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Green Synthesis of ZnO NPs Using Chamomile Flower Extract Via Pulsed Laser Ablation and Its Effect on Surface Wounds

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Abstract

Zinc oxide (ZnO) plays a pivotal role across various industries due to its remarkable properties, which are further enhanced when engineered at the nanoscale. In recent years, the green synthesis of nanoparticles using biological systems, particularly plant extracts, has emerged as a rapidly advancing field within nanotechnology. This eco-friendly approach aims to reduce the environmental impact associated with conventional methods. In the present study, zinc oxide nanoparticles (ZnO NPs) were synthesized using zinc oxide and an aqueous extract of *chamomile flowers* as a reducing and stabilizing agent. The field emission-scanning electron microscopy (FESEM) results of ZnO pure and ZnO green, respectively, showed that the shape of the nanoparticles is semi-spherical, and the nano size ranges between 20nm and 50nm, and atomic force microscopy (AFM) provided detailed information on the topographical features at the nanoscale. Together, these characterization techniques confirmed the successful synthesis of ZnO NPs, highlighting the potential of green synthesis methods in producing nanoparticles with desirable properties for biological applications. The ZnO NPs were made utilizing an eco-friendly technique using extracts of chamomile flower (*Matricaria chamomilla L.*) and showed their direct effect on healing the surface wounds in mice.

Keywords: Chamomile Flower, ZnO, PLAL, Green Synthesis, Mice.

التخليق الأخضر لـ ZnO NPs باستخدام مستخلص زهرة البابونج عن طريق الاستئصال بالليزر النبضي وتأثيره على الجروح السطحية

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الخلاصة

يلعب أكسيد الزنك (ZnO) دورًا محوريًا في مختلف الصناعات نظرًا لخصائصه الرائعة، والتي يتم تعزيزها بشكل أكبر عند هندستها على نطاق النانو. في السنوات الأخيرة، برز التركيب الأخضر للجسيمات النانوية باستخدام الأنظمة البيولوجية، وخاصة المستخلصات النباتية، كمجال سريع التقدم في تكنولوجيا النانو. يهدف هذا النهج الصديق للبيئة إلى تقليل التأثير البيئي المرتبط بالطرق التقليدية. في الدراسة الحالية، تم تصنيع جسيمات أكسيد الزنك النانوية (ZnO NPs) باستخدام أكسيد الزنك ومستخلص مائي من أزهار البابونج كعامل اختزال وتثبيت. كانت نتائج FESEM لـ ZnOpure و ZnO green على التوالي، حيث

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أظهر المسح أن شكل الجسيمات النانوية شبه كروي، ويتراوح حجم النانو بين 20 نانومتر و 50 نانومتر، كما قدمت المجهر الذري للقوة (AFM) معلومات مفصلة عن السمات الطبوغرافية على نطاق النانو. وقد أكدت تقنيات التوصيف هذه معًا نجاح تخليق جسيمات أكسيد الزنك النانوية، مما يسלט الضوء على إمكانات طرق التخليق الخضراء في إنتاج جسيمات نانوية ذات خصائص مرغوبة للتطبيقات البيولوجية. وقد أظهرت جسيمات أكسيد الزنك النانوية المصنوعة باستخدام تقنية صديقة للبيئة استخدام مستخلصات زهرة البابونج (*Matricaria chamomilla L.*) وأظهرت تأثيرها المباشر على التئام الجروح السطحية للفنران.

Introduction

Nanoparticles (NPs) are a group of different kinds of particulate matter that have a single dimension less than 100 nm.

They are becoming more and more common as drug delivery methods to get around issues that most medications have historically had, like poor solubility, poor bioavailability, non-specificity, and/or toxicity [1]. The properties of nanoparticles have generated a great deal of interest in biomedical research [2]. Metal nanoparticle synthesis has been described using a variety of techniques over the years, including chemical, biological (green synthesis), and physical techniques. The solid target material is covered with a thin layer and exposed to pulsed laser light in the laser ablation process. The substance is exposed to laser radiation, which breaks the solid into nanoparticles that stay in a liquid around the target and create a colloidal solution. The ablation efficiency and characteristics of the produced metal particle are influenced by a number of factors, including the laser pulse duration, wavelength, ablation time, laser fluence, and the surrounding active liquid medium. This method is comparatively easy and efficient for creating many tiny (nano-sized) particles in suspension form. By choosing the laser parameter and the liquid's composition appropriately, the properties of the nanoparticles can be altered [3,4]. In this study, zinc was chosen as it is a vital mineral that is present in the skin, hair, nails, brain, muscles, and immune system. Additionally, compared to other organic and inorganic compounds, zinc has a strong ionic contact with oxygen that is more selective and heat resistant, even if zinc oxide has a little covalent property. Zinc oxide's small size increases its rate of dissolution, which makes it more biocompatible with healthy human cells and potentially useful as an antibacterial, anti-inflammatory, and anticancer agent [5]. It was discovered that a coating of ZnO deposited on medical textile fabrics had potent antibacterial qualities, significantly lowering the risk of bacterial infections [6,7]. Green synthesis of nanoparticles is a method of green chemistry that blends plant biotechnology and nanotechnology. Metal ions are biologically reduced to create nanoparticles using plant extracts. It has been demonstrated that plant metabolites such as sugars, terpenoids, polyphenols, alkaloids, phenolic acids, and proteins are crucial for reducing metal ions to nanoparticles and maintaining their stability afterward [8,9]. One of the world's most traditional, often used, and extensively researched medicinal plants, chamomile has been suggested for a range of therapeutic purposes [10,11]. The chemical components of chamomile have an impact on its biological uses. Since fresh or dried flowers contain the majority of the active ingredients, infusions or essential oils are utilized in medical medicines [12,13]. According to pharmacological studies, chamomile flowers have several biological actions, such as antifungal, antioxidant, antiparasitic, and anti-infective qualities. This plant has been demonstrated in numerous studies to have therapeutic effects on various ailments, including skin conditions and wound healing [14]. Green synthesis of nanoparticles is increasingly preferred over conventional methods due to its compatibility with sustainable and environmentally friendly practices. This approach uses biological entities, including plant extracts as reducing and stabilizing agents, which gives it several advantages, such as environmental sustainability, as green synthesis eliminates the need for

toxic chemicals typically used in conventional methods, thus reducing environmental pollution and promoting safer manufacturing processes. Low cost and ease of preparation, as renewable natural resources such as plant extracts are used, and are inexpensive and easy to prepare, as their preparation does not require complex equipment and hazardous reagents. Biocompatibility, as nanoparticles synthesized through green methods show increased compatibility with biological systems, making them particularly suitable for medical and pharmaceutical applications [15-17]. If traditional techniques had been adopted as a point of comparison to this green synthesis using chamomile extract, the size of resultant ZnO nanoparticles would have been much smaller, while it can still pose high antibacterial efficacies worthy of both biomedical and environmental applications. While the former can be noted in somewhat reduced crystallinity with wider XRD peaks compared to, in contrast, highly crystalline material with excellent purity obtained in other traditional methods of higher energetic input values using advanced equipment like pulsed laser ablation. The aim of this study was to green-synthesize zinc oxide nanoparticles using extracts of chamomile flower (*Matricaria chamomilla* L.) and discuss their effect on healing surface wounds.

Materials and Methods

Plant extract preparation

Prepare chamomile flower extract. After being cleaned with deionized water, the flowers were dried and pulverized into a fine powder. Using a hot-stirrer, 5g of powder was mixed with 100 ml of deionized water for 30 minutes at 70°C. The choice of 70°C and 30 min for extract preparation is often based on optimizing the balance between efficiently extracting the desired active compounds and maintaining their stability. Many bioactive compounds from plants (e.g. polyphenols, flavonoids, and alkaloids) are efficiently extracted at moderate temperatures such as 70°C. This temperature helps to break down plant cell walls and release the bioactive components. After cooling at room temperature, the mixture was centrifuged for twenty minutes and then filtered the extract using filter paper, as shown in Figure 1.

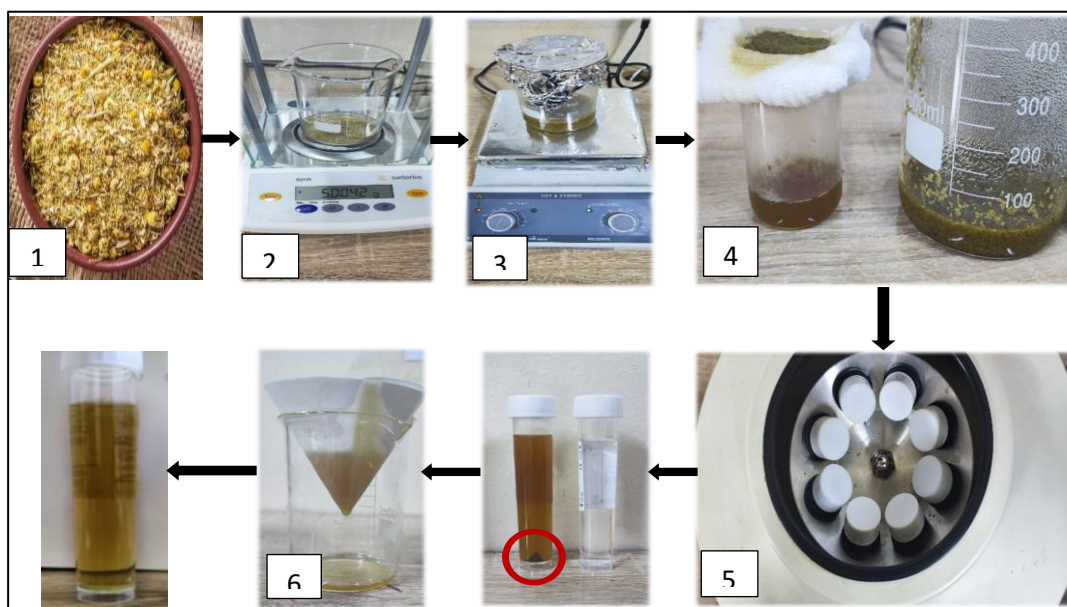


Figure 1: Steps to prepare chamomile flower extract. 1- Cleaned *chamomile* flower with deionized water then grinding it. 2- 5g weight of powder. 3- Mixed the powder with 100ml of deionized water using hot-stirrer for 30 minutes at 70°C. 4- Filter the extract with gauze. 5- Centrifuged the mixture 6- Filtered the extract using filter paper

Synthesize ZnONPs in a physical method (pulsed laser ablation PLA)

A sheet of pure zinc metal (99%) is prepared with 10 ml of deionized water. Struck the sheet using a pulsed laser beam (Nd: YAG) with a wavelength of 1064 nm, an energy of 300 mJ, a frequency of 8 Hz, and a rate of 400 shots. Figure 2 explains the steps of the work.

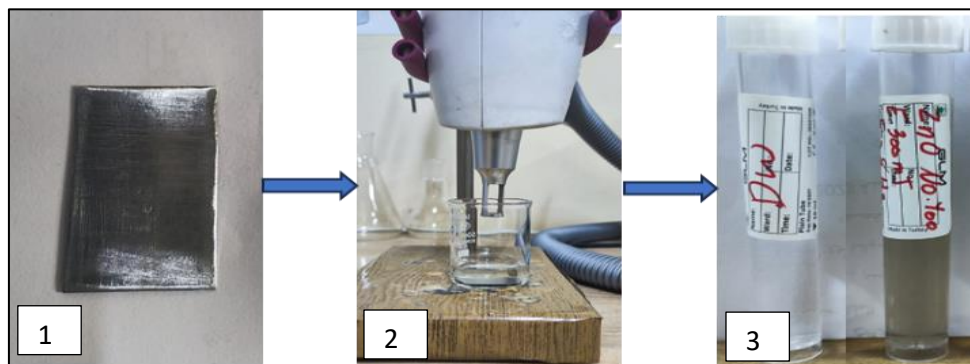


Figure 2: Steps of (PLA) pulsed laser ablation to synthesis ZnONPs. 1- A sheet of zinc metal of pure (99%). 2- Struck the sheet (Placed in a beaker with 10 ml of deionized water) using a pulsed laser beam. 3- Color change in deionized water indicates formation of ZnONPs.

Synthesize ZnONPs using a green method

Preparation is carried out with the same previous steps, but replacing the deionized water with a mixture consisting of 1ml chamomile flower extract and 9 ml deionized water. A 1:9 ratio of extract to deionized water is often used in green synthesis to balance the concentration of bioactive compounds in the extract and ensure proper dilution for efficient nanoparticle synthesis. The extract contains phytochemicals (such as flavonoids, polyphenols, or enzymes) that are responsible for reducing metal ions and stabilizing the nanoparticles. The 1:9 ratio provides a moderate concentration of these compounds, preventing insufficient or excessive bioactive content, which may affect the nanoparticles' size, shape, or yield.

In order to test the effect of the prepared zinc nanoparticles on superficial skin wounds, tests were conducted on mice, where seven groups of mice were prepared, and each group included three mice. The test was conducted for seven days, where on the first day a superficial wound of 10 ml was made for each mouse, and treatment was applied to the groups of mice as follows: The first group (-ev control) did not receive any treatment; the treatment of the second, third, and fourth groups was with ZnO NPs at a concentration of 200, 400, and 600, respectively; the treatment of the fifth group was with chamomile flower extract, the treatment of the sixth group was with green ZnO NPs, and the treatment of the seventh group was with Fucidin ointment. After 72 hours of the application, the length of the wounds made on the mice was measured using a vernier. On the seventh and last day of testing, histological images were taken of the mice.

Results and Discussion

X-Ray Fluorescence analysis

As shown in Figure 1, the XRF test findings display the amounts of the chemical components in the chamomile flower that give it its antioxidant characteristics. In the green production of nano ZnO, the plant extract directly facilitates the reduction of the metal and the stabilization of the nanoparticles. The bioactive chemicals in the extract, including flavonoids, tannins, and polyphenols, engage with the solid metal surface via corrosion or

partial dissolving processes induced by acidic environments or chemical interactions of organic molecules. These chemicals facilitate the oxidation of solid zinc into nano ZnO while creating a coating layer on the surface of the resultant particles, reducing agglomeration and maintaining stability. In the synthesis using chamomile extract, phenolic components interact with the metal surface, facilitating the progressive removal of atoms from the metal and their conversion into ZnO. This method offers a direct approach to diminish particle size and enhance stability, eliminating the need for dangerous or supplementary substances [18].

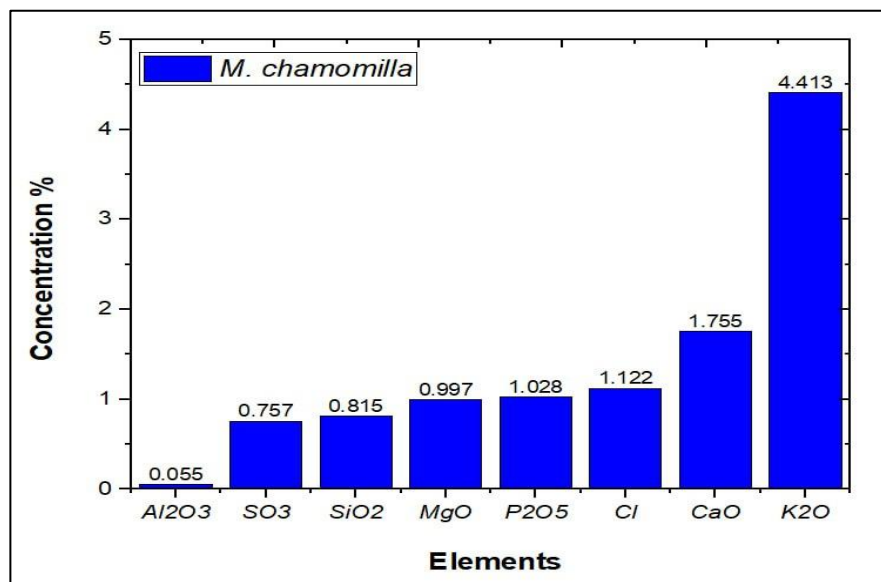


Figure 3: XRF analysis showing the concentrations of the chemical components of *chamomile flower*

UV-Vis Spectroscopy

Figure 4 shows the UV-Vis absorption spectrum of the synthesized ZnO nanoparticles, the surface plasmon resonance (SPR) appears at a wavelength of 373 nm for pure ZnO and 382 nm for green ZnO. The energy gap was calculated using Planck's law [19], where the energy gap for pure ZnO was 3.3 eV and for green ZnO was 3.2 eV.

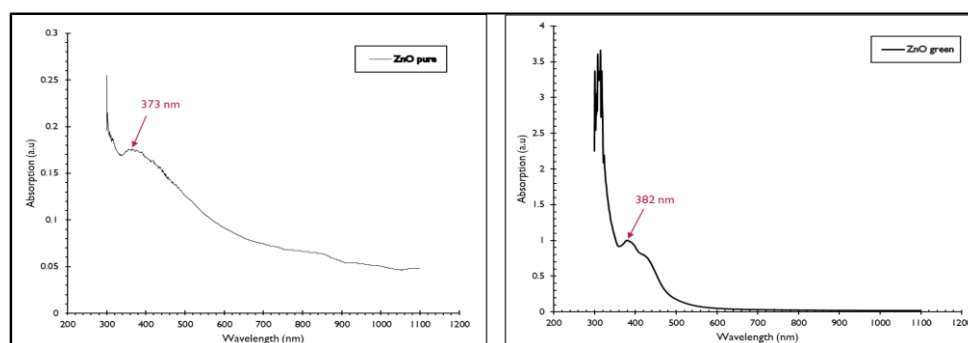


Figure 4: UV-Vis analysis shows SPR peak at wavelength 373nm for ZnO pure and 382nm for ZnO green

X-Ray Diffraction analysis

When crystalline atoms permit X-rays to diffract and interferences arise, X-ray diffraction (XRD) is an invaluable technique for ascertaining the atomic and molecular structure of the crystal. The distance between atoms in a layer or row, the orientation of the crystal, strain,

and grain size, as well as the identification of crystal structures belonging to unidentified species, can all be calculated using XRD [20].

Figure 5 displays the results of the XRD test. The identity of the material was established by comparing the values of the Miller coefficients with the values of the ZnO reference pattern. The observed values in the coordinates ($2\theta = 31.75, 34.35, 36.25, 47.45, 56.55, 62.85, 66.35, 67.95,$ and 69.05) are associated with the lattice diffraction peaks (100,002,101,102,110,103,200,112, and 201) respectively, which confirm that the structure of the nanoparticles is hexagonal wurtzite, which is consistent with the standard values of (JPCDS card number: 36-1451) [21]. The comparative study of ZnO nanoparticles synthesized by pulsed laser ablation and green synthesis using chamomile extract shows significant differences in crystallinity and particle size. In the XRD pattern, peaks for the ZnO sample prepared by pulsed laser ablation were pronounced and strong, indicating high crystallinity, very good purity, and consequently high energy involved in the synthesis of nanoparticles. In contrast, the sample prepared using the green methods presented wider and weaker peaks due to low crystallinity caused by chemical interaction of bioactive chemicals from chamomile extract but also in a hexagonal wurtzite structure. Furthermore, nanoparticles from the green method of manufacture had a smaller size that was appropriate for use in medicine and the environment. On the other hand, pulsed laser ablation showed outstanding purity and crystallinity for electrical and optical applications. This comparative study emphasizes choosing the synthesis process based on the application while balancing performance with environmental considerations.

Fourier transform infrared spectroscopy analysis

Fourier transform infrared spectroscopy (FTIR) is one of the best analytical methods. Infrared spectrograms display the absorbance against wavelengths ranging from 700 to 400 000 nm and wavenumbers between 14,000 and 25 cm^{-1} . The molecules vibrate more as the compounds absorb energy, expanding and bending according to their geometry. The absorption spectrum pattern can be compared to a fingerprint that uniquely identifies a molecule, making it amenable to both qualitative and quantitative study [22]. FTIR examination revealed the absorption spectra and chemical bonds of chamomile extract, ZnOpure, and ZnOgreen samples. Figure 6 shows the spectrum of the mentioned samples, and Table 1 shows the chemical bonds and the appearance of ZnOpure at wavenumber (549.21 cm^{-1}) and ZnOgreen at wavenumber (468.60 cm^{-1}). Chamomile extract contains certain chemicals, such as phenols, that could interact with the zinc surface. The latter may establish non-covalent interactions like hydrogen bonding or interact with the -OH groups on the zinc oxide surface. This might increase stability and inhibit agglomeration. Moreover, the chamomile extract can act as a medium for modifying the surface contacts of zinc oxide. Resultant nanoparticles with less roughness may provide the possibility for interactions on the surface, leading to the creation of protective layers or coatings, hence affecting the behavior of the particles in medical applications such as wound healing.

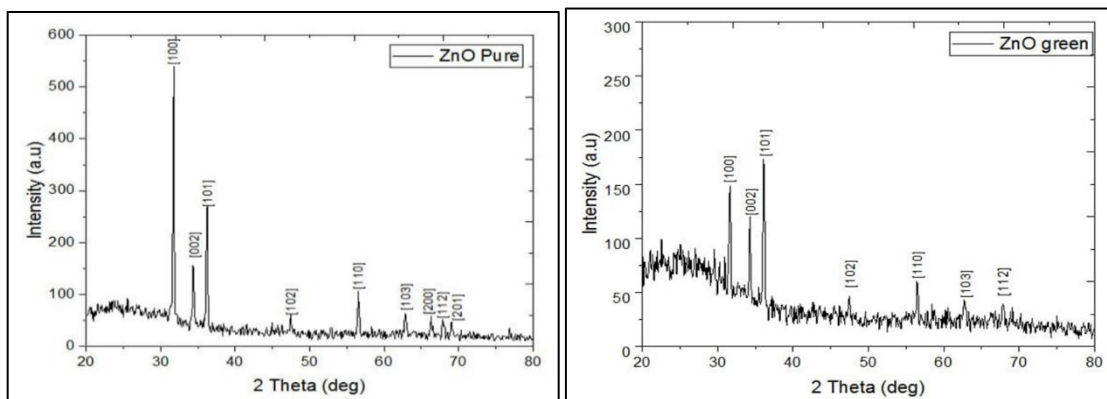


Figure 5: XRD pattern of ZnOpure and ZnOgreen

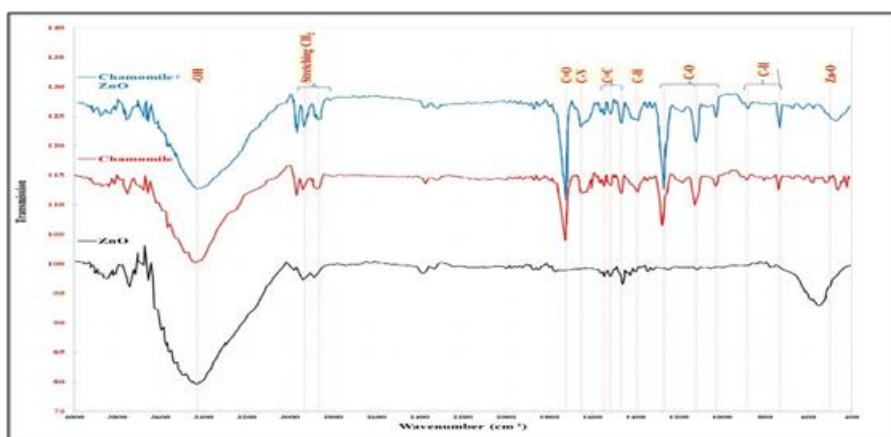


Figure 6: FTIR absorption spectra of the chamomile flower extract, ZnOpure and ZnO green.

Table 1: The chemical bonds of the extract, ZnOpure and ZnOgreen

Band Type	Zn-O	Chamomile	Chamomile + ZnO
O-H	3439.30	3439.16	3428.87
C-H stretch	2940.07	2938.35	2931.49
C-H stretch	2886.90	2871.46	2864.60
C=O	-	1724.06	1722.34
C-N	-	1646.88	1652.02
C=C	-	1543.97	1543.97
	-	1514.82	1516.53
	-	1465.08	1465.08
C-H	-	1389.61	1393.04
C-O	-	1276.42	1267.84
	-	1123.77	1120.34
	-	1029.44	1026.01
C-H	-	883.66	881.94
ZnO-O	549.21	-	468.60

Field Emission-Scanning Electron Microscopy

Scanning electron microscopy (SEM) is a highly adaptable, esteemed, and widely used method in both the industrial and research domains. This type of electron microscopy produces images with high magnification and resolution by scanning the samples with a high-energy electron beam. This technique provides a multitude of data about the materials being studied, such as their elemental makeup, surface morphology, and crystallinity, among other things. A variation of standard scanning electron microscopy (SEM) that offers a wider energy range and higher resolution images is called field emission scanning electron microscopy (FE-SEM) [23,24].

Figure 7 and Figure 8 shows the FE-SEM results of ZnO pure and ZnO green, respectively, where the scanning showed that the shape of the nanoparticles is semi-spherical, and the nano size ranges between 20nm and 30nm. The results also showed that the extract coated the nanoparticles, determined their size, and preserved them from agglomeration, as the extract is an insulating material that prevents the particles from attracting and agglomerating. Several nanoparticles, such as zinc oxide, exhibit antibacterial activity that prevents infection, which could delay the healing process. They also contribute to the facilitation of wound healing since some nanoparticles can enhance tissue regeneration and, therefore, accelerate the healing process. Besides, nano-zinc possesses antibacterial activity that reduces the chance of infection in wounds.

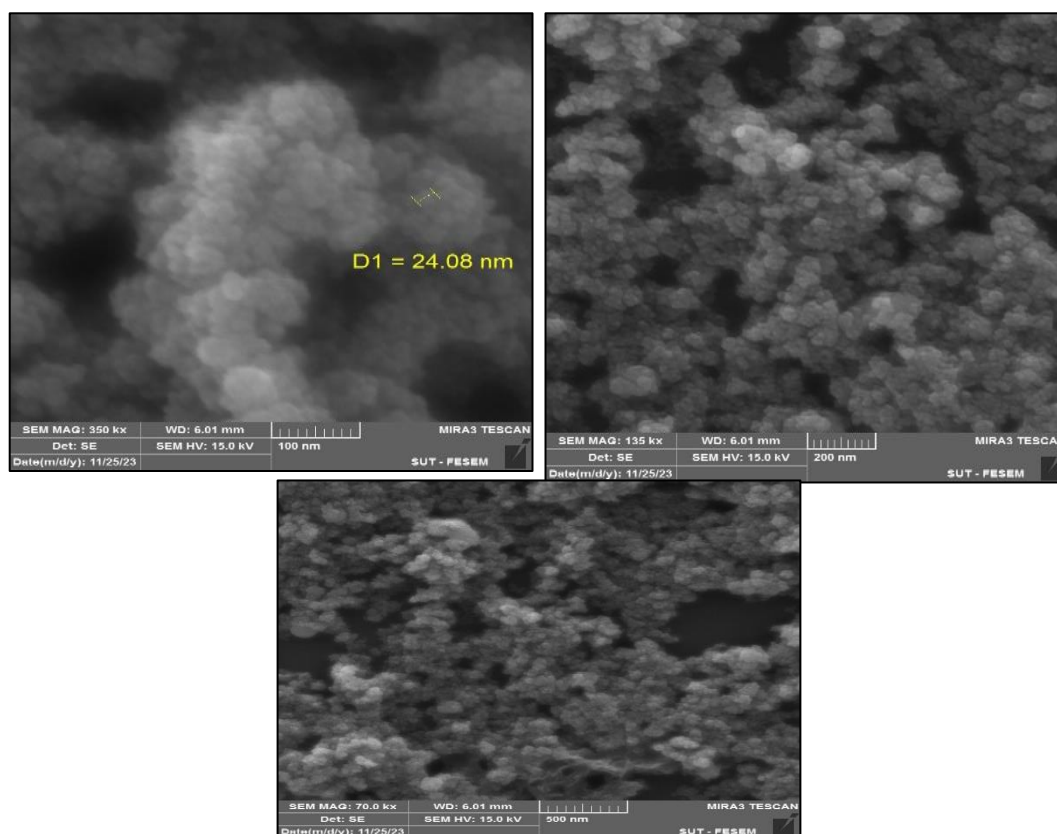


Figure 7: FE-SEM images of the ZnO pure show: The formation of nanoclusters representing Zn nanoparticles

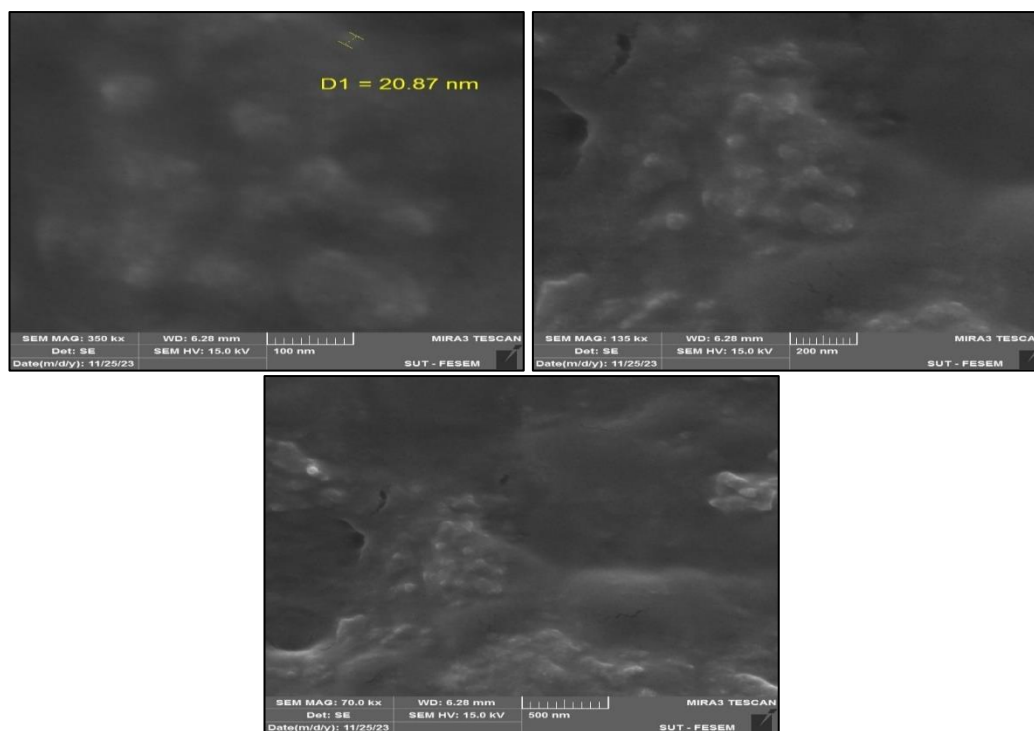


Figure 8: FE-SEM images of the ZnO green show: The formation of nanoclusters representing Zn nanoparticles

Energy Dispersive X-Ray Spectroscopy analysis

This method is used to determine the specimen's elemental composition. The specimen was subjected to an electron beam bombardment using the scanning electron microscope during the EDX investigation. Every element's atom releases different energy levels of X-rays during the collision process. Thus, measuring the energy of the X-rays emitted by a specimen during electron beam bombardment can reveal the identity of the atom from which the X-ray originated [25,26]. Based on the EDX plot of the SEM image, Figure 8 displays the elemental composition analyses of ZnOpure and ZnOgreen, respectively. The resulting zinc nanoparticles were verified to be exceedingly pure by EDX spectra, and the molecular weights of the material were revealed; its identity was established by determining the chemical elements and their distribution on the surface. Plant chemicals can also coat the nanoparticles' surfaces, reducing the detectable amount of zinc in the EDX measurement. This coating might be too tiny or not recognizable in the study, resulting in a lower concentration of zinc than the actual concentration in normally produced particles, as shown in the figure 9.

Atomic Force Microscopy analysis

Sample surfaces are analyzed using AFM (atomic force microscopy). In contrast to scanning electron microscopy and transmission electron microscopy, this method, which depends on the interactions between atoms, is comparatively non-destructive. It can also create three-dimensional maps of the surface. Ten-tenths of a nanometer, or 10^{-10} m, atomic resolution can be obtained by the use of AFM [27]. The results of the AFM analysis show the roughness, root mean square, and average diameter for each ZnO pure and ZnO green, as shown in Figure 10 (a,b). The results show that the average grain diameter of ZnO pure is 39.45 and that of ZnO green is 73.13, the roughness of ZnO pure was 3 and that of ZnO green was 2, while the (R.M.S) of ZnO pure was 4 and that of ZnO green was 5, as shown in Table 2. According to these results, it is clear that ZnO green is less rough, indicating that the plant

extract (chamomile extract) coated the ZnO nanoparticles and prevented them from attracting each other and causing agglomerations. A lower value of surface roughness would mean a smoother surface of ZnO and therefore enhanced biocompatibility with human tissue cells. This implies that the nanoparticles interact more effectively with cells at a wound site, hence facilitating cell and tissue growth and regeneration. Consequently, a smooth surface of ZnO may improve particle interaction with proteins and cells at the wound site, increasing the ability of the body to regenerate tissue and speeding up the healing process.

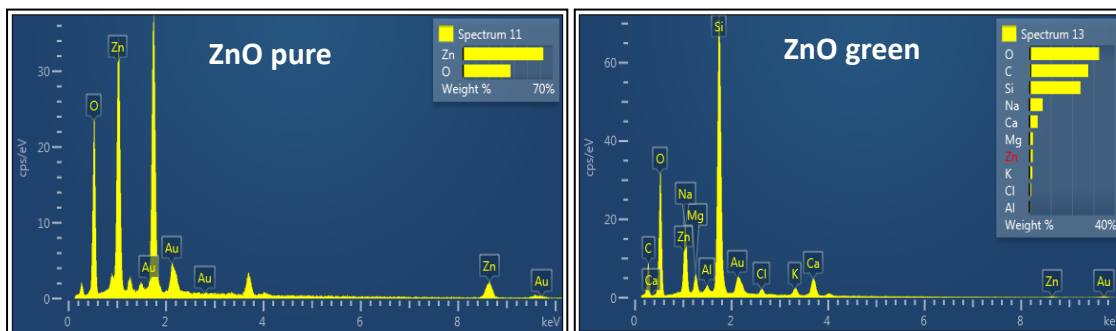


Figure 9: EDX pattern of ZnOpure and ZnOgreen NPs shows the components of the nanocomposite and proves the purity of the compound

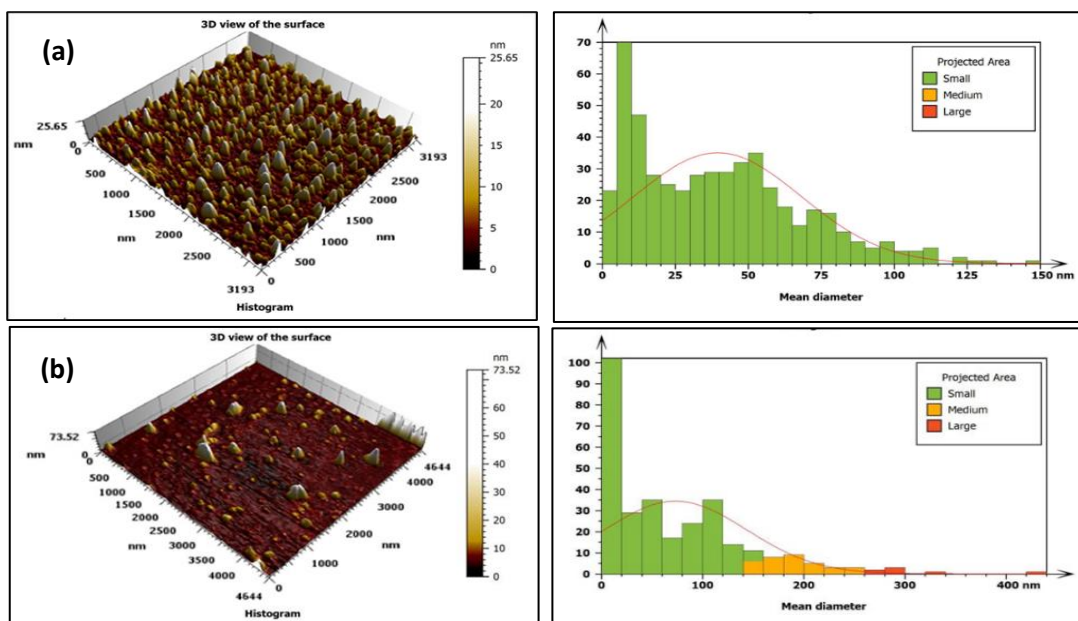


Figure 10: (a): AFM and their granularity accumulation distribution for ZnO pure NPs (b) AFM and their granularity accumulation distribution for ZnO green NPs

Table 2: Shows the difference in Avg.d, Roughness and R.M.S values between ZnOpure and ZnOgreen NPs

Sample	Avg. d	Roughness	R.M.S
ZnO Pure	39.45	3	4
ZnO green	73.13	2	5

Effect of zinc oxide nanoparticles prepared by physical and green methods on wound healing

After conducting all the required tests on the zinc oxide nanoparticles manufactured by physical and green methods, as well as identifying and researching their properties, mice

were used to evaluate the ability of zinc oxide nanoparticles to heal wounds. Five groups of mice were prepared, A 10 ml wound was made for the mice, and the treatment was given for a week, seven days after the start of treatment, histological examination images were collected, and the results were as follows, the first group (G1) was a control group, the histopathological figures of the skin revealed thick layer of necrotic tissue covered the injured epidermis with focal dermatitis that infiltration of mono nuclear leukocytes and marked line of reepithelization revealed immature epidermal stratified keratocytes. The dermis revealed mature fibrous connective tissue with normal constituents of fibrocytes, fibroblast sebaceous glands with hair follicles (Figure 11 a,b). The second group (G2) was treated with zinc nanoparticles prepared by the physical method, the histopathological figures of the skin revealed thin line of newly regenerated epidermis that composed of non-keratinized stratified squamous epithelium, the dermis composed of mature fibrous connective tissue with marked formation of hair follicle and little degree of collagen degeneration (Figure12 a,b). The third group (G3) was treated with chamomile flower extract; the histopathological figures of the skin revealed normal stratified squamous epidermis cells. The dermis is composed of dense mature fibrous tissue with fibroblasts (fibroplasia), proliferation of hair follicles with sebaceous glands, and no dermatitis or inflammatory reaction (Figure 13 a,b). The fourth group (G4) was treated with zinc nanoparticles prepared by the green method; the histopathological figures of the skin revealed marked irregular epidermal hyperkeratosis that showed marked hyperplasia of keratocytes with little figure of degeneration. The dermis revealed normal mature fibrous connective tissue with marked proliferation of hair follicles with sebaceous glands (Figure 14 a,b). The fifth group (G5) was treated with Fucidin ointment; the histopathological figures of the skin revealed a marked very thin epidermis. The dermis revealed marked degeneration of fibrous connective tissue with necrosis of hair follicles and sebaceous glands (Figure15 a,b). Fibroblasts are important cells during wound healing and play a significant role in the synthesis of collagen and extracellular matrix. Green ZnO thus suggests that nano zinc oxide may facilitate cell proliferation at various levels by stimulating cellular growth. Some reports pointed out that micro ZnO may enhance the proliferation of fibroblasts due to increased cellular activity or by influencing any signaling pathways like MAPK or PI3K/Akt, which are part of cell growth. Besides the enhancement of growth factor generation, green nano zinc oxide has favored the growth factor release of TGF- β , which potentially initiates the cell proliferation and tissue regeneration [28].

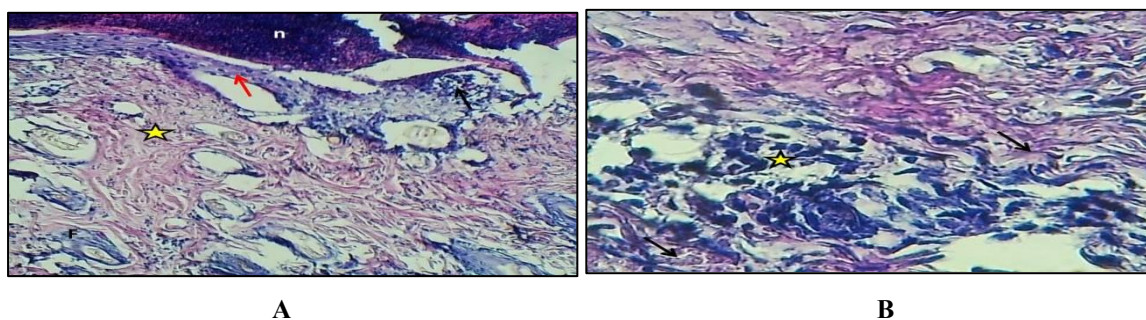


Figure 11.(A): section of skin (G1) shows: thick layer of necrotic tissue (n), focal dermatitis with infiltration of leukocytes (Black arrow), line of reepithelization revealed immature stratified keratocytes (Red arrow), dermal mature fibrous connective tissue (Asterisk) & sebaceous glands (F). H&E stain.100x. (B): section of skin (G1) (Control negative) shows: normal epidermis keratinized stratified squamous epithelium (E), dermal collagen fibers (Asterisk) & hair follicle (arrows). H&E stain.400x.

As the predominant cell type in the uppermost strata, keratinocytes play a very important role in wound healing. This cell type needs to migrate from the edge of the wound to the site of injury so that closure of the wound and restoration of skin integrity may be achieved. Green ZnO nanoparticles might influence the migration of keratinocytes by the activation of cell signaling pathways such as MAPK/ERK and NF- κ B, which are involved in the inflammatory response and promoting migration. Phenolic or flavonoid chemicals from plant extracts, such as chamomile extract, could interact with the surface of green ZnO nanoparticles and directly enhance the activity of keratinocytes by interacting with the surface ligand or cellular receptors [29].

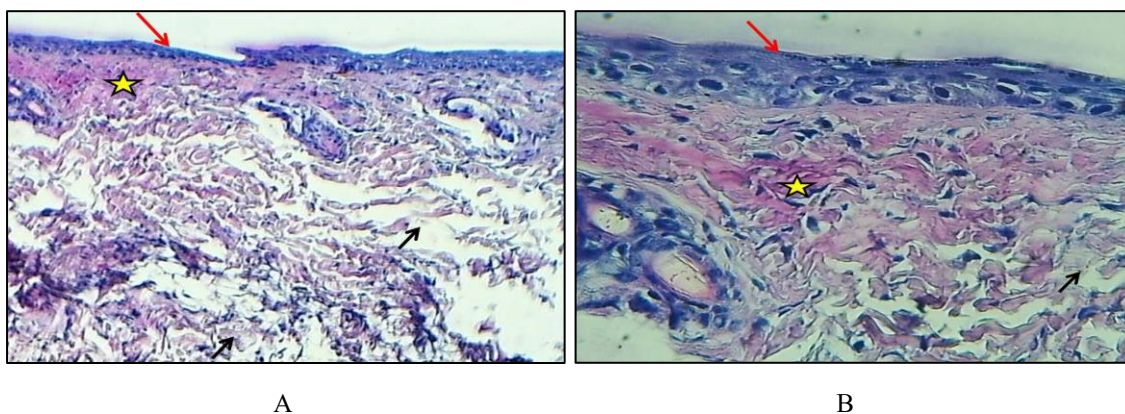


Figure 12.(A): section of skin (G2) shows: thin line of newly regenerated epidermis (Red arrow), mature fibrous connective tissue of dermis with marked formation of hair follicle (asterisk) little degeneration of collagen fibers (Black arrows). H&E.100x. (B): section of skin (G2) shows: thin line of newly re-epithelization epidermis (Red arrow), mature fibrous connective tissue of dermis with marked formation of hair follicle (asterisk) little degeneration of collagen fibers (Black arrows). H&E.400x.

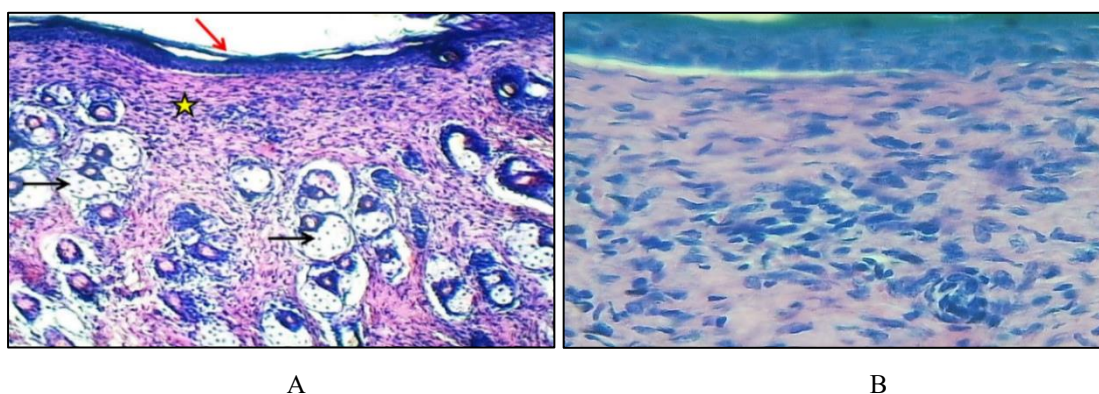


Figure 13.(A): section of skin (G3) shows: normal epidermis (Red arrow), mature fibrous connective tissue of dermis with fibroplasia (asterisk) with proliferation of hair follicle (Black arrow). H&E.100x. (B): section of skin (G3) shows: normal stratified squamous cells, mature fibrous connective tissue of dermis with fibroplasia. H&E.400x.

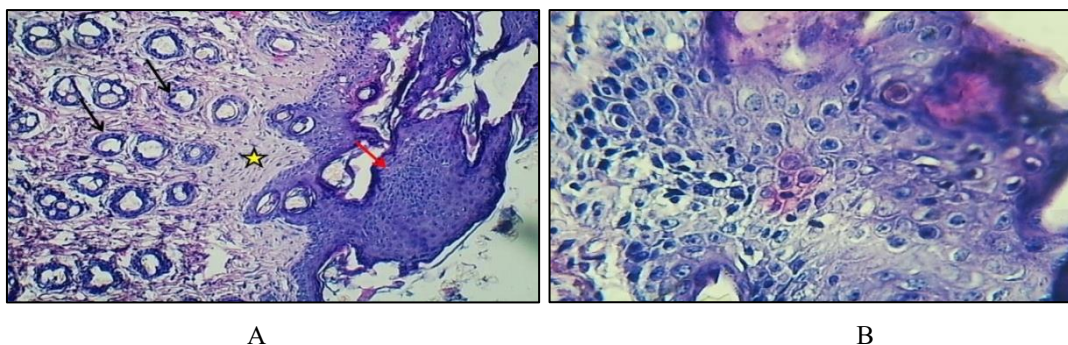


Figure 14: (A): section of skin (G4) shows: thick irregular epidermal proliferation (Red arrow), mature fibrous connective tissue of dermis (asterisk) with proliferation of hair follicle (Black arrow). H&E.100x. (B): section of skin (G4) shows: hyperkeratosis with degeneration of keratinocytes. H&E.400x.

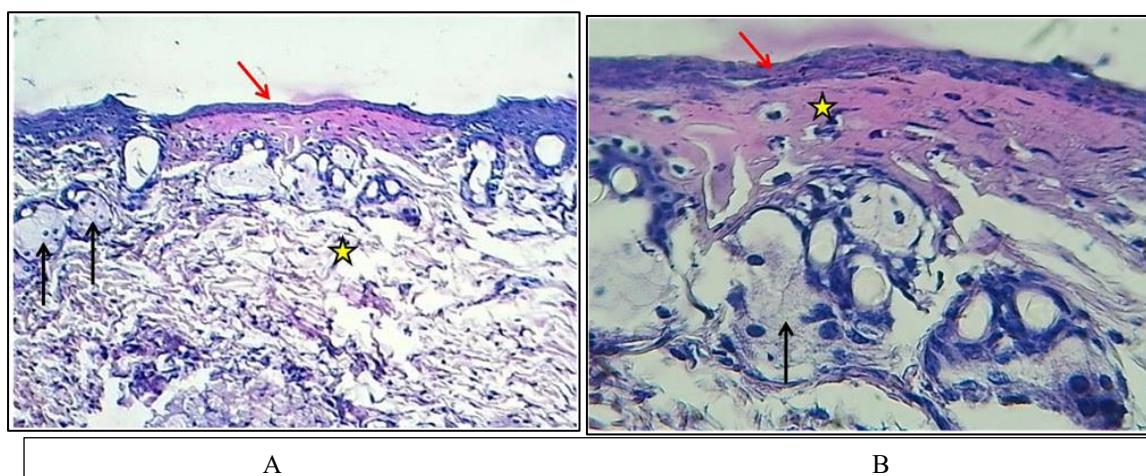


Figure 15:(A): section of skin (G7) shows: poor reepithelization of epidermis (Red arrow), degeneration with necrosis of fibrous connective tissue (asterisk) with little normal sebaceous glands (Black arrow). H&E.100x. (B) section of skin (G5) shows: poor reepithelization of epidermis (Red arrow), necrosis of fibrous connective tissue (asterisk) with degeneration of sebaceous glands (Black arrow). H&E.400x.

Conclusions

The series of techniques, like XRF, UV-visible, XRD, FE-SEM, EDX, FTIR, and AFM, confirmed the successful synthesis of Zinc oxide (ZnO) NPs pure and ZnO NPs using green technique by Pulsed Laser Ablation in Liquid. This novel synthesis method enhances the biological activities of the nanoparticles while additionally offering an eco-friendly approach, owing to the use of chamomile flowers extract as a capping and reducing agent. The FE-SEM pictures show that both the ZnO pure and ZnO green nanoparticles have a semi-spherical morphology with a size distribution of 20 to 30 nm. Using the EDX spectra, the resulting zinc nanoparticles were verified to be exceedingly pure, and the molecular weights of the material were revealed, and its identity was established by determining the chemical elements and their distribution on the surface. AFM analysis showed the surface roughness of ZnO green is less than that of pure ZnO, indicating that the chamomile extract coated the ZnO nanoparticles, inhibiting their attraction, which causes agglomeration. These particles can be used in medical applications. The use of ZnO NPs and chamomile flower (*Matricaria chamomilla* L.) showed their direct effect on healing the surface wounds for mice, where phenolic or flavonoid chemicals from chamomile flower extract can interact with the surface of green zinc oxide nanoparticles and directly enhance the activity of keratinocytes, thus enhancing the healing process.

Conflict of Interest

The authors declare no conflicts of interest.

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