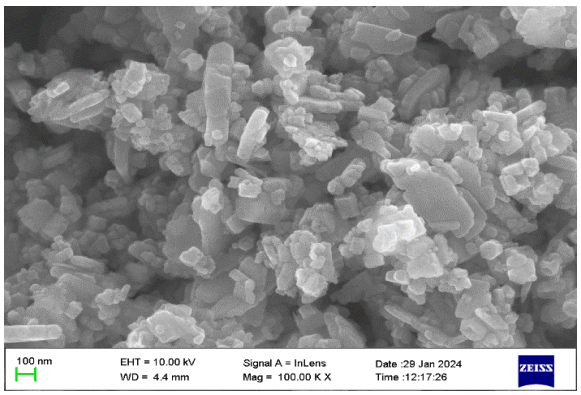
An advanced microscopy method called a scanning electron microscope (SEM) is used to create high-resolution images of a sample's surface.

Principle of Operation: An electron beam that is focused is scanned over a sample's surface by a SEM. Different signals are produced by the interaction between the electron beam and the sample's atoms, such as backscattered electrons, secondary electrons, and distinctive X-rays. The topography, composition, and other surface characteristics of the sample are all revealed by these signals.

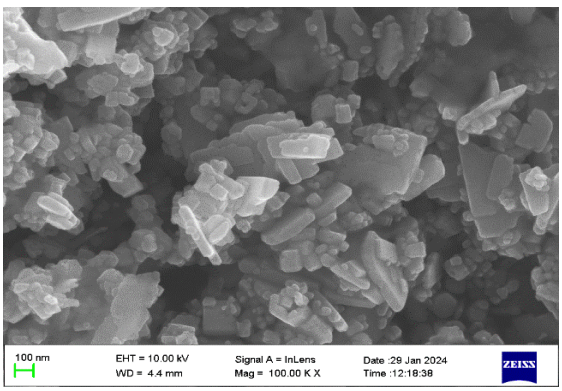
High-resolution surface images can be obtained in microscopy with the use of the FESEM technique. It is especially helpful for researching the morphology and topography of materials at the nanoscale. To detect the signals produced by the interaction between the electrons and the sample surface, a focused electron beam is scanned across the surface of the sample using a FESEM. Depending on the capabilities of the instrument, this technique allows for detailed imaging at magnifications ranging from 10x to over 1,000,000x. Many scientific and industrial domains, such as materials science, nanotechnology, biology, and semiconductor research, make extensive use of FESEM.

Data: shape and structure of nanoparticle: By analyzing SEM TEST images we could know the structure, and shape of minute particles. we here considered it as a mixture of rod-like and spherical forms. down below shows a pillar-like structure of the ZnO nanoparticle obtained.

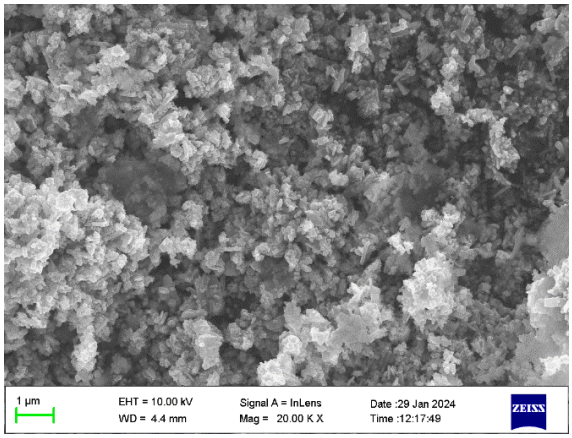
FESEM images



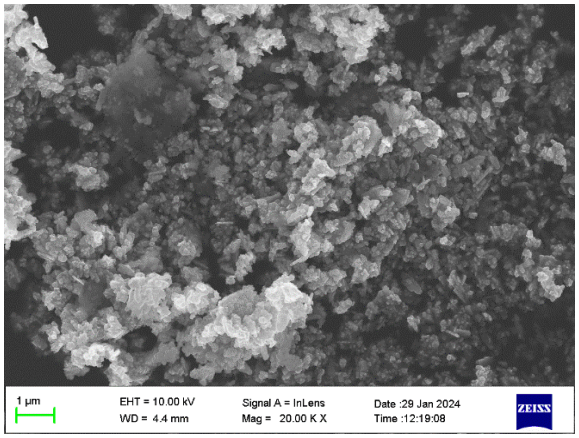
fig(5.31)



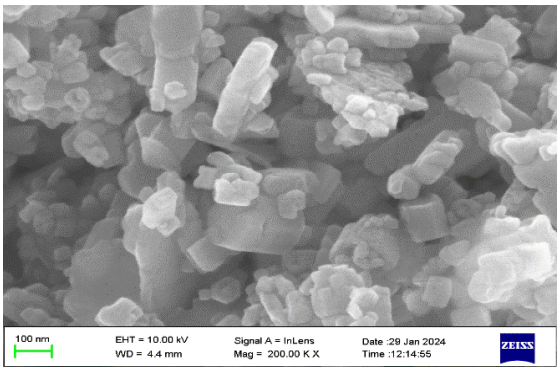
fig(5.32)



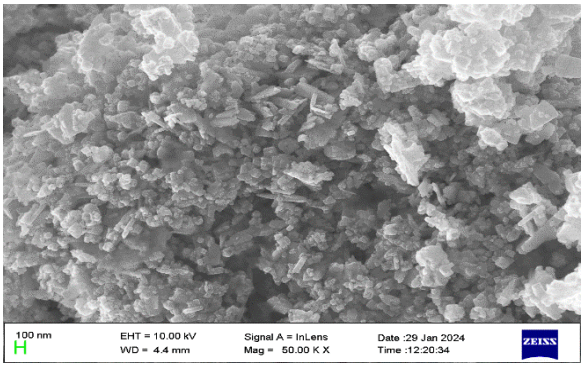
fig(5.33)



fig(5.34)



fig(5.35)



fig(5.36)

5.4 Fourier-Transform Infrared Spectroscopy (FTIR)

FTIR testing- Fourier Transform Infrared Spectroscopy is referred to as FTIR. Based on how well chemical compounds absorb infrared light, this analytical method is highly effective in identifying and characterizing them. FTIR spectroscopy measures the amount of light absorbed at various wavelengths by passing infrared radiation through a sample. Every compound has a different absorption pattern, which reveals important details about the molecular makeup and functional groups of the substance.

The infrared beam in the FTIR instrument is divided into two sections using an interferometer. While the other part acts as a reference, the first part goes through the sample. The sample's infrared spectrum is then obtained by recombining the beams and analyzing the resulting interference pattern using Fourier transform techniques.

Specific vibrational modes of the chemical bonds present in the sample are represented by peaks in the spectrum. Analysts can determine which functional groups and chemical bonds are present in the sample by comparing these peaks to databases or reference spectra.

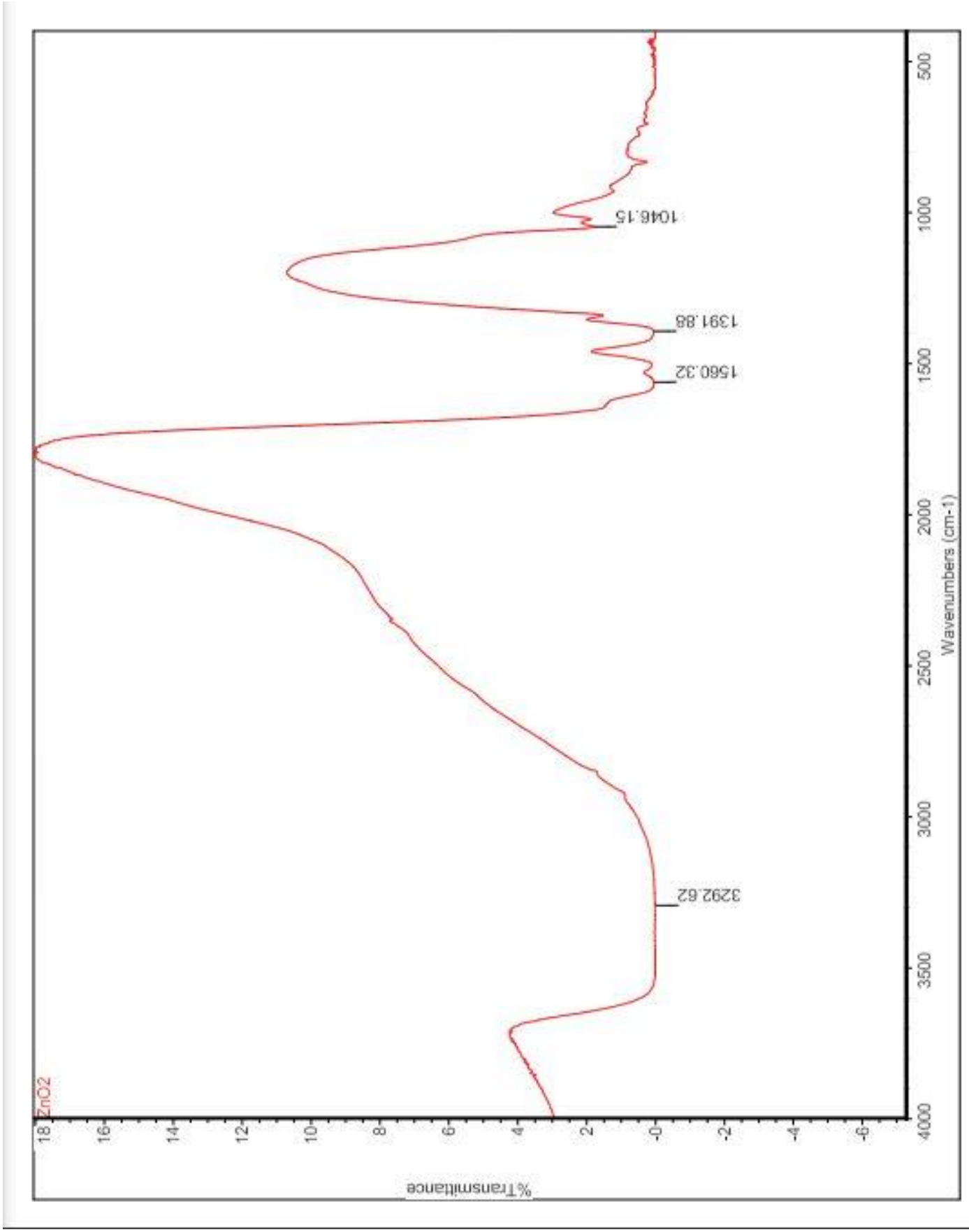
Pellet **hydraulic presser**

fig(5.41) **fig(5.42)**

then, hydrolic pressed the powder in(10mm diameter),pellet made by pressing in 11 pascal.we added the pellet in the testing machine(ftir) and the system (computer)shows the graphical representation of the sample, test is taken.

Other tests are done like uv, XRD,…

FTIR, or Fourier Transform Infrared Spectroscopy, is a potent analytical method that uses a chemical compound's absorption of infrared light to identify and describe it.


ZnO2
%Transmittance
Wavenumbers (cm-1)
3292.62
1560.32
1391.88
1046.15

fig(5.43)

Chapter 6

Conclusion

We obtained nanoparticles through a chemical method. subjected ZnO nanoparticle to various tests followed by conducting an application of it, that is the toxicity in plants. the toxicity could give us the maximum effect at low concentrations, similar to high concentrations.

As the whole process, finally get to know more about nanoparticles, but much more about ZnO nanoparticles.

Thus we conclude our project with our experimentation and observation of this particle through XRD, SEM, FTIR, and UV studies to confirm the nanostructure for the prepared ZnO nanoparticles.

Chapter 7

Reference

[1] Khan I, Saeed K, Khan I (November 2019). "Nanoparticles: Properties, applications and toxicities". Arabian Journal of Chemistry. 12 (7): 908–931. doi:10.1016/j.arabjc.2017.05.011

[2] Klingshirn C (April 2007). "ZnO: material, physics and applications". ChemPhysChem. 8 (6): 782–803. doi:10.1002/cphc.200700002. PMID 17429819.

[3] Wiberg E, Holleman AF (2001). Inorganic Chemistry. Elsevier. ISBN 978-0-12-352651-9.

[4] Greenwood NN, Earnshaw A (1997). Chemistry of the Elements (2nd ed.). Butterworth-Heinemann. ISBN 978-0-08-037941-8.

[5] Vitha, Mark F. (2018). "Chapter 2". Spectroscopy: principles and instrumentation. Hoboken, NJ: John Wiley & Sons. ISBN 978-1-119-43664-5.

[6] Skoog, Douglas A.; Holler, F. James; Crouch, Stanley R. (2007). Principles of Instrumental Analysis (6th ed.). Belmont, CA: Thomson Brooks/Cole. pp. 169–173. ISBN 978-0-495-01201-6.[dead link]

[7] R. S. Drago (1992). Physical Methods for Chemists, 2nd Edition. W. B. Saunders. ISBN 0030751764.

[8] Seabra AB, Durán N (June 2015). "Nanotoxicology of Metal Oxide Nanoparticles". Metals. 5 (2): 934–975. doi:10.3390/met5020934