Research Article

Zinc oxide nanoparticles green synthesis using *Moringa oleifera* and tulasi a formulation and its anti-inflammatory activity

T. Vinaya Swetha, S. Rajeshkumar*, T. Lakshmi, Anitha Roy

ABSTRACT

Aim: To study the synthesis of zinc oxide nanoparticles using plant formulation and its anti-inflammatory activity. **Introduction:** Zinc oxide nanoparticles (ZnO NPs) are metal oxide nanoparticle paves the way for new field of biomedical application from diagnosis to treatment. ZnO semiconductor is a material that can be easily synthesized with different structures due to its chemical behaviors. ZnO NPs can be synthesized from various plants. **Materials and Methods:** The ZnO NPs were synthesised using herbal formulations and anti-inflammatory activity using protein denaturation method. **Results:** ZnO NPs synthesized from *Moringa oleifera* and tulasi extract have high anti-inflammatory activity that can be used for many medicinal purpose and many health-related diseases such as arthritis, cancer, and thyroid function.

KEY WORDS: Antimicrobial, Diseases, Inflammation, Nanoparticles, Plant extract

INTRODUCTION

Zinc oxide nanoparticles (ZnO NPs) are metal oxide nanoparticle paves the way for new field of biomedical application from diagnosis to treatment. It is a wide gap semiconductor with 3.37 ev at room temperature. ZnO semiconductor is a material that can be easily synthesized with different structures due to its chemical behaviors.[1] ZnO NPs can be synthesized from various plants such as *Azadirachta indica*, *Gamella sinensis*, *Rosa canina*, and *Moringa oleifera*. There are many methods are used to synthesize ZnO NPs such as chemical reduction,[2] thermal decomposition,[3] laser irradiation[4] etc. The ZnO NPs can also be synthesized from microorganisms, biorenewable source, and are of immense interest in the current area of biomedical research.[5] NPs can also be synthesized using angiospermic plant extracts, especially with medicinal plants, it is called as "nature nano factory."

Nanoparticle synthesis can also be mediated by plant extracts, water-soluble plant biomolecules that are present in the plant extracts such as alkaloids, polyphenol, flavonoid, and terpenoid reduce the metal ion to nanoparticle in a small time.[6,7] Biomedical applications of ZnO NPs are that it has antibacterial activity, photothermal therapy, photoimaging, cosmetics and daily care products, food supplements and packaging, nanosensors and drug delivery, environmental remediation, medical devices, and drugs. Apart from that, ZnO NPs are used in ointments as antimicrobial and antifungal agents.

ZnO NPs have some adverse effects also; human exposure to ZnO NPs is increased due to their vast application in cosmetics and daily care products. The food drug administration does not require manufacturers to determine the nanoscale ingredients for safety use. Pre-research data state that NPs have biological, chemical, and physical properties.[8] It states that ZnO NPs are highly reactive and induce highly reactive oxygen species (ROS)-mediated single-stranded DNA that breaks at a relatively low concentration ZnO NPs in already synthesized products. The greater effect of ZnO NPs is highly used in many industries. Another effect is fetus and children, whose cells divide in faster rate mediate ROS-mediated DNA damage. If it is not treated, it may lead to health hazards.[8]

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ZnO NPs have a vast use as an antibacterial activity in that ZnO NPs have a positive charge at their surface and the bacterial membrane has a negatively charged surface, in which ZnO NPs enter the bacterial surface by endocytosis, many mechanisms have said that its main antibacterial effects as they cause destruction of bacterial cell wall and cause release of Zn^{2+} ions, and they also cause damage cellular metabolism.[9] Antibacterial agents which containing ZnO NPs are used in dental composites.[10] ZnO NPs are termed as an effective cancer clearing agent; it exhibits preferred toxicity among cells when compared with normal cells.[11] In electrical industry, ZnO nanocomposites had showed high stability and high retention power.[12] They also used for the detection for neurotransmitter inside the brain.[13] ZnO NPs contain fluorescent properties used for imaging cancer cells and bacteria.[14] ZnO NPs also synthesized using microorganisms such as viruses, bacteria, algae, fungus, and other biorenewable sources such as plant extracts; they are mainly used in biomedical research purpose.[15-17] In the field of dentistry, eradication of caries-causing bacteria and reparation of tooth blemishes where decay has adhered to, are made possible with the help of nanorobots in accordance with a computer to control them.[18] Antibiotic resistance can be reduced with the help of ZnO NPs; it enhances the antibacterial activity of ciprofloxacin against microorganisms.[19]

MATERIALS AND METHODS

Collection and Preparation of Plant Extract

Moringa oleifera and tulasi leaves were collected from Chennai. The collected leaves were washed 3–4 times using distilled water, then dried it in shade for 7–14 days. The well-dried leaves were made into a powder using mortar and pestle. The collected powder was stored in an airtight container. One gram of leaves powder was dissolved in distilled water and boiled for 5–10 min at 60–70°. The solution was filtered using Whatman No.1 filter paper. The filtered extract was collected and stored in 4° for further use.

Synthesis of ZnO NPs

One millimolar of zinc sulfate dissolved in 90 ml of double-distilled water. The plant extracts of *M. oleifera* and tulasi were added with the metal solution and were made into 100 ml solution. The color change was observed visually and photographed. The solution is kept in magnetic stirrer for NPs synthesis.

Characterization of ZnO NPs

The synthesized NPs solution is preliminarily characterized using ultraviolet (UV)-visible spectroscopy. Three milliliter of the solution is taken in the cuvette and scanned in double-beam UV-visible spectrophotometer from 300 nm to 700 nm wavelength. The results were recorded for the graphical analysis.

Preparation of NPs Powder

The NPs solution is centrifuged using Lark refrigerated centrifuge. The ZnO NPs solution is centrifuged at 8000 rpm for 10 min and the pellet is collected and washed with distilled water twice. The final purified pellet is collected and dried at 100–150°C for 2/24 h, and finally, the NPs powder is collected and stored in airtight Eppendorf tube.

Inhibition of Albumin Denaturation Assay

Bovine serum albumin (BSA) was used as a reagent for the assay. BSA makes up approximately 60% of all proteins in animal serum. It is commonly used in culture, particularly when protein supplementation is necessary and the other components of serum are unwanted. BSA undergoes denaturation on heating and starts expressing antigens associated with Type III hypersensitive reaction which is related to disease such as rheumatoid arthritis, glomerulonephritis, serum sickness, and systemic lupus erythematosus.

Two milliliter of 1% bovine albumin fraction was mixed with 400 µl of plant crude extract in different concentrations (500–100 µg/mL) and the pH of reaction mixture was adjusted to 6.8 using 1 N HCl. The reaction mixture was incubated at room temperature for 20 min and then heated at 55°C for 20 min in a water bath. The mixture was cooled to room temperature and the absorbance value was recorded at 660 nm. An equal amount of plant extract was replaced with dimethyl sulfoxide for control. Diclofenac sodium in different concentrations was used as standards. The experiment was performed in Chennai.

% Inhibition was calculated using the formulae:

% Inhibition = Control O.D–sample O.D/Control O.D

RESULTS AND DISCUSSION

Medical plants are the major source of drug to cure human disease. Since NPs have wide uses of medicinal values, it is used in many drug industries along with plant extract. India is rich in herbal plants and has more medicinal values that are used in Unani system, Ayurveda, and other traditional medications. In medicine, ZnO and ZnO NPs have many uses such as wound healing, thyroid function, immune system maintenance, and much more.

The ZnO NPs are synthesized from *M. oleifera* and tulasi plant extract. The ZnO NPs exhibit pale yellow color [Figure 1]. Initially, the zinc sulfate was added to aqueous solution with plant leaves extract; they exhibit a color change from dark brown to pale yellow color. This color change is due to the reduction of zinc ions and this is observed in UV-visible spectroscopy [Figure 2].

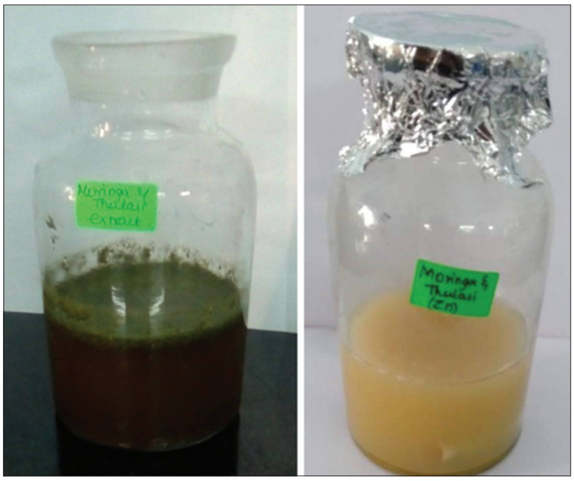

Figure 1: Visual observation

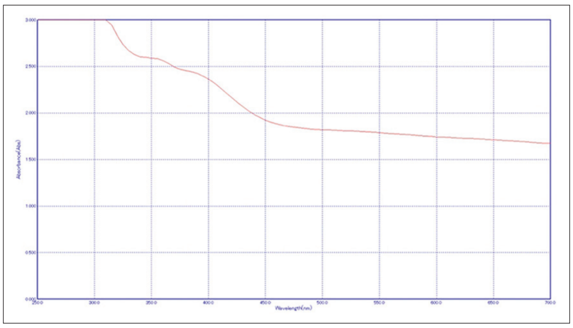

Figure 2: Ultraviolet-visible spectroscopy

The UV-visible analysis of ZnO NP was analyzed in the absorbance range between 250 nm and 700 nm. The peak was found to be maximum at 320 nm. Thus, ZnO NPs were synthesized at 320 nm due to the reduction of ions in aqueous solution.

Anti-inflammatory Activity

The anti-inflammatory activity of ZnO NPs using *M. oleifera* and tulasi plant extract was measured by the inhibition of albumin denaturation assay [Figures 3 and 4]. Inflammation is a complex event linked to tissue damage by various causes such as microorganisms, trauma, chemical factors, and other various phenomenons.[20] The pathological response of inflammation includes redness, swelling, heat, and pain. It is a main pathogenic cause for many diseases such as cancer, stroke, and arthritis. The ability of the NPs to inhibit the denaturation of proteins indicates the potential for anti-inflammatory activity. There is no zone of inhibition seen in ZnO NPs synthesis since the zone of inhibition is not seen with this. The anti-inflammatory activity of the extract is same as its stranded substance.

In the study of Mina Zara, the synthesis of ZnO NPs was at the peak of 370 nm; in the present study, the synthesis of NPs was occurred at the peak of 320 nm. In the study of Chaudhuri and Malodia, ZnO NPs synthesis was at a peak of 350 nm.[21]

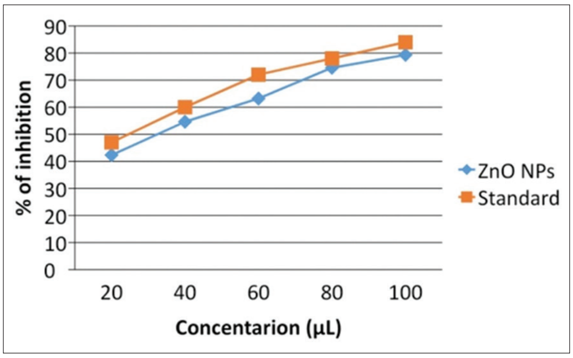

Figure 3: Anti-inflammatory activity of ZnO NPs

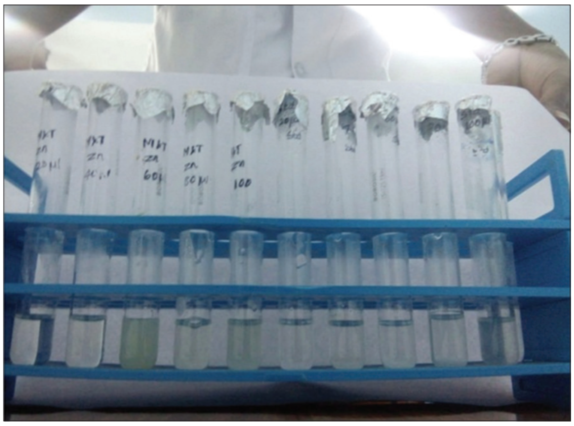

Figure 4: Anti-inflammatory activity

The most commonly used drugs for inflammation such as ibuprofen and diclofenac sodium are belonged to nonsteroidal anti-inflammatory drug group of drugs. They have high anti-inflammatory activity and routine usage of these ZnO NPs with anti-inflammatory activity has good result in many diseases such as Thurmond dysfunction and cancer issues. According to research, ZnO NPs produced from *M. oleifera* and tulasi extract have considered as potential candidate for anti-inflammatory activity. Thus, it reduces the risk of cancer, thyroid dysfunction, and other health-related problems.

CONCLUSION

The ZnO NPs are synthesized from *M. oleifera* and tulasi plant extract. All these data shows that ZnO NPs produced from plant extract have high anti-inflammatory activity. Further research has to be done to develop the new strategies in the use of plants for medicinal purpose.

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