



زانکۆی سه‌لاحه‌دین - هه‌ولێر
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Theoretical Study Green-Based ZnO Nanoparticle preparation and Their Characterization.

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بِسْمِ اللَّهِ الرَّحْمَنِ الرَّحِيمِ
قَالُوا سُبْحَانَكَ لَا عِلْمَ لَنَا إِلَّا مَا عَلَّمْتَنَا إِنَّكَ أَنْتَ الْعَلِيمُ الْحَكِيمُ
صدق الله العظيم

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Supervisor Certificate

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*This project is dedicated
to:*

*Allah Almighty, my
Creator and my Master,*

*My great teacher and
messenger, Mohammed
(May Allah bless and
grant him), who taught us
the purpose of life,*

*My homeland Kurdistan,
the warmest womb,*

The Salahadin University; my second magnificent home;

My great parents, who never stop giving of themselves in countless ways,

My beloved brothers and sisters;

To all my family, the symbol of love and giving,

my friends who encourage and support me,

All the people in my life who touch my heart.

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TABLE OF CONTENTS

ACKNOWLEDGEMENTS	iv
SUMMARY	6
CHAPTER 1. Introduction	
1.1 Introduction	7
1.2 Zinc oxide (ZnO) nanoparticles	8
CHAPTER 2. Material and Method	
2.1 Synthesis of ZnO Nanoparticles	11
2.2 Physical synthesis processes	3
2.3 Chemical synthesis processes	12
2.4 Biological Method	15
CHAPTER 3. Results and Discussion	
3.1 Microscopy Techniques	17
3.1.1 SEM	17
3.1.2 TEM	18
3.1.3 UV–Visible Spectrophotometry	19
3.1.4 Energy Dispersive X-Ray Spectroscopy	19
3.1.5 Xray Diffraction	20
3.2 Application of ZnO NPs.	22
3.3 Comparison Table.	24
CHAPTER 4. Conclusion and Future work	
4.1 Conclusion	25
REFERENCES	26

SUMMARY

Zinc oxide nanoparticles (ZnO NPs) are a popular metal oxide nanoparticle with numerous industrial and academic applications. To fulfill the rising demand for ZnO NPs, many synthesis methods have been used. The environmental and economic implications of most methods of synthesizing ZnO NPs have led to the search for more environmentally and economically beneficial alternatives. Biosynthesis employing plant sources has been proven to be suitable for producing ZnO NPs due to several health, environmental, economic, and therapeutic benefits. The unique properties of ZnO NPs generated from plant extracts have increased their use in agriculture for fertilizers, insecticides, and fumigants. They are used in the synthesis of disinfectants, antifungals, anticancer, antioxidants, anti-inflammatory, and anti-diabetic med. this review highlights the importance of zinc oxide as a multifunctional material with various preparation and modification methods and a wide range of applications in different industries. Further research and development in the field of zinc oxide are necessary to explore its full potential in various applications. it is important to consider the potential environmental and health impacts of the use of ZnO NPs in different applications. While they have been shown to have many benefits, there are also concerns about their toxicity and potential negative effects on human health and the environment. Therefore, it is crucial to continue researching and developing safe and sustainable methods of producing and using ZnO NPs, as well as to monitor their impact on the environment and human health. Overall, the use of ZnO NPs has the potential to significantly benefit various industries and improve human health, but it is important to balance these benefits with potential risks and ensure responsible use.

CHAPTER ONE

GENERAL INTRODUCTION

1.1 Introduction:

Nanoparticles are defined as ultrafine particles sized between 1 and 100 nanometers in diameter. In recent decades, there has been wide scientific research on the various uses of nanoparticles in construction, electronics, manufacturing, cosmetics, and medicine.(Mohajerani et al., 2019, Pushpalatha et al., 2022)

The concept of a “nanometer” was first proposed by Richard Zsigmondy, the 1925 Nobel Prize Laureate in chemistry. He coined the term nanometer explicitly for characterizing particle size and he was the first to measure the size of particles such as gold colloids using a microscope(Krukemeyer et al., 2015)

Modern nanotechnology was the brain child of Richard Feynman, the 1965 Nobel Prize Laureate in physics(Hulla et al., 2015)

One of the most promising aspects of nanotechnology, both now and in the future, is Nano medicine. Nanotechnology has the potential to revolutionize the way medicine is practiced and how humans approach their own healthcare. Nano medicine technologies currently being used include enhanced images for medical diagnostics, procedures for finding clogged arteries and detecting the early stages of Alzheimer’s disease, focused treatments for different forms of cancer, and the creation of artificial tissues to repair diseased organs and nerves. As with any new technology or field of study. (Thema et al., 2015). Nanotechnology is defined as the understanding and control of matter at dimensions between 1 and 100 nm (Hulla et al., 2015). Nanoparticles are not simple molecules and consist of three layers shown in Figure 1.1(Röhr, Sá and Konezny, 2019).

1.2 Zinc oxide (ZnO) nanoparticles

Zinc oxide (ZnO) nanoparticles have wide technological applications in catalytic, photocatalytic, electrical and optoelectronic processes and systems(Noman et al., 2019) For these diverse applications, size and morphology of ZnO particles are of primary consideration. Consequently, studies on the synthesis, characterization and properties of nanosized ZnO have received significant attention in the recent years. ZnO belongs to a family of metal oxide semiconductors. ZnO is an n-type semiconductor with wide band gap (3.37 eV) and having large exciton binding energy (60meV) (Adam, Pozina, Willander and Nur, 2018) Researchers have been concluded that the photocatalytic performance of nanomaterials significantly depends upon the synthesis route, size, shape, crystallinity and dimension. At present, different researchers have prepared ZnO nanostructures by different synthesis methods i.e., sol–gel, hydrothermal, co-precipitation, green, electrospinning and sonochemical(Bandeira et al., 2020)

The ZnO NPs occurring in a very rich variety of size and shape will provide a wide range of properties. The methods for stable ZnO NPs preparation have been widely developed in recent years, which mainly include the chemical precipitation method, sol-gel method, solid-state pyrolytic method, solution-free mechanochemical method, and biosynthesis method.(Agarwal et al., 2017)

ZnO NPs have exhibited promising biomedical applications based on its anticancer, antibacterial, antidiabetic, anti-inflammatory, drug delivery, as well as bioimaging activity. Due to inherent toxicity of ZnO NPs, they possess strong inhibition effects against cancerous cell and bacteria, by inducing intracellular ROS generation and activating apoptotic signaling pathway, which makes ZnO NPs a potential candidate as anticancer and antibacterial agents. In addition, ZnO NPs have also

been well known to promote the bioavailability of therapeutic drugs or biomolecules when functioning as drug carriers to achieve enhanced therapy efficiency. Moreover, with the ability to decrease blood glucose and increase in insulin levels, ZnO NPs have shown the promising potential in treating diabetes and attenuating its complications, which can be further evaluated.(Jiang et al., 2018)

The nanoparticles thus prepared were characterized by regarding scanning electron microscopy (SEM), X-ray diffraction (XRD) and (EDX) (Becheri, Dürr, Lo Nostro and Baglioni, 2007).

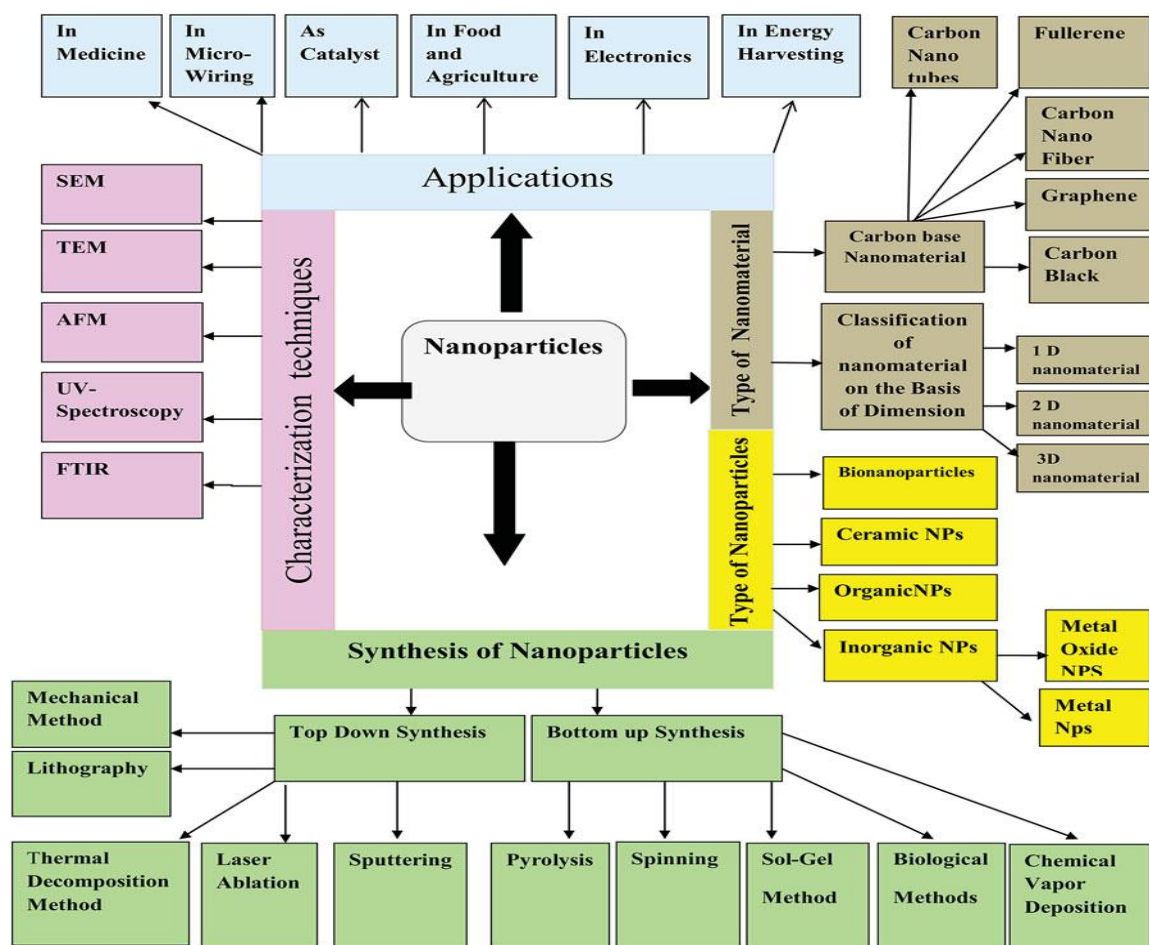


Figure 1.1 Application and Synthesis of Nanoparticles

Recently many researchers have shown that ZnO nanostructures are suitable materials for manufacturing a biosensor. There is ongoing research to observe the bio-imaging sensing purpose. It has shown positive results only on mice so far. The requirements of this sensor are non-allergenic, noncarcinogenic, high sensitivity, good reproducibility, and non-toxic characteristics. It can help in diagnosing the early stages of cancer germs. Moreover, sensing is important for monitoring and tracking the disease of a patient. (Jiang et al., 2018)

CHAPTER TWO

MATERIALS AND METHODS

2.1 Synthesis of ZnO Nanoparticles

ZnO nanoparticles have been developed by using several processes, such as physical, chemical, and biological synthesis processes. Physical and chemical processes are performed based on a thermodynamic and kinetic equilibrium approach. In physical and chemical synthesis processes, development rates of ZnO nanoparticles are very significant and mostly utilized for industrial advancement. Biological synthesis processes are performed through plants, fungi, algae, bacteria, etc. Large-scale productions of ZnO nanoparticles by plants show free additional impurities. The plant parts like fruits, seeds, stems, leaves, and roots have been used for nanoparticle development. Their extract is prosperous in photochemical, which can act as both stabilization and reducing agents (Thema et al., 2015).

ZnO NPs can be obtained using chemical, physical or biological methods. Chemical methods include precipitation, microemulsion, chemical reduction, sol-gel and hydrothermal techniques, which may lead to high energy consumption when high pressure or temperature conditions are required in the process. Among the chemical methods, the most commonly used is the sol-gel synthesis, developed by Spanhel and Anderson (1991), which uses a zinc precursor salt (nitrate, sulphate, chloride, etc.) and a chemical reagent in order to regulate the solution pH and avoid the precipitation of Zn (OH)₂. After, this solution will be exposed to thermal treatment under temperatures up to 1000 °C to obtain the ZnO NPs (Espitia et al., 2012). Chemical stabilizers, such as citrates or polymers like polyethylene glycols, polyvinylpyrrolidone and amphiphilic block copolymers, can be added to the ZnO NPs synthesis for controlling the size of the nanoparticles and avoid particle agglomeration. In addition, a significant factor to be considered on the chemical synthesis is that the concentration of the chemicals used in the process can influence the size and shape of the particles considerably. It is known that it is possible to

obtain particles from few nanometers (5-10 nm) up to micrometric size using the same process but different concentrations and ratios of chemicals. Although less used than the chemical method, ZnO NPs can be synthesized *via* physical techniques by vapor deposition, plasma and ultrasonic irradiation. Nonetheless, these techniques usually require a high amount of energy and robust equipment, which increases the cost of the products. Another approach to obtain ZnO NPs is using biological synthesis, which has arisen as an alternative eco-friendly process. The interest in synthesizing ZnO NPs *via* biological methods has increased considerably in the last decade. The development of this new approach and the significant interest in it is mainly related to the absence of toxic chemicals or high amount of energy applied to the biological synthesis, which makes the process more cost-effective and eco-friendlier (Thema et al., 2015).

2.2 Physical synthesis processes

Physical synthesis can be divided into high energy ball milling, physical/chemical vapor Physical synthesis can be divided into high energy ball milling, physical/chemical vapor deposition, and laser ablation processes. C. Prommalikit et al. reduced the particle size from micron to nano scale of ultrafine ZnO powder by using a high energy ball milling process. The authors used the starting material of commercial grade ZnO powder with an average size of 0.8 μm . They investigated the crystalline structure, surface morphology, and particle size of milled samples by using the X-ray diffractometer (XRD), scanning electron microscopy (SEM), and particle analyzer, respectively. According to XRD results, the authors suggested that the XRD pattern had indicated the hexagonal crystalline structure. The SEM images indicated the particle size of ZnO nanopowders distinctly decreased due to the increase of milling time and speed. After a milling process, particle size was found in ultrafine nanopowers in the range between 200

and 400 nm. These results indicated the commercial ZnO powders minimized the particle size with specific milling speed and time. Finally, the authors recommended that the milling parameters (speed and time) of this process showed a significant influence on the reduction of particle size (Thema et al., 2015). This process has been used in the production of ZnO NPs with diverse shapes like nanorods, nanobelts, nanocombs, and nanorings. ZnO NPs synthesized using this approach have potential photocatalytic activity which is recommendable for degradation. The physical methods suffer some limitations because they involve the usage of expensive equipment, large space for machine set up, and high pressure and temperature. (Ali et al., 2018)

2.3 Chemical synthesis processes

A variety of chemical synthesis processes can be used for the development of ZnO nanoparticles. The chemical synthesis processes can be generally divided into two categories, such as gas and liquid phase. The gas phase process can be divided into processes as spray pyrolysis and gas condensation. The liquid phase process can be divided into processes as precipitation or co-precipitation, colloidal, sol-gel, oil microemulsion, hydrothermal, and solvothermal. prepared the ZnO nanoparticles by using a spray pyrolysis process from zinc acetate dihydrate ($\text{Zn}(\text{CH}_3\text{COOH})_2 \cdot 2\text{H}_2\text{O}$) at a concentration of 0.5 M solution with distilled water. The solution was used as a precursor for spray by sonication into a vertical quartz reactor by being allowed to flow without a carrier gas in a 900°C furnace. The authors studied the optical sensing characteristics conducted by temperature-dependent PL measurements. They observed the low temperature PL spectra of the ZnO nanoparticles that displayed a strong exciton emission peak with multiple side band regions. In this process, other researchers synthesized the ZnO particles/nanoparticles, depending on various reactor temperatures, solution concentrations, and atomizing pressures. The ultraviolet-visible spectroscopy,

FTIR, XRD, SEM, and TEM characterization methods were used in their experiment. The authors recommended that all parameters showed a significant influence on particle sizes of synthesized materials. Their experimental results showed the particle size lay between 10 and 400 nm with uniform morphologies that indicated the wide distribution of particle size. In conclusion, the authors recommended that ZnO nanoparticles could enhance the photo catalytic efficiency and controlling the crystalline dimension by using this process. (Jiang et al., 2018)

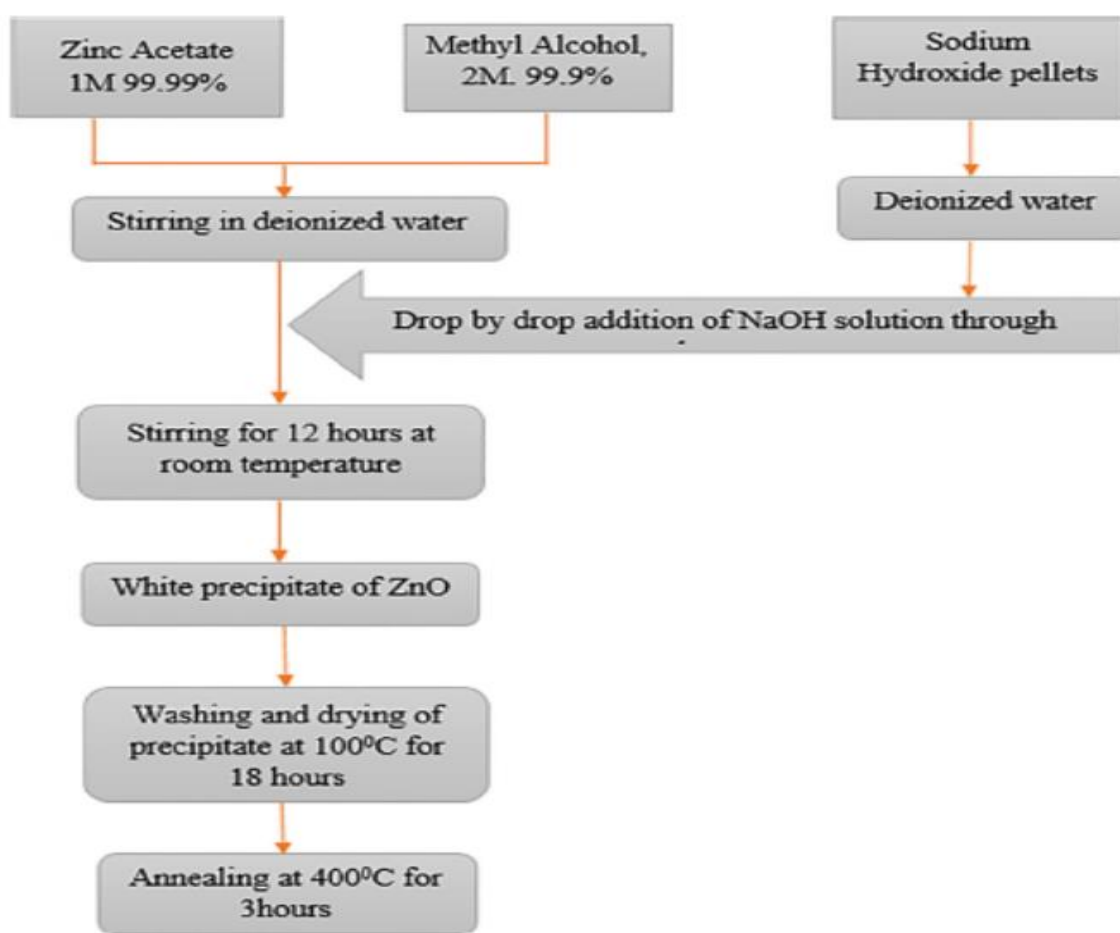


Figure 2.1 Flow chart illustrating the chemical method of synthesis of ZnO NPs

2.4 Biological Method

The biological method of synthesizing ZnO NPs is referred to as green synthesis or biosynthesis; this method involves the use of microorganisms such as algae, fungi, yeast, bacterial, and plant extracts as the reducing agent. Despite the advantages attached to the use of microorganism as reducing agent in the synthesis of ZnO NPs, the extreme safety required due to the toxicity of some microorganisms and challenges of incubation are the major problems. The high effectiveness of plant extracts in the synthesis of ZnO NPs has been attributed to the presence of high concentrations of chemical constituents termed phytochemical or secondary metabolites they contain (Espitia et al., 2012).

Phytochemicals such as methylxanthines, tannins, phenolic acids, terpenoids, flavonoids, alkaloids, and saponins have been reported as good reducing agents of zinc precursor [30]. This use of plant extract has some benefits such as been safe, cost effective, environment friendly, nonhazardous, bio-compatible, and large-scale production is plausible. The procedure for the biological method of synthesis of ZnO NPs is shown in Fig. 3. (Agarwal et al., 2017).

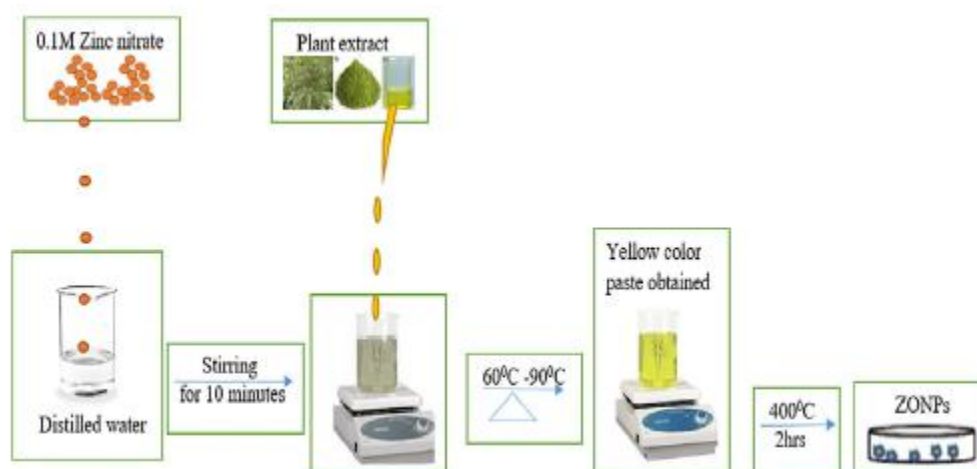


Figure 2.2 Flow chart illustrating the biological method of synthesis of ZnO NPs

ZnO nanoparticles have been successfully synthesized using the green method. Exact mechanisms explaining the biosynthesis procedure of ZnO nanoparticles is still under research, however, the main idea behind the biosynthesis of ZnO nanoparticles may be that the natural materials contain phytochemicals. They reduce the metal (zinc) to the 0-valence state and then through calcinations, oxide may be added to the metal. Another very convincing mechanism is that zinc ions in the solution of the natural extract form complexation with the polyphenols (or other phytochemicals) with zinc in the form of Zn^{+2} . This is then followed by the formation of zinc hydroxide ($Zn(OH)_2$) through hydrolysis and finally after calcinations, the complex decomposes, thereby favoring the formation of ZnO nanoparticles. Several mechanisms for possible formation of ZnO nanoparticles using bio-extracts are depicted in figure 3.4, as collected from different literature. Moreover, the images from reported work have been shown in figure 3.4, depicting the mechanisms involved in the biosynthesis process. (Faisal et al., 2021)

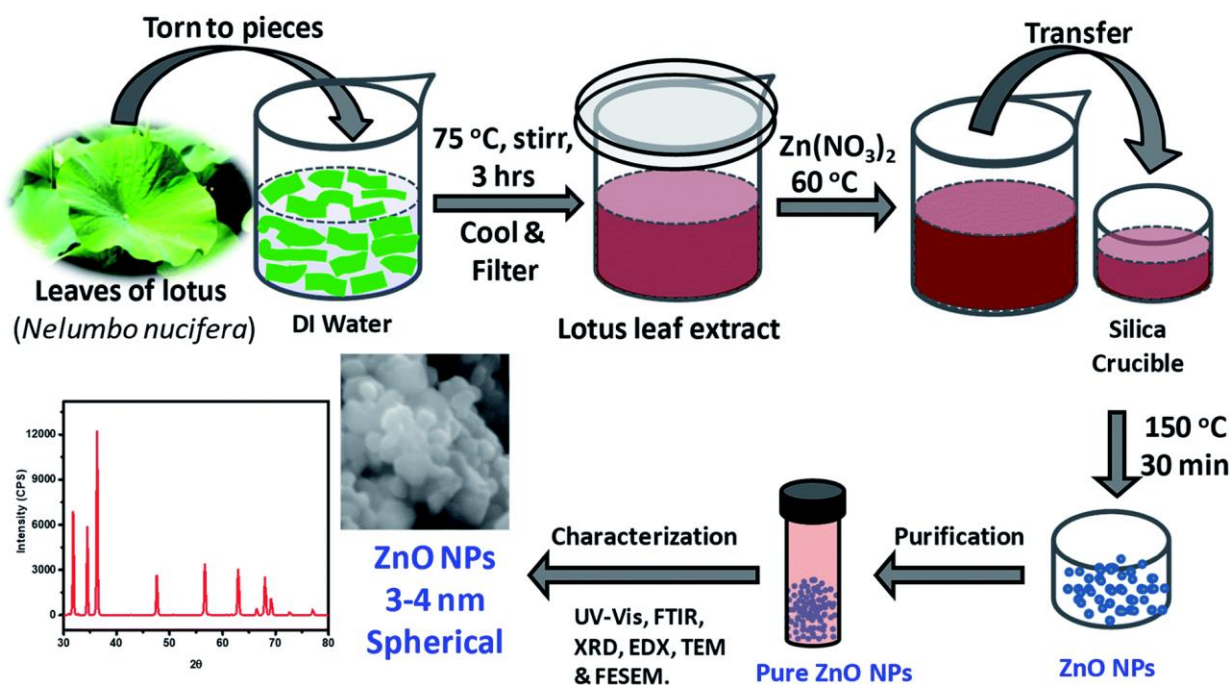


Fig. 2.3 Formation of ZnO nanoparticles using bio-extracts

CHAPTER THREE

RESULTS AND DISCUSSION

3.1 Microscopy Techniques

Microscopy technique is an important tool for characterization and imaging of ZnO NPs. Electron microscopy gives an exact information about the size, structure, shape, spatial resolution, and composition of ZnO NPs. The following techniques, scanning electron microscopy (SEM) fluorescent microscopy, atomic force microscopy (AFM), and transmission electron microscopy (TEM), are used for the morphological identification of ZnO NPs. Among the listed techniques, SEM and TEM are the techniques commonly used by researchers.

3.1.1 SEM

SEM is a technique for high-resolution surface imaging utilized in obtaining information about the structures of samples at nano and micro scale via electron beam. Depending on the electron density of the surface, the larger field depth and high magnification makes SEM images useful for ZnO NPs surface topological assessment. The determination of the morphological identity using this technique is via direct visualization. When ZnO NPs are exposed to electron beams, signals are generated and recorded by the detector. From the recorded signal, information about the morphology, orientation, and crystalline structure of the ZnO NPs are deduced. (Agarwal et al., 2017).

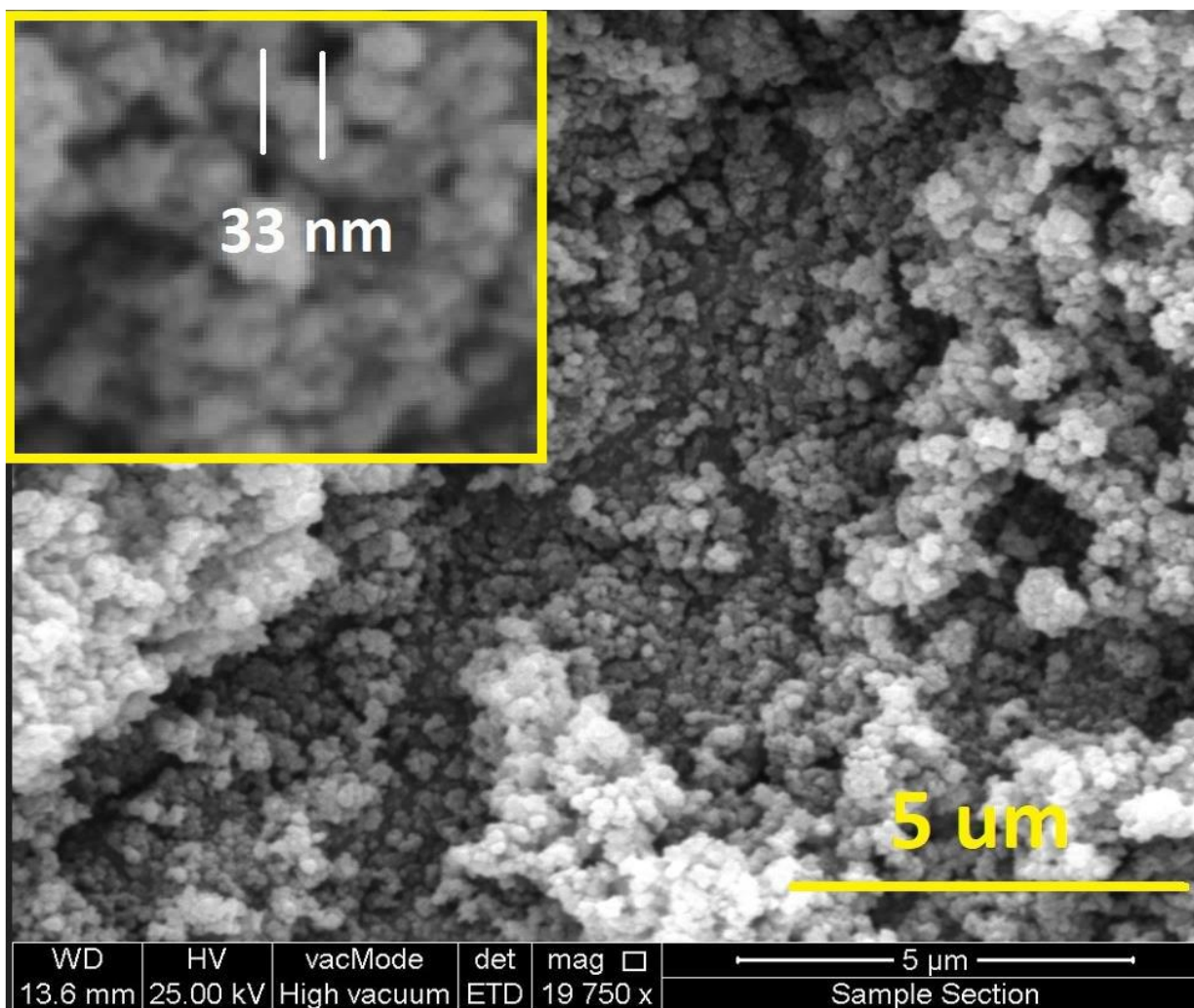


Figure 3.1: SEM analysis of ZnO NPs using *Anethum graveolens* leaf extract

3.1.2 TEM

The application of TEM as a characterization tool is based on the interaction of a thin ZnO NPs sample and current density electron beam (Santos et al., 2011).

When the electron beam and sample are in contact, the electrons are either transmitted or scattered. An image which reveals the morphological properties of ZnO NPs are produced from the transmitted electrons from the TEM machine and the extent of the interaction between the transmitted electron and the sample influences the size and shape of the ZnO NPs.

3.1.3 UV–Visible Spectrophotometry

UV-visible spectrophotometry is a common and cost-effective instrument used for the confirmation of the formation of ZnO NPs. Synthesized ZnO NPs are scanned in the UV region of the electromagnetic wave around 200-700 nm. The interaction between light and the mobile surface electrons of ZnO NPs produced the SPR. Findings from previous studies have established the peaks at wavelength 289-385 nm as the SPR of ZnO NPs (Pai et al., 2019).

3.1.4 Energy Dispersive X-Ray Spectroscopy

Energy dispersive x-ray spectroscopy (EDX) has been reported as a suitable technique for the analysis of the elemental composition of ZnO NPs. The elemental composition analysis is achievable because each element has a unique atomic structure which produces distinct peaks on the X-ray spectrum. EDX technique can also function as a tool for the examination of the extent of ZnO NPs purity. The chemical constituent of the plant extract used as reducing agent has been reported to be the source of other elements such as oxygen and carbon on the EDX. The EDX spectrum obtained from the analysis of the elemental composition of ZnO NPs biosynthesized using jujube extract confirmed the presence of zinc and oxygen with the weight percentages of 70% and 27% respectively; small peaks of some unknown chemical compounds which could be from the extract were also observed in the in the EDX spectrum. In another study on the synthesis of ZnO NPs from *Morus nigra*, the EDX result indicated the existence of zinc and oxygen. (Kumar et al., 2013).

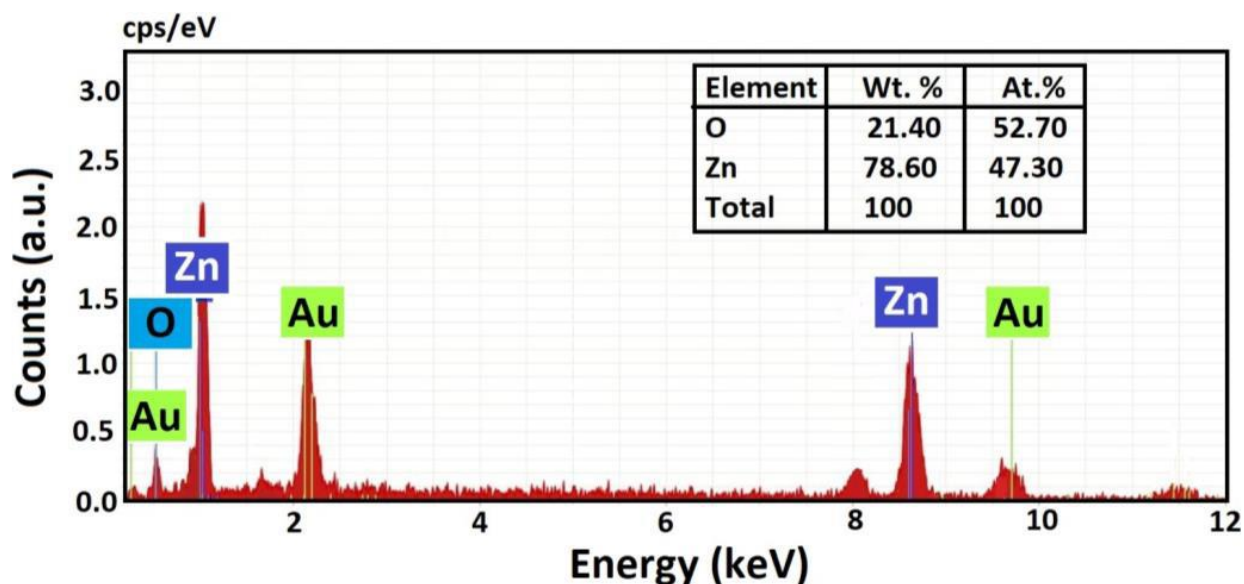


Figure 3.2: EDX Analysis of ZnO NPs using Anethum graveolens leaf extract.

3.1.5 X-Ray Diffraction

X-ray diffraction (XRD) technique is a recommended tool for the crystallinity assessment of ZnO NPs. During XRD analysis, ZnO NPs are exposed to energetic rays from the XRD machine which penetrates through it to provide useful data about its structure [90]. This technique cannot be used for estimating the amorphous structure of nanoparticles due to the absence of diffraction peak. The broadening of the XRD pattern signifies the nano size. Debye-Scherrer's equation is used for the average size estimation of ZnO NPs size.

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

Where K = Scherrer constant (0.9), λ = x-ray wavelength, β = full width at half maximum of the diffraction peak, θ = measured Bragg angle, and d = size of ZnO NPs in nanometers. The sharp peaks found at the lattice planes of (100), (002), (101), (102), (110), (103), (112), and (202) have been reported to indicate the purity and crystallinity of ZnO NPs (Faisal et al., 2021)

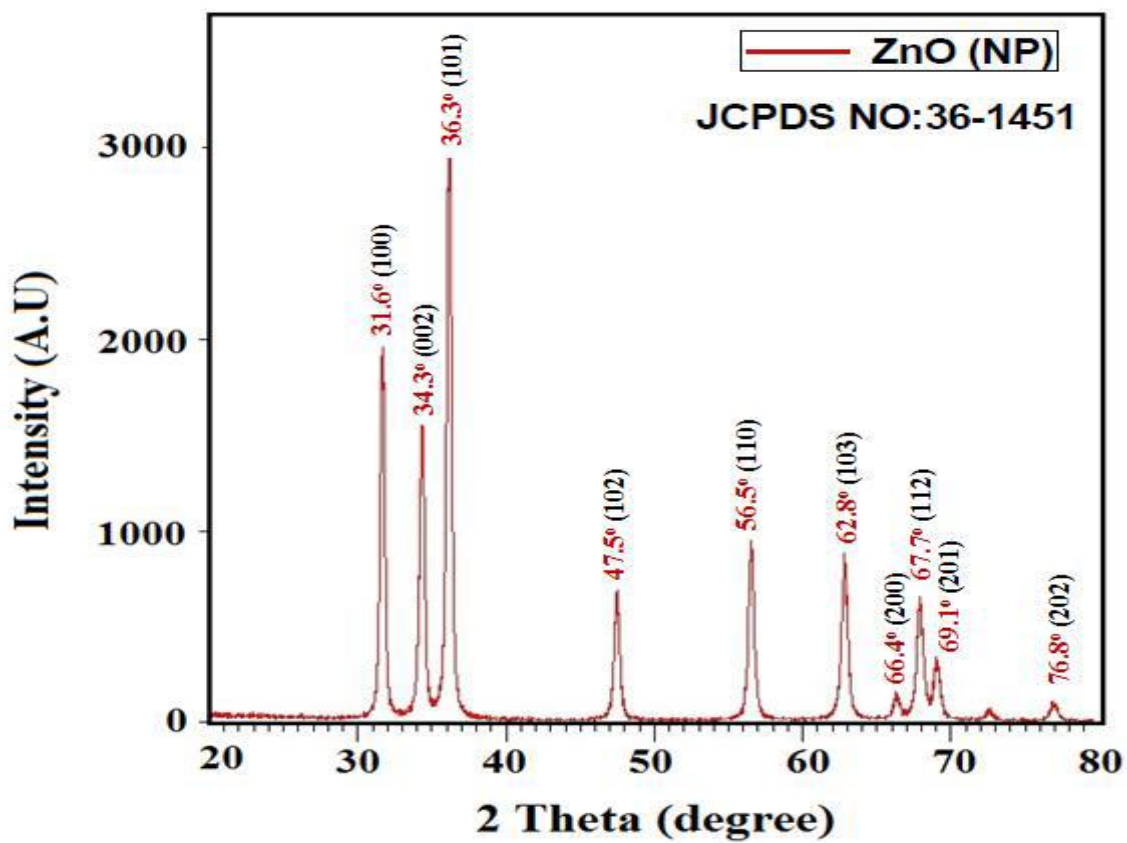


Figure 3.3: Precipitate XRD configuration of ZnO nanoparticles.

3.2 Applications of Zinc Oxide Nanoparticle:

Zinc oxide is widely used in many areas. It plays an important role in a very wide range of applications, ranging from type to ceramics, from pharmaceuticals to agriculture, and from paints to chemicals. Figure 11 shows worldwide consumption of zinc oxide by region. In the Figure 12 summarized application paths of ZnO are presented. (Vinotha et al., 2019)

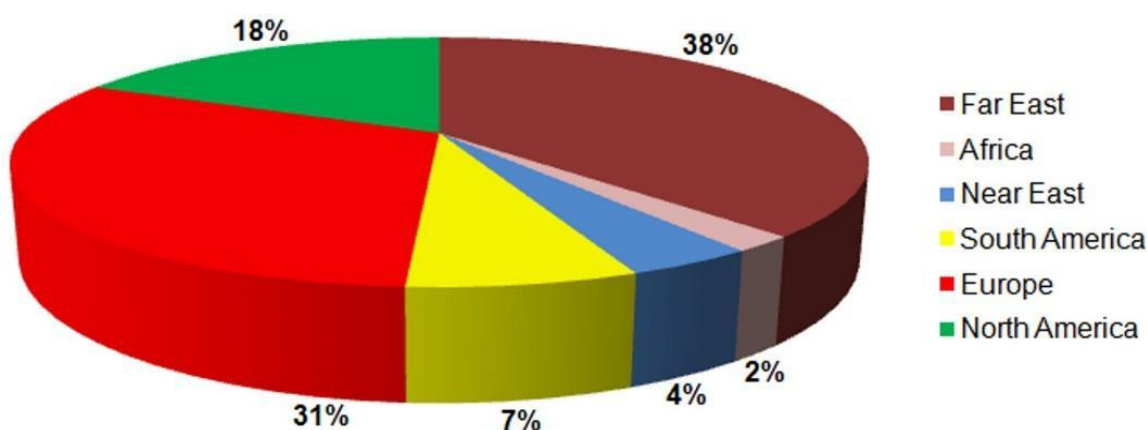


Figure 4.1 summarized application paths of ZnO

Green preparation of zinc oxide nanoparticles involves the use of environmentally friendly methods to synthesize nanoparticles. This method involves the use of natural products such as plant extracts or microorganisms as reducing and stabilizing agents, which reduces the use of toxic chemicals and solvents. Here are some potential applications of green prepared zinc oxide nanoparticles:

1. **Sunscreens:** Zinc oxide nanoparticles are widely used as a key ingredient in sunscreens as they have the ability to absorb and scatter harmful UV rays from the sun. Green preparation of zinc oxide nanoparticles can help produce more environmentally friendly sunscreens. (Pushpalatha et al., 2022)
2. **Environmental remediation:** Green synthesized zinc oxide nanoparticles can be

used in environmental remediation as they have the potential to remove heavy metals and organic pollutants from contaminated soil and water. (Mirzaei and Darroudi, 2017, Thareja and Shukla, 2007)

3. Antimicrobial agents: Zinc oxide nanoparticles possess antimicrobial properties that can be used in the development of antibacterial, antifungal, and antiviral agents. These nanoparticles have been shown to be effective against a wide range of pathogens. (Ali et al., 2018).
4. Catalysts: Green synthesized zinc oxide nanoparticles can be used as catalysts in various industrial processes, such as the production of biodiesel and degradation of pollutants. (Ali et al., 2018)
5. Biosensors: Zinc oxide nanoparticles can be used as sensing elements in biosensors due to their high surface area and sensitivity to changes in the environment. Green preparation of these nanoparticles can help to produce more biocompatible and environmentally friendly biosensors. (Faisal et al., 2021)

Also some different applications that mentioned in fig 4.1

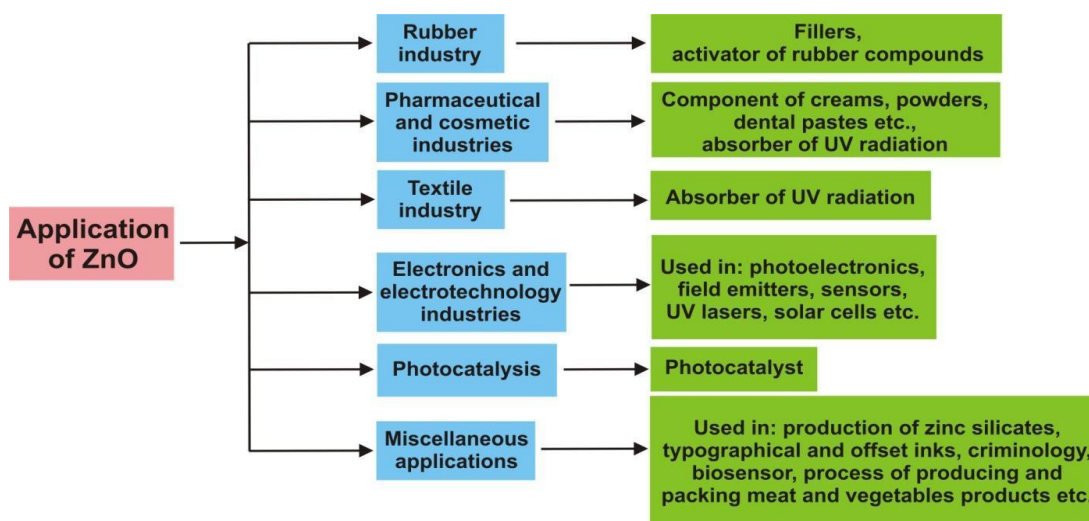


Figure 4.2. Schematic representation all the application of ZnO.

4.4 Summary Table of characterization and application ZnO NPs.

<i>References</i>	<i>Plants name</i>	<i>Plants parts</i>	<i>Type of zinc salt</i>	<i>Grain size</i>	<i>Applications</i>
(Vinotha et al., 2019)	Castusignes	Leaf	Zinc acetate	26-57	Antidiabetic, antibiofilm
(Begum et al., 2018)	Averrhoa bilimbi (cucumbertre)	Fruit	Zinc acetate	12.4	Photoelectrode
(Zare et al., 2017)	Cuminum cyminum	Seed	Zinc nitrate	15-50	Antibacterial
(Jamdagni et al., 2016)	Prosopis farcta (Syriam mesquite)	Aerial	Zinc sulfate	20-40	Antifungal, cytotoxic
(Salari et al., 2017)	Lavandula vera (loddon pink)	Leaf	Zinc sulphate	30	Cytotoxic, antioxidant
(Kairyte et al., 2013)	Mentha pulegium(peppermin)	Leaf	Zinc nitrate	17-59	Antimicrobial
(Jamdagni et al., 2016)	Vaccinium arctostophylos	Leaf	Zinc Chloride	12.4	Photoelectrode
(Akintelu and Folorunso, 2020)	Prunus sp (plum)	Fruit	Zinc acetate	33	photocatalytic
(Rajashekara et al., 2020)	Calotropis gigantean	Leaf	Zinc nitrate	52	cytotoxicity
(Velsankar et al., 2020)	Echinochloa frumentacea	Grain	Zinc acetate	55	Cytotoxicity
(Vijayakumar et al., 2018)	Glycosmis pentaphylla	Leaf	Zinc acetate	19	antimicrobial
(Vinotha et al., 2019)	Costus igneus	Leaf	Zinc acetate	39	Antidiabetic, antibiofilm, antimicrobial
(Wang et al., 2019)	Marsdenia tenacissima	Leaf	Zinc acetate	18	anticancer
(Begum et al., 2018)	Averrhoa carambola L	Leaf	Zinc nitrate	60	Agricultural
(Sirelkhatim et al., 2015)	Lagerstroemia speciosa	Leaf	Zinc acetate	15	Hemolytic
(Ali et al., 2018).	Lavandula vera	Leaf	Zinc sulphate	2.8	Dentistry

CHAPTER FOUR

CONCLUSION

4.1 Conclusion

The use of plant extracts in the biosynthesis of nanoparticles offers several advantages over conventional methods, such as being cost-effective, non-toxic, and environmentally friendly. Additionally, plant extracts contain a variety of secondary metabolites such as polyphenols, flavonoids, and terpenoids, which act as reducing and stabilizing agents for nanoparticle synthesis. Further research in this field can help to optimize the production process and increase the yield of nanoparticles using plant extracts. In particular, understanding the mechanisms involved in nanoparticle synthesis using plant extracts can lead to the development of more efficient and sustainable production methods. Additionally, the identification and characterization of phytochemicals involved in nanoparticle synthesis can lead to the development of novel plant-based drugs and therapeutics. The potential applications of plant-mediated nanoparticles are vast, ranging from drug delivery to water treatment. For instance, nanoparticles synthesized using plant extracts have shown promise as anti-cancer agents, antimicrobial agents, and catalysts for environmental remediation. With continued research, plant-mediated nanoparticle synthesis could become a major area of research in the coming years with significant implications for various industries.

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