

RESEARCH PAPER

## Green Synthesis of Zinc Oxide Nanoparticles Using *Achillea Wilhelmsii* Extract: A Biological Approach

Muslimanova Gulnar <sup>1\*</sup>, Mohaned Mohammed Hani <sup>2</sup>, Mohammed AbedJawad <sup>3</sup>, Ghufran Lutfi Ismaeel <sup>4</sup>, Safaa Sabah Mohammed <sup>5</sup>, Fakhri Alajeeli <sup>6</sup>, Hameed Ghazy <sup>7</sup>, Zainab Al-Hawraa Riyad Muedii <sup>8</sup>, Xodjiyeva Dilbar Tadjiyevna <sup>9</sup>, Kadirova Dilbar Normuminovna <sup>10</sup>, Akhmedova Dilafroz Bahodirovna <sup>11</sup>, Kulmukhanova Damira Rustemkyzy <sup>12</sup>

<sup>1</sup> D. Serikbayev East Kazakhstan Technical University The Republic of Kazakhstan, Ust-Kamenogorsk

<sup>2</sup> Department of Medical Instrumentation Engineering Techniques, Imam Ja'afar Al-Sadiq University, Iraq

<sup>3</sup> Department of anesthesia, Al-Nisour University College, Baghdad, Iraq

<sup>4</sup> Department of Pharmacology, College of Pharmacy, University of Al-Ameed, Karbala, Iraq

<sup>5</sup> Department of Optical Techniques, AlNoor University College, Iraq

<sup>6</sup> Department of Optical Techniques, Al-Hadi University College, Baghdad, 10011, Iraq

<sup>7</sup> Department of Pharmacy, Al-Manara College For Medical Sciences/ (Maysan), Iraq

<sup>8</sup> College of Medical Technology, Medical Lab techniques, National University of Science and Technology, Dhi Qar, Iraq

<sup>9</sup> Bukhara State Medical Institute, Uzbekistan

<sup>10</sup> Termiz State University, Uzbekistan

<sup>11</sup> Bukhara State Medical Institute, Uzbekistan

<sup>12</sup> Abai Kazakh National Pedagogical University, Dostyk 13. Almaty, Kazakhstan

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### ABSTRACT

Nanotechnology is a branch of science and engineering that involves the manipulation, production, and design of materials and devices at a nanoscale level. The prefix "nano" represents one billionth of a meter, and at this size, the physical and chemical characteristics of materials can be distinct from those of larger objects. This can result in new properties that can be exploited for a variety of uses. In recent times, biological methods for the synthesis of nanoparticles have become popular because they offer several advantages over traditional chemical and physical processes. One significant benefit is that biological approaches use less energy and time. Additionally, they are environmentally friendly because they do not use toxic solvents or dangerous materials. In this experiment, zinc oxide nanoparticles (ZnO NPs) were synthesized using *Achillea wilhelmsii* extract as the reducing agent. 40 mL of *Achillea Wilhelmsii* extract was gradually added to 60 mL of zinc nitrate solution with a concentration of 1 M. Spectrophotometric analysis of the solution revealed an absorption maximum at 370 nm, signaling the existence of ZnO NPs. Scanning Electron Microscopy, or SEM analysis showed the nanoparticles to have a spherical morphology with diameter ranging from 38-60 nm. Particle Size Analysis (PSA) showed that the nanoparticles were between 35 and 218 nm in size. This study demonstrated that it is possible to create ZnO NPs using a safe biological process that does not require the use of hazardous chemicals.

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\* Corresponding Author Email: [gMuslimanova@edu.ektu.kz](mailto:gMuslimanova@edu.ektu.kz)



## INTRODUCTION

Metal oxide nanoparticles are a significant type of nanomaterials widely utilized in various industries [1-3]. Among these, zinc oxide nanoparticles (ZnO NPs) hold great importance in medicine, pharmacy, and other sectors [4]. ZnO NPs are extensively utilized in the manufacturing of nanoscale materials, following silver oxide, titanium dioxide, and silicon dioxide nanoparticles [5]. The collective production of 41 different nano-products using ZnO NPs is estimated to exceed 600 tons annually [6,7]. These nanoparticles are typically synthesized through chemical methods. However, their excessive application has resulted in the generation of nano-sewage, with a substantial portion entering aquatic environments, estimated to be over 410,000 tons per year [8].

The presence of nanoparticles in water has led to genetic alterations and elevated mortality rates among aquatic organisms, including fish [9]. Additionally, the chemical processes involved in nanoparticle production have proven to be environmentally costly. Consequently, researchers have turned their attention towards biological synthesis methods, also known as green synthesis, for producing nanoparticles [10]. These methods are being explored primarily due to their potential environmental benefits along with other compelling reasons. The emerging methods, known as green synthesis, involve harnessing the potential of plants, microorganisms, or fungi as a foundation for nanoparticle production. These approaches have been recognized as highly effective in utilizing the inherent biological

components present in nature [11-13]. Plants, in particular, offer significant advantages for widespread adoption, as they are abundant, easily accessible. In the past few years, there have been notable achievements in utilizing various plant species to quickly and effectively produce gold, silver, copper, and other types of nanoparticles outside of living cells [14]. The extracts derived from *aloe vera* [15], *Glycosmis pentaphylla* leaf [16], and *Limonia acidissima* [17] have demonstrated successful outcomes in synthesizing ZnO NPs. The techniques employed for synthesizing nanoparticles from various plant components are comparatively simpler from a technical standpoint than chemical methods. As a result, biological materials assume a crucial role in nanoparticle synthesis, representing innovative alternatives to traditional chemical approaches. While the importance of utilizing plants for nanoparticle synthesis has been emphasized, regrettably, only a restricted number of studies have focused on this subject in Iraq [18,19]. Due to the significant variety of plant species found in Iraq, further research is necessary to explore this aspect.

Mohammadi *et al.* [20] used the aqueous extract of *Euphorbia petiolata* to synthesize ZnO NPs with a particle size range between 30-40 nm using the green synthesis method. Yassin *et al.* [21] also reported the preparation of ZnO NPs by using extracts of *Origanum majorana*. Another study was conducted where they utilized the *Achillea wilhelmsii* flower to extract cadmium oxide nanoparticles. The researchers found that certain



Fig. 1. Image of *Achillea Wilhelmsii* plant.

compounds present in the extract, including tannins, flavonoids, alkaloids, and carotenoids, played a crucial role in reducing cadmium oxide ions and ensuring the stability of the resulting nanoparticles [22]. Furthermore, the blossoms of this particular species have been harnessed effectively in generating gold nanoparticles [23].

*Achillea*, a member of the *Asteraceae* plant family, possesses numerous medicinal and therapeutic characteristics. It is rich in polyphenolic compounds, phenolic compounds, sesquiterpene lactones, alkaloids, and flavonoids. *Achillea* is employed for various health purposes, including managing hypoglycemia, acting as a nerve tonic, alleviating hemorrhoids, treating diarrhea, providing antacid effects, acting as a carminative and appetizer, possessing anthelmintic properties, and serving as an antibacterial remedy [24-26]. *Achillea Wilhelmsii* is extensively utilized in various food items within Iraq [Fig. 1].

It has a rich history in traditional medicine, known for its diverse medicinal properties that include antioxidant, antifungal, antibacterial effects, and more [27,28]. Additionally, *Achillea Wilhelmsii* has been found to possess LDL-cholesterol-lowering properties and is abundant in oily and proteinaceous substances [29]. Given that the extract from this particular genus contains a high concentration of substances that possess antioxidant and reducing agents, it can be deemed significant and noteworthy in terms of its potential as a secure biological source for reducing zinc ions into nanoparticles. Therefore, this study was conducted with the aim of synthesizing green nanoparticles of ZnO using *Achillea* plants.

## MATERIAL AND METHODS

In January 2023, *Achillea Wilhelmsii* flower shoots were collected from the Baghdad Forest & Agricultural Research Center and identified at the College of Agriculture, Baghdad University. The flower branches were dried in the shade before

being washed with deionized water. To prepare the solution, 25 grams of dried flowers were placed in an Erlenmeyer flask and brought up to a volume of 100 milliliters. The mixture was boiled for 10 minutes and allowed to cool. The extract was then filtered using clean paper and 0.45 µm filters. Under reflux conditions at a temperature of 75°C, a volume of 40 mL of *Achillea Wilhelmsii* extract was slowly introduced to 60 mL of 1 M zinc nitrate solution. This addition was done drop by drop. The process lasted for 90 minutes and aimed to observe an alteration in hue caused by the phenomenon of the surface plasmon resonance effect. The changes were monitored using the ultraviolet-visible spectroscopy (UV-vis) technique. Simultaneously, white precipitates started forming. To separate the precipitation completely, centrifugation was performed at 8000 rpm. The resulting powder was then washed with methanol and distilled water to eliminate any potential contaminants. Subsequently, the powder underwent annealing in a muffle furnace at 450°C for a duration of 90 minutes [Fig. 2].

The nanoparticles obtained from this process were comprehensively studied via various analytical techniques such as UV-vis spectroscopy, X-ray diffraction (XRD), energy-dispersive X-ray spectroscopy (EDX), scanning electron microscopy (SEM), and Fourier-transform infrared spectroscopy (FTIR).

## RESULTS AND DISCUSSION

### UV-vis Spectrometry

The UV-vis of the sample revealed a distinct peak in light absorption at 370 nm (T80 series of UV-Visible Spectrophotometers, England) [Fig. 3]. This matches earlier findings that ZnO NPs absorb light maximally between 360-380 nm [30-32].

### XRD analysis

XRD provides a distinct diffraction pattern for each unique crystalline solid, akin to a

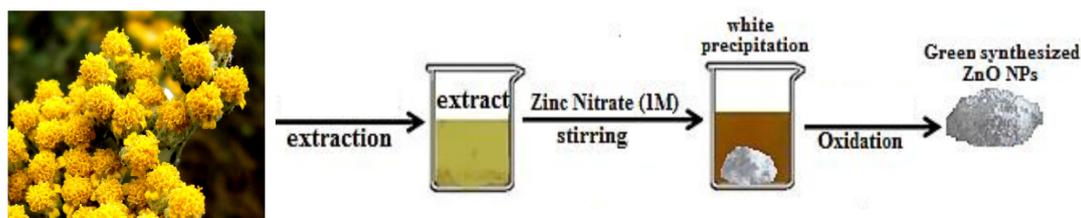


Fig. 2. The process of producing ZnO NPs using the extract derived from *Achillea Wilhelmsii*.

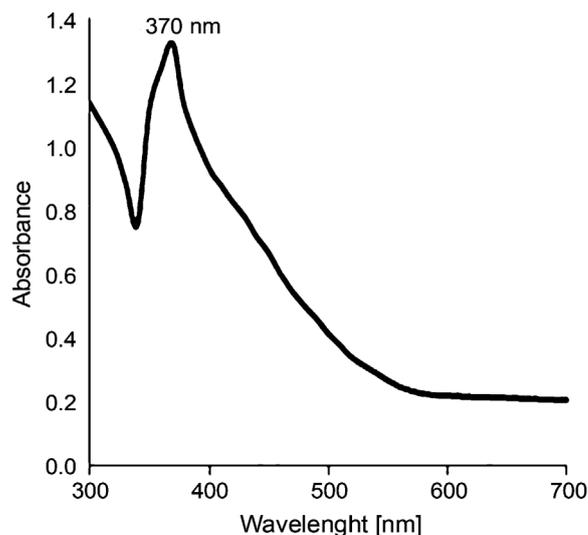


Fig. 3. UV-vis spectrum ZnO NPs synthesized by the extract derived from *Achillea Wilhelmsii*.

“fingerprint” for identifying materials. The X-ray diffraction pattern of the green synthesized ZnO NPs showed several distinct peaks between 0 and 90° (2θ), indicating the crystalline structure of the nanostructures. The detected signals at specific 2θ values of 31.8°, 34.5°, 36.3°, 47.6°, 56.6°, 62.9°, 66.4°, 68.0°, 69.1°, and 77.0° correspond to different crystallographic planes of ZnO (STOE Company, Germany) [Fig. 4]. The peak positions match well with standard diffraction data for crystalline monoclinic ZnO. The lack of extra peaks suggests high phase purity of the green synthesized ZnO NPs. The XRD results confirm the successful production of crystalline ZnO NPs

using the green synthesis method, consistent with previous reports [19, 20, 31].

The average crystallite size of the synthesized ZnO NPs, as estimated from X-ray diffraction data, was determined to be 16 nm. This size aligns with the nanoparticle dimensions reported in prior studies [6].

#### Energy-Dispersive X-ray Analysis (EDX)

EDX analysis was used to semi-quantitatively determine the elemental composition of ZnO NPs produced from *Achillea Wilhelmsii*. The EDX results confirmed the presence of zinc and oxygen as the primary elements, with approximately

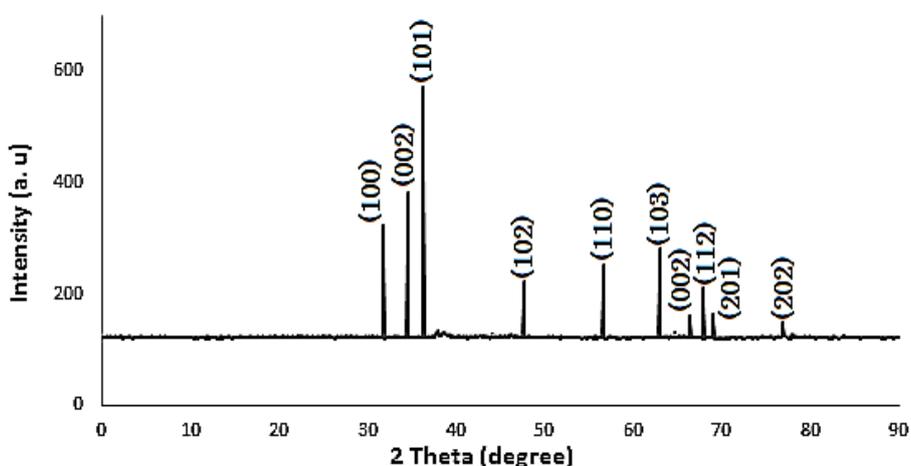


Fig. 4. XRD profile of ZnO NPs produced via green synthesis.

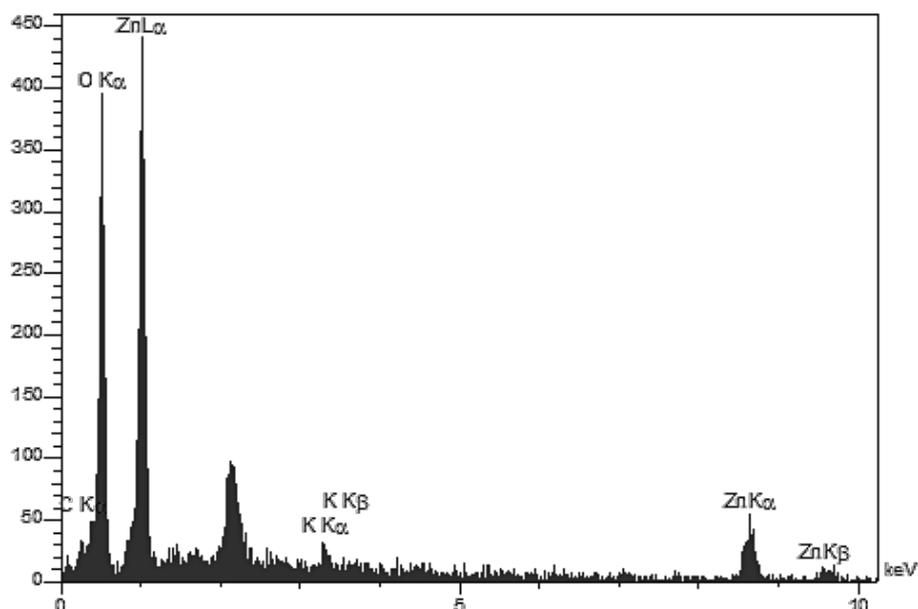


Fig. 5. The EDX Spectrum of ZnO NPs by *Achillea Wilhelmsii*.

51.3% oxygen and 36.8% zinc by weight. Carbon was also detected at 13.2% by weight. The EDX spectrum showed dominant zinc peaks around 1 keV, consistent with ZnO NPs as the primary component. Additional smaller peaks at higher energies around 3 keV and 8.5 keV corresponded to the minor presence of other elements [Fig. 5].

Carbon presence in the spectrum signifies plant phytochemical group activity in limiting and reducing ZnO NPs synthesis [33]. At an energy of 1 keV in the ultra-peak spectrum, ZnO NPs are detected, aligning with other studies [19].

#### SEM analysis

The synthesized ZnO NPs were analyzed using SEM (JSM-6360) to determine their morphology and size. SEM images revealed spherical-shaped

ZnO NPs with diameters ranging from 38-60 nm [Fig. 4]. The particles appeared clustered together with rough surfaces. These results closely matched findings by Jose *et al.* [15], who synthesized similar ZnO NPs using a *Aloe Vera* plant extract and observed spherical ZnO NPs of 38-60 nm diameter. Spherical ZnO NPs of 35-60 nm diameter were also reported using orange fruit peel extracts [34]. Overall, the spherical morphology and 38-60 nm diameter of the ZnO NPs synthesized here using *Achillea Wilhelmsii* align with previous reports that employed plant extracts like *Aloe vera*, *Azadirachta indica*, *Abutilon indicum*, and *Clerodendrum inerme* [35]. The SEM analysis confirms the synthesis of spherical-shaped ZnO NPs with diameters within the typical range of 38-60 nm using the *Achillea Wilhelmsii* plant extract.

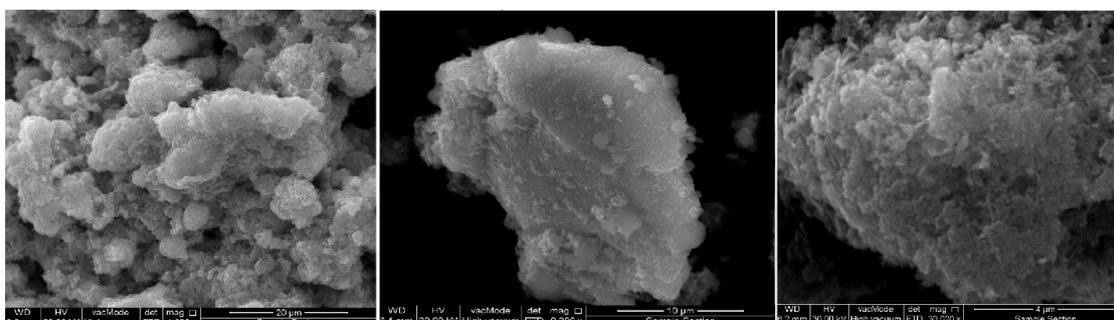


Fig. 6. SEM micrographs of green synthesized ZnO NPs.

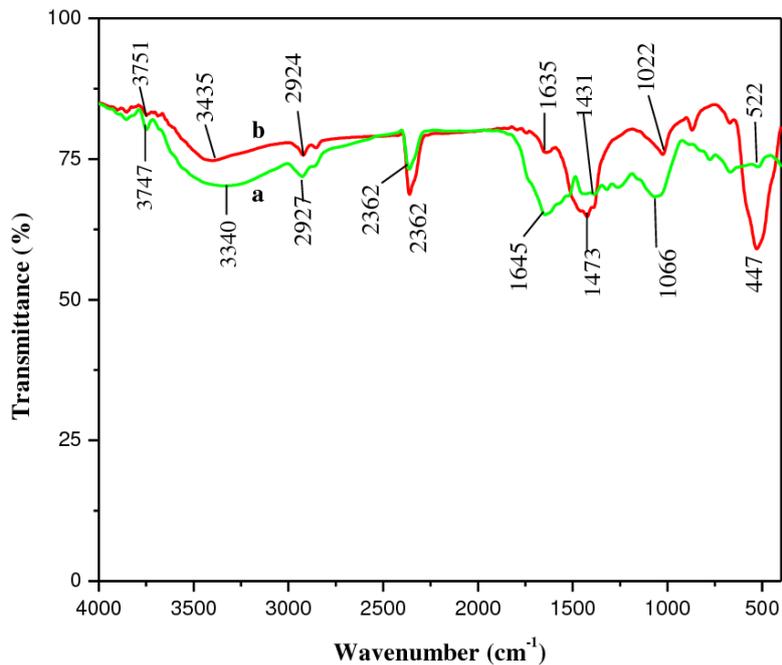


Fig. 7. The FTIR spectrum of (a) *Achillea Wilhelmsii* extract and (b) green synthesized ZnO NPs.

#### FTIR analysis

The measurements of FTIR spectra analysis were performed at room temperature using the KBr pellet technique. Fig. 7(a) displays the FTIR spectrum for ZnO NPs synthesized via the bio-synthesis approach, while Fig. 7(b) represents the spectrum obtained from *Achillea Wilhelmsii* extract. In the *Achillea Wilhelmsii* extract spectrum, several absorption bands were observed at specific wavenumbers: 3747, 3340, 2927, 2362,

1645, 1431, 1066, and 522  $\text{cm}^{-1}$ . After synthesizing the ZnO NPs, these bands underwent a shift and appeared at slightly different wavenumbers: 3751, 3435, 2924, 2362, 1635, 1473, 1022, and 447  $\text{cm}^{-1}$ , respectively. The peak noted at 447  $\text{cm}^{-1}$  in the lower energy zone is of particular note, as it indicates the bending vibrational mode of the ZnO bond in the produced nanoparticles.

The metal-oxygen region is between 400 and 600  $\text{cm}^{-1}$ . In the FTIR spectra, there is a broad

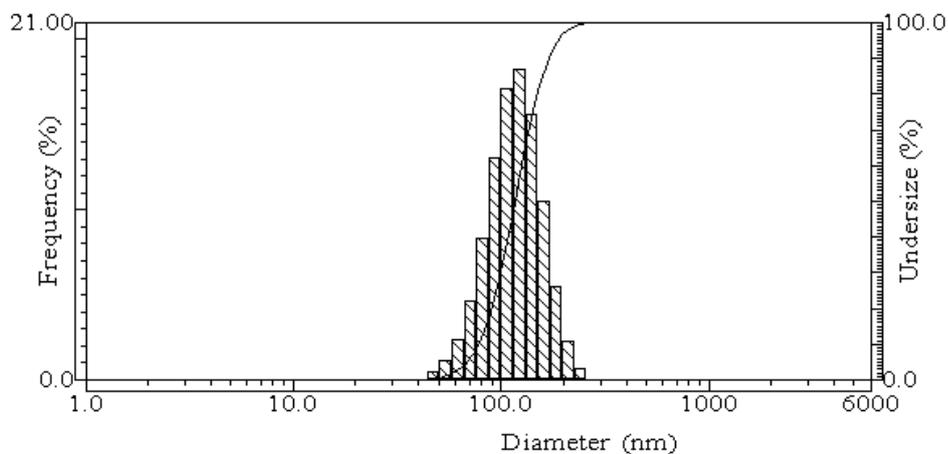


Fig. 8. Size distribution diagram of biologically produced ZnO NPs using PSA device.

peak in the higher energy region which can be attributed to the stretching of O-H bonds and the presence of alcohols and phenols that are hydrogen-bonded (at approximately 3340 and 3435  $\text{cm}^{-1}$ ). The vibration band occurring around 2927 and 2924  $\text{cm}^{-1}$  signifies the stretching of C-H bonds, implying the existence of an alkane group [36]. The enol form of 1,3-diketones and hydroxy aryl ketones is evidenced by medium and sharp peaks observed at around 2362  $\text{cm}^{-1}$ . Mononuclear aromatics, cis-tri substituted, and vinyl exhibit absorption peaks at about 1645 and 1635  $\text{cm}^{-1}$  [37]. Vibrations corresponding to O-H bond stretching in the polyols group are observed at wavenumbers of (1431, 1473)  $\text{cm}^{-1}$ . Lastly, Vibrations corresponding to stretching modes occur at around 1066 and 1022  $\text{cm}^{-1}$ , representing the stretching of C-N bonds in aliphatic amines.

#### Particle Size Analyzer

The dynamic light scattering analyzer is commonly used to examine and study particles, emulsions, or molecules that have been dispersed or dissolved in a liquid. It finds application in determining the sizes of metallic nanoparticles or quantum dots, as well as analyzing the estimated population of both large and small aggregates for small molecules. After obtaining a solution through interaction with distilled water, a volume of 1 mL was diluted to 10 mL and subjected to analysis using the PSA analyzer Model: 90Plus/BI-MAS. The findings revealed that the nanoparticles generated have a size range of approximately 35-218 nanometers, with the majority of particles measuring around 117 nm [Fig. 8].

#### CONCLUSION

This investigation synthesized ZnO NPs with *Achillea Wilhelmsii* extract, which possesses antioxidant and regenerative chemical entities. Nanoparticle specifications were determined via existing instrumentation. In conclusion, an effective, green, biological, inexpensive and sustainable protocol for synthesizing ZnO NPs has been established, utilizing *Achillea Wilhelmsii* extract as a reducing agent. Uv-vis, FTIR, XRD, SEM and EDX analysis definitively confirmed nanoparticle biosynthesis and characteristics. Benefits afforded by this method consist of rapid reaction times, simple one-pot preparation, direct and sustainable nanoparticle synthesis, avoidance of toxic chemicals, extra surfactants/reductants,

organic solvents, and a simple work-up procedure.

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#### CONFLICT OF INTEREST

The authors declare that there is no conflict of interests regarding the publication of this manuscript.

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