

# Antibacterial Efficiency of the Extracts of *Euphorbia condylocarpa* Tuber Roots and Leaves on Three Multidrug Resistance Bacterial Isolates using Green Synthesis of Zinc Oxide Nanoparticle Techniques

Halala R Qader<sup>1</sup>, Abdulghany O I Sarmmamy<sup>2</sup>

<sup>1</sup>Department of Health and Environmental Science, Salahaddin University, Erbil, Iraq

<sup>2</sup>Department of Biology, College of Science, Salahaddin University, Erbil, Iraq

Received: 23<sup>rd</sup> Jun, 2025; Revised: 11<sup>th</sup> Aug, 2025; Accepted: 23<sup>rd</sup> Aug, 2025; Available Online: 25<sup>th</sup> Sep, 2025

## ABSTRACT

The perform study was executed to inspect the phytochemical complement, antimicrobial activity of chloroform (ChL), ethyl acetate (EA), as well as ethanol (ET) extracts in the leaves and root tubers of spurge *E. condylocarpa* along with green synthesis, characterization and antibacterial attain of zinc oxide nanoparticles on multidrug-resistant bacterial isolates such as *staphylococcus aureus*, *Pseudomonas aeruginosa*, and *Escherichia coli* by Minimum Inhibitory Concentrations (MIC). The inclined ZnO NPs were categorized along UV-vis, FT-IR, SEM, XRD, together with EDX. This experiment applied in the laboratories of Biology Department, College of Science Salahaddin University, and conducted according to a completely randomized design (CRD) with three replications. *E. condylocarpa* leaves and root tubers have been retainer as reducing, capping and stabilizing agents, the configuration of ZnO NPs portrait by shift in color from dark yellow to yellowish white. X-ray diffraction (XRD) valid the crystallinity of ZnO NPs. FT-IR spectroscopy corroborated the functional groups of leaves and root tubers extracts Qualitative phytochemicals analysis focuses on the presence or absent of several chemical groups in the plant parts. Total condensed tannin, and total phenol was done in both parts. The results showed that the amount of total condensed tannin was more in leaves as comparison to root tubers, while total phenolic content in ethanol extract more than in ethyl acetate then in chloroform in both leaves and root tubers. The crude extracts of leaves and root tubers showed that the plant contains different phytochemicals such as terpenoids, steroids, saponin, flavonoids, phenol, tannin, coumarins, quinone, and alkaloids. The lowest significant MIC values with *E. coli* was 4.17 mg/ml for ethanol leaf extract of *E. condylocarpa*.

**Keywords:** *Euphorbia condylocarpa*, ZnO Nanoparticles, Antibacterial activity, Medicinal plants, Bacteria.

**How to cite this article:** Halala R Qader, Abdulghany O I Sarmmamy. Antibacterial Efficiency of the Extracts of *Euphorbia condylocarpa* Tuber Roots and Leaves on Three Multidrug Resistance Bacterial Isolates using Green Synthesis of Zinc Oxide Nanoparticle Techniques. International Journal of Drug Delivery Technology. 2025;15(3):1006-16. doi: 10.25258/ijddt.15.3.15

**Source of support:** Nil.

**Conflict of interest:** None

## INTRODUCTION

Depending on their attending uses, aromatic and medicinal plants can be sold as both fresh and dried goods. Additionally, medicinal plants are utilized as raw elements to make pharmaceutical industry- marked medications<sup>1</sup>. Plants create bioactive substance called phytochemicals to defend themselves. Some important phytochemicals include carotenoids polyphenols, isoprenoids, phytosterols, saponin, dietary fibers, and certain polysaccharides. They can be obtained from foods, including whole grains, fruits, vegetables, nuts, and herbs<sup>2</sup>. *Euphorbia condylocarpas* root is used in traditional medicine to cure cancer, migraines, and costiveness. It is also used as an emollient and to extract various phenolics from the plants root<sup>3</sup>. Phytochemicals are the bioactive natural chemical substances that give plants their medicinal properties. Primary and secondary metabolites are the two general categories into which phytochemicals fall<sup>4</sup>. The glabrous perennial herb spurge (*Euphorbia condylocarpa* M. Bieb.) is a portion of family Euphorbiaceae, which is one of most significant groups of

medicinal in the world. Antioxidant flavonoids, especially flavones, anticancer effects, antibacterial effects, anti-inflammatory, wart treatments, decrease risk of blood cancer, osteoporosis, and wound healing are some of this plant's therapeutic qualities. In Turkey, Iran, Northern Iraq, and the Caucasus (Georgia, Armenia, Azerbaijan, and Russia)<sup>5,6</sup> *Euphorbia condylocarpa* grows in limestone on mountain slops in oak trees, numerous Iraqi forest zones, such as Rowanduz Gorge, Gali Warte, Haji Omaran, Darbandi Basian, and Qopi Qaradax, have documented biodiversity of various kinds of this plant<sup>7</sup>. In traditional medicine *Euphorbia condylocarpa* root and leaf extracts are used in treat constipation, gonorrhea, and skin conditions<sup>8</sup>. Another options to antibiotics, nanoparticles are being employed progressively to intent bacteria. Using nanoparticles in bacterial coatings and pharmaceutical products to stop infections, as well as encourage wood healing are two cases to how nanotechnology may be very helpful in serving bacterial infections<sup>9</sup>. Multi- drug resistance bacteria have emerged as a direct result of the

\*Author for correspondence: halala.qader@su.edu.krd

extensive usage of antibiotics, according to several studies, therefore attention focused on use of nanoparticles with antibacterial activity<sup>10</sup>. The nanoparticles functional antibacterial activity stems from their inherit high- surface to volume ratio due to their smaller particle size. These characteristics ensure close bonding interactions between nanoparticles and microbial membrane. The relatively large surface area and small size of NPs enables them to penetrate cell membrane of pathogens increase their antibacterial effectiveness<sup>11</sup>. Zinc oxide (ZnO) nanoparticles represent a noble class of inorganic nanomaterials, exhibiting a range of unique properties such as photocatalytic effects, antioxidant abilities, antibacterials and antifungal activity making them highly valuable across various research domains<sup>12</sup>. According to<sup>13</sup> zinc oxide nanoparticles inhibit the proliferation of bacterial species such as *E. coli*, *S. aureus*, and *P. multiseptus* bacterial and fungal species<sup>14</sup> investigated that there several variables which effect on the green synthesis of nanoparticles from plant extract such as solvents, temperature, PH, and pressure levels.

The intention of this study examines the antibacterial tracts of nanoparticles produce across green synthesis, along with ethanol, ethyl acetate, chloroform leaves besides tuber root extract of *E. condylocarpa* against three multidrug resistant pathogenic bacteria including *S. aureus*, *P. aeruginosa*, and *E. coli*. Additionally phytochemical screening was also conducted in both parts.

## MATERIALS AND METHODS

### Extraction of Plant

The fresh plant root tubers and leaves were harvested, washed thoroughly in water and shade dried for 14 days for root tubers and 7 days for leaves. The dried plant was milled to fine powder and stored in a tight plastic bottle until needed for extraction purposes. Plant extracts were prepared by using three different solvents (Chloroform, Ethyl acetate, and Ethanol), as elucidate in figure (2)

The extraction of plant parts (leaves and tubers) of *E. condylocarpa* M. Bieb., concluded by Soxhlet methods. In this method, 10 grams of dried powdered materials (leaves and root tubers) were subjected with extraction by three different solvents (ethanol, ethyl acetate, and chloroform) by using a Soxhlet apparatus at 60 °C for 8 hours for tubers



Figure 1: The whole *Euphorbia condylocarpa* plant

Table 1: Yield of crude extracts of different solvents of leaves and root tubers of *E. condylocarpa* M. Bieb.

Plant parts	Dry weight (g)	Chloroform extract (g)	Ethyl acetate (g)	Ethanol extract (g)
Leaves	10.0	0.75 b	0.65 c	0.93 a
Tuber root	10.0	0.68 b	0.47 c	0.74 a

The means in each category of column followed by the same letters are not significantly different at  $p < 0.05$  according to Duncan's Multiple Range test.

Table 2: The percentage of yield of crude extracts of different solvents of leaves and root tubers of *E. condylocarpa* M. Bieb.

Plant parts	Yield %		
	Chloroform	Ethyl acetate	Ethanol
Leaves	7.5 b	6.4 c	9.2 a
Tuber Root	6.8 b	4.7 c	7.3 a

The means in each category of column followed by the same letters are not significantly different at  $p < 0.05$  according to Duncan's Multiple Range test.

and 4 hours for leaves. A rotary evaporator. The performed extracts were clarified and condensed by operating a rotary evaporator operating at 40°C with reduced pressure was used to filter and concentrating the resulting extracts. Before being employed, the extract were kept in the freezer<sup>15</sup>.

### Determination of Plant Extract Yields

The yield of evaporated dried *E. condylocarpa* leaf, and root extract based on the dry weight of the extract was calculated as described by<sup>15</sup> from the subsequent calculation:

Yield (g/10 g of dry plant materials) = weight of the extract(g)  $\times$  100/weight of the dry plant material.

### Nanoparticle Preparation

#### Preparation of Plant Extract

The Fig. (3) provides a pictorial representation of the plant extraction procedure. It can be seen that the fresh leaves and root tubers of *E. condylocarpa* plant were dripped with distilled water besides left in a dried place for a while to be dried. After that, the dried leaves and root tubers were crushed along with converted into powder. To obtain its finest form, the resulting powder was sieved through a mesh screen. After the process of shading, drying, and grinding, (5) grams of each dried leaves and root tubers were mixed with 100 ml of deionized water. Then, the coalesce was boiled for 30 minutes at 60 °C by using a magnetic stirrer with a hot plate. Next, the mixture was put under reflux conditions to be extracted. After that, the aqueous plant extract of the leaves and root tubers were gained by filtering the mixture using filter paper Whatman No. 1. Then, this aqueous plant extract was prepared to be used in further step testing<sup>16</sup>.

#### ZnO NPs Synthesis by Green Methods

Zinc oxide NPs were generated via green synthesis methods as outlined in (Fig. 3). In a beaker dissolve 2gm of zinc sulphate in 100 ml of distilled water and maintain stirring for 20min at 60°C, subsequently, add 100 ml of plant extract dropwise to the solution, followed by the addition of 1 M NaOH solution until the color transitions from dark yellow

Table 3: Qualitative Phytochemical screening of varied extracts of *E. condylocarpa* in leaves and root tubers

compound extract	plant parts	Terpenoid	steroid	Saponin	Flavonoids	phenol	Tannin	Coumarin	quinone	alkaloid
Ethanol	Leaves	+	+	-	+	+	+	+	+	+
	Tuber root	+	+	-	+	+	+	+	+	+
Ethyl acetate	Leaves	+	+	+	+	-	+	-	+	-
	Tuber root	+	+	+	+	+	+	+	+	-
Chloroform	Leaves	-	+	+	-	-	-	-	+	+
	Tuber root	+	+	+	+	+	+	+	+	+

Key: + = present; - = absent

to yellowish-white. Upon resolution of the reactions, 50ml of deionized water was introduced to the paste and allowed to sort out for 24 hours to rushed the nanoparticles (Fig.4), which were subsequently dried in an oven at 90°C to isolate the nanoparticles. The paste was placed in a crucible and calcinated at 400°C for 2 hours to obtain clear ZnO NPs. To terminate the basic solution, the contents were thoroughly and constantly washed through distilled water<sup>17</sup>. The powdered samples were kept in an area that's dry for beyond characterizations.

#### Culture Media and Activation of Bacterial Strains

For the cultivation of bacteria, the media of Muller Hinton broth was used. The inoculum was primed by transferred numerous single colonies of bacteria into a sterile broth media furthermore crossbred together, all at once incubated at 37°C for 24 hr. inoculum of the culture solution was accommodated to the Mc Farland scale 0.5 and verified by spectrophotometer at 580nm, which is equal to  $12 \times 10^8$  CFU/ML for bacteria. The initial solutions were diluted in 1:10 ratio for the suspension of bacteria<sup>18</sup>.

#### Preparation of Bacterial Test Isolates

For this study, three multidrug-resistant bacteria were sort out, which were one gram-positive bacteria *Staphylococcus aureus*, with two gram-negative bacteria *Pseudomonas aeruginosa*, *Escherichia coli*. The microorganisms were acquired from the laboratory of microbiology, at the Salahaddin University-Erbil, College of Science. Each strain was grown in a separate test tube contains 5m of sterile Muller Hinton broth at 37 for 24 hours.

#### Activation of Pure Farms of Microorganisms

The insulate of the three bacterial species were actuated for 24 hours before testing the plant extracts and the

Table 4: The ZnO NPs particle size calculation using Dybe-Scherers formula along data form

No. of peaks	Planes	Pos. (°2Th.)	FWHM (°2Th.)	Size (nm)
1	100	31.8024	0.8659	10.4
2	002	34.5111	0.6298	14.02
3	101	36.2124	0.2755	32.34
4	111	44.7127	0.6298	14.48
5	102	47.5555	0.7872	11.63
6	110	56.6148	0.6298	15.21
7	103	62.9501	0.7872	12.48
8	112	67.9333	0.7872	12.83
average size				15.42

nanoparticles, Muller Hinton broth media was used to activate microorganisms.

#### Assessment of Antimicrobial Activities of Plant Extract and ZnO NPs by Microdilution

Antibacterial exertion of extracts of the plants and nanoparticles were assayed by the Microdilution method in accordance with to former ways with, that used for ascertain of the Minimum Inhibitory Concentration (MIC) of the leaves as well as root tubers of *E. condylocarpa* M. Bieb. The assessment was carried out in 96-well barren microplates, afterwards 100µl of Muller Hinton broth was introduced to each well of the microplate. The first well was filled with 100 µl of the sample. Next, by transferring 100µl content from the initial well into the next well of the same row of the microplate, next to adding the content of the well, here procedure was frequented for the following well of the analogous row to obtain serial dilutions (3.125, 6.25, 12.5, 25, 50 mg/ml). In this instance, the concentration of the sample will decline, while the broth media with pure

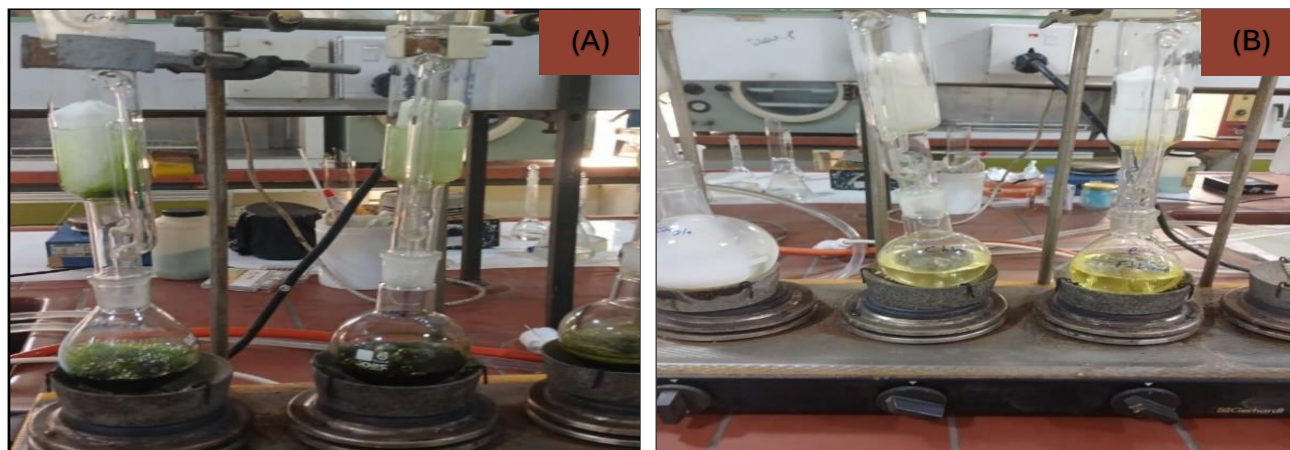


Figure 2: Soxhlet extraction of *E. condylocarpa* (A) leaves, (B) root tubers using (chloroform, ethyl acetate and ethanol) solvents





Figure 3: Preparation process of ZnO nanoparticles from leaf and tuber roots of *E. condylocarpa*

dimethyl sulfoxide were handled as a negative control, whereas Ciprofloxacin (antibacterial) was deliberated as references. A microbial suspension of the working solution was infused in each well. The absorbance for each well was deliberated at 630nm before incubation time: by using ELISA Microplate Reader, the microplate was incubated at 25 °C for 24 h for growth of bacteria. The absorbance was measured again after incubation time to approach with initial measurement. By contrasting the absorbance before and after incubation time, the MIC was assessed at the point of concentrations. The experiment was carried out in triplicating for each microbial growth.

#### *Qualitative Analysis of Phytochemicals in Leaves and Tuber of E. condylocarpa*

According to the methodology adopted by<sup>19</sup>, plant parts (leaves and root tubers) extract crudes were fulfilled in its own rudiments solvents (mg/ml) (ethanol, ethyl acetate, and chloroform), to obtain stock solution. For phytochemical analysis the stock solution employed according to the standard procedure as described as follows:

##### *Detection of Terpenoids*

The prevalence of terpenoids is verified by the emergence of a reddish or brown interface, by melding 0.5 ml of extracts of plant due to 2 ml of concentrated sulphuric acid and chloroform V/V,

##### *Detection of Steroids*

The existence of steroids is proved by adding 1 ml of sulphuric acid besides 2 ml of chloroform to 0.5 ml of plant

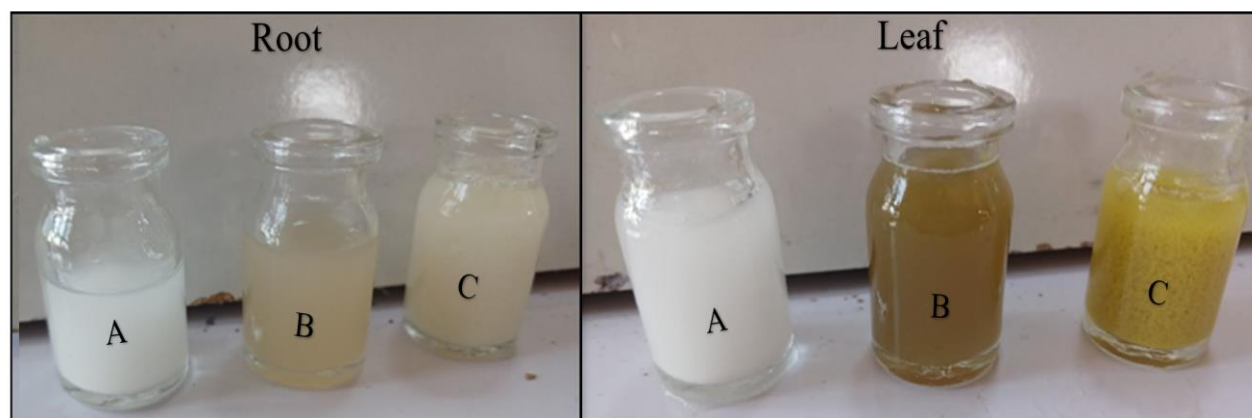


Figure 4: Discoloration of zinc sulphate after addition of plant extract (A) zinc sulphate solution, (B) plant extract (Root and Leaf), (C) nanoparticles with plant extract

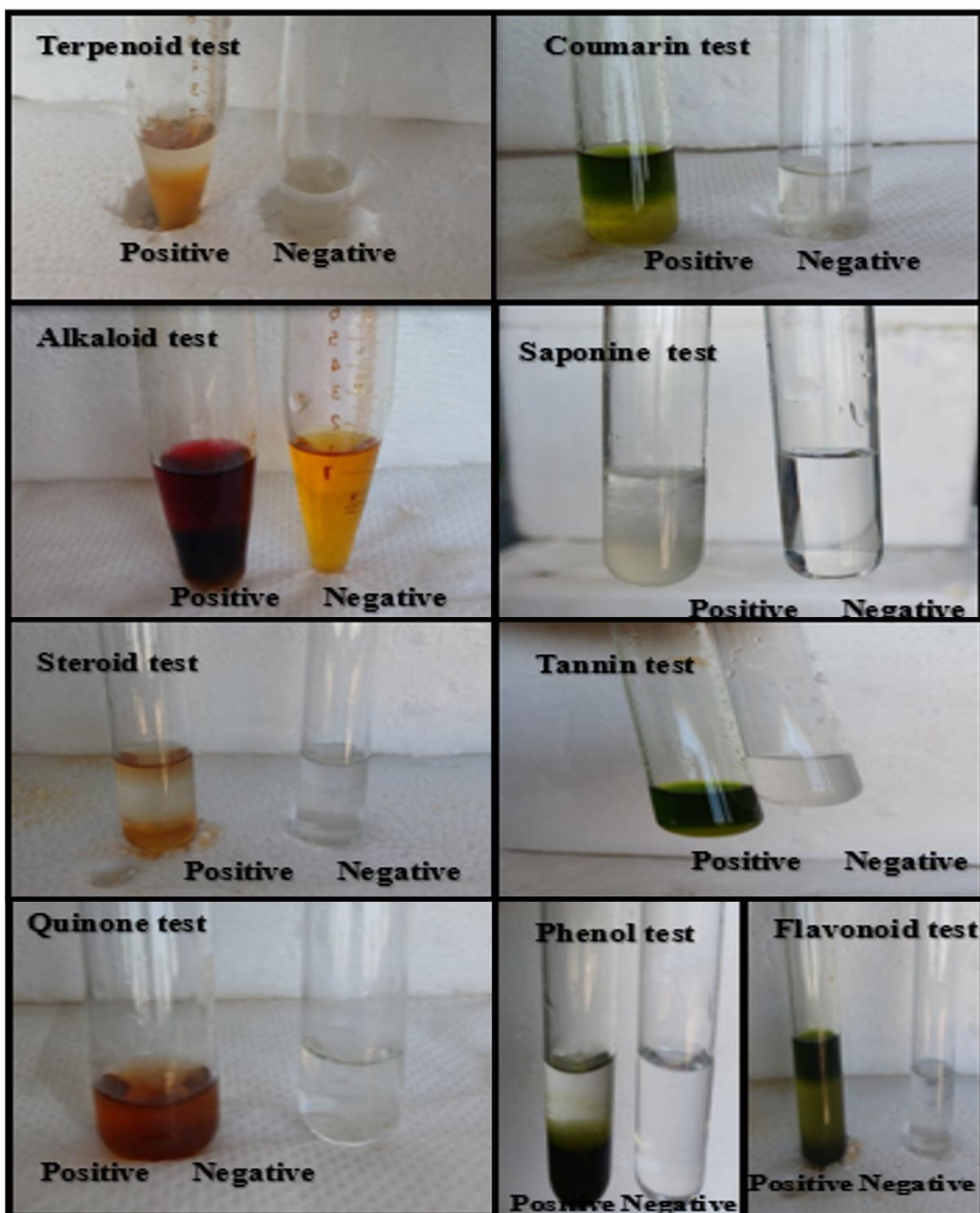


Figure 5: Phytochemical test for leaves and tuber roots of *E. condylocarpa*

extract. Formation of reddish-brown interface rings confirmed that steroids are present.

#### Detection of Saponins

To identify saponins, 2ml of plant extract and 2ml of distilled water were stirred in a measured cylinder for 15 minutes. Saponins were detected if 1-centimeter foam was formed.

#### Detection of Flavonoids

Flavonoids are represented by yellow coloration, by combining 2 ml of plant extract and 1 ml of 2N of sodium hydroxide.

#### Detection of Phenols

2 ml of distilled water was merged with 1ml of plant extract in addition to a few drops of 10% ferric chloride. Phenols are present if blue/ green color development took place.

#### Detection of Tannins

1 ml of 5% ferric chloride was added to 2 ml of the plant extract. Indicator of tannin seems in dark blue or greenish-black color.

#### Detection of Coumarins

1ml of 10% of sodium hydroxide was combined with 1 ml of plant extract to recognize the coumarins. The appearance of yellow color is a sign of coumarins.

#### Detection of Alkaloids

From 2 ml of plant extract, two drops of Wagner reagent were inserted and incorporate well. The aspect of a reddish color denotes the presence of alkaloids<sup>20</sup>.

#### Quantitative Analysis of Phytochemicals in Leaves and Tuber Roots of *E. condylocarpa*

##### Determination of total Condensed Tannin

This analyze was executed by UV- vis spectrophotometer. Firstly, a solution extraction was processed by mixing 0.05 g of FeSO<sub>4</sub>, 95 ml of N- butanol and 5ml hydrochloric acid (HCl) (35%). Then, for determine the condensed tannin, for 0.01g of both plant powder roots, leaves and mimosa tannin were 10ml of extraction solution were add independently and placed in a hot water bath 1h. Finally, the absorbance was deliberated at wavelength 580nm<sup>21</sup>.

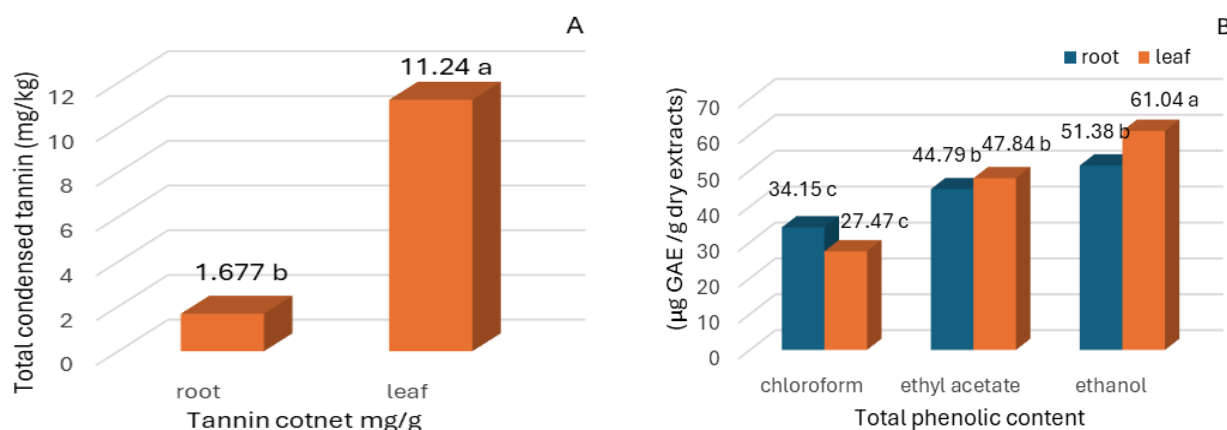


Figure 6: (A)Total condensed tannin (B) total phenolic content of leaves and tuber roots of *E. condylocarpa*

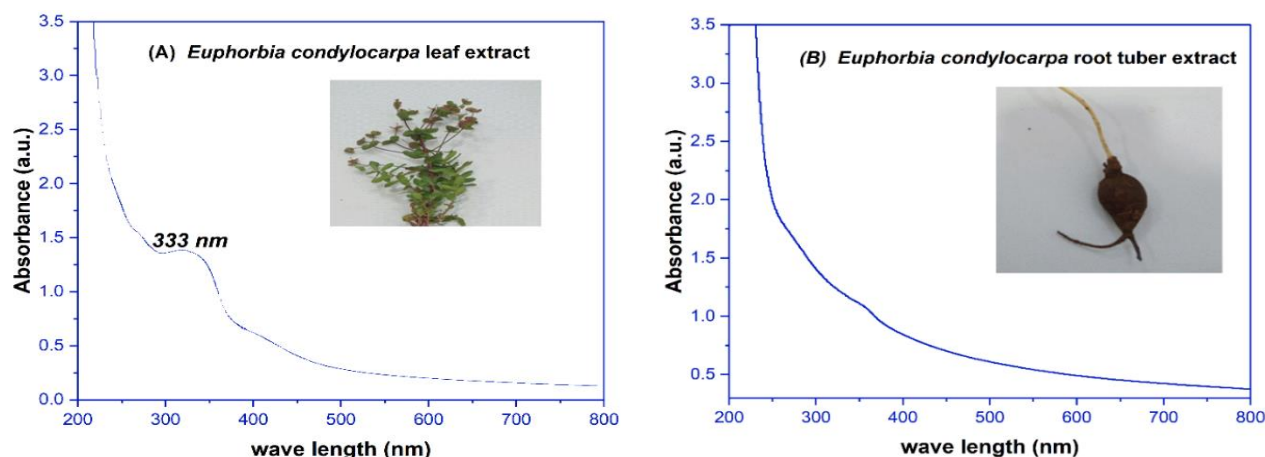


Figure 7: UV-vis spectrum of *Euphorbia condylocarpa* leaves and root tubers extract

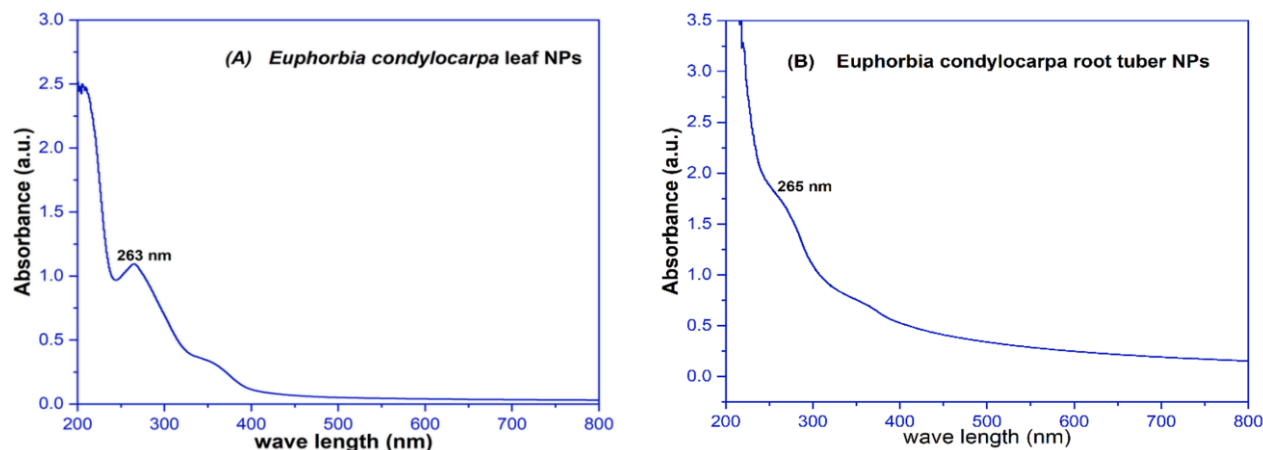


Figure 8: UV-vis spectrum of ZnO NPs synthesis using *Euphorbia condylocarpa* aqueous leaves and root tuber aqueous extract



### Assessment of Total Phenolic Compounds in Tuber Roots and Leaves of *E. condylocarpa* (TPC) (mg GAE /DW)

The total phenolic content was appraised by Folin-Ciocalteu reagents as methods described<sup>22</sup> by from the extract was prepared from spurge (leaves and root tubers). The properate amount 0.1 ml of filtered (chloroform, ethyl acetate, and ethanol) crude extracts (1mg/ml) were oxide by folin ciocalteu reagents and after 5 minutes was the reaction was relieved with saturated Sodium Carbonate (5%), furthermore the blend was authorized to stand for one hour in the shade with recurring vibrating. The absorbance was measured at 765nm. The phenolic contents of the extract were explicated in dry weight of the sample as mg Gallic acid equivalents per gram (mg.g).

### Statistical Analysis

The Statistical Package for Social Sciences (SPSS) (version 26) software was applied to data analysis, descriptive analyses and the means were equated using the Duncan's Multiple Range Test with a probability of 0.05, and for drawing graphs excel was used.

## RESULTS AND DISCUSSION

### The Yield and Percentage of Crude Extracts of *E. condylocarpa* M. Bieb

The quantity of crude extract of each of leaves and root tubers of *E. condylocarpa* by using different solvents were

Table 5: Determination of MIC of *E. condylocarpa* M. Bieb extract against three isolated bacteria

Plant parts	Extracts	MIC (mg/ml)		
		<i>Staphylococcus aureus</i> G <sup>+</sup>	<i>Pseudomonas aeruginosa</i> G <sup>-</sup>	<i>E. coli</i> G <sup>-</sup>
Leaves	Chloroform	41.66 a	10.41 c	25.00 abc
	Ethyl acetate	20.83 a	20.83 bc	25.00 abc
	Ethanol	37.50 a	18.75 bc	4.17 c
Tuber Root	Chloroform	20.83 a	18.75 bc	10.41 bc
	Ethyl acetate	41.66 a	20.83 bc	8.33 bc
	Ethanol	41.66 a	33.33 ab	16.66 bc
Leaf nanoparticles		29.16 a	41.66 a	41.66 a
Root tubers nanoparticles		25.00 a	33.33 ab	10.41 bc
Ciprofloxacin		29.16 a	14.58 bc	29.16 ab

shown in (Table 1). The highest value was recorded in ethanol, then in chloroform, the lowest value in ethyl acetate extracts.

The order of yield extract were ethanol>chloroform>ethyl acetate. The percentage of leaves and root tubers extracts varies from 4.7% to 9.2 % (Table 2)

### Qualitative Phytochemicals Determination in Leaves and Tuber Roots of *E. condylocarpa* M. Bieb

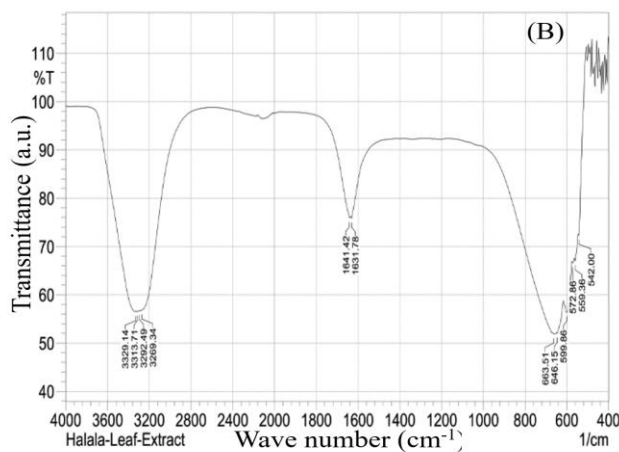
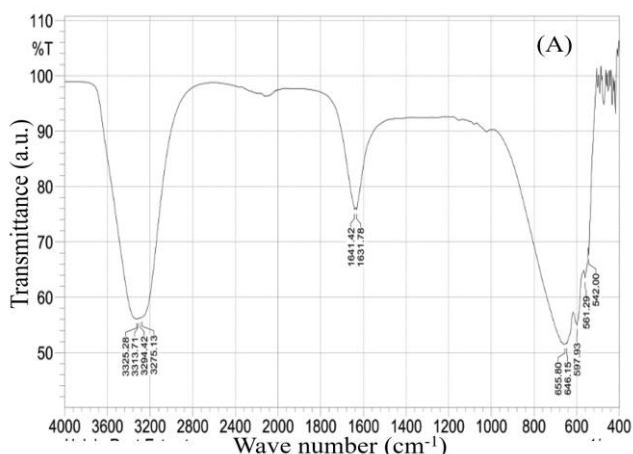


Figure 9: FTIR of *E. condylocarpa* from (A) Root tubers extract, (B) Leaves extract

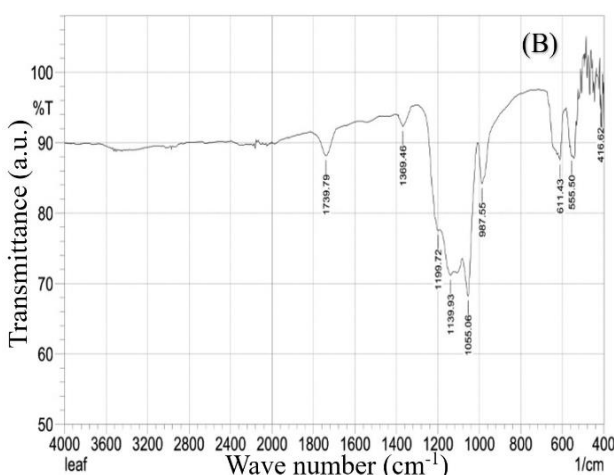
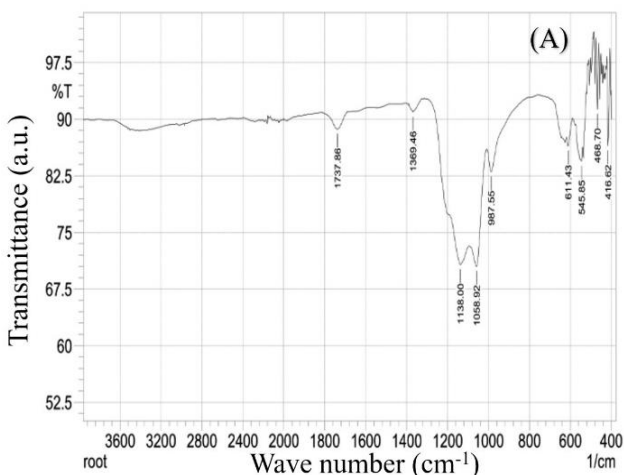


Figure 10: FTIR of biosynthesis of ZnO nanoparticles of (A) root tubers, (B) leaves of *E. condylocarpa*

Qualitative phytochemicals were analyzed by using different solvents chloroform, ethyl acetate, and ethanol of leaves and root tubers of *E. condylocarpa*. The data revealed that the contiguity of major phytochemicals groups such as terpenoids, alkaloids, phenols, and the results assigned as (+) for the occurrence together with (-) for lack of phytochemicals (Fig.5). The outcomes exhibited that the presence of different phytochemicals in leaves and tuber roots of *E. condylocarpa* (Table3). It's attainable that these phytochemicals liable for of therapic tracts. Terpenoids display myriad pharmacological activity operating as quick agent against inflammation, cancer, viruses in addition to bacteria. Flavonoids are possessed to have antioxidant outcomes impeding as well as succession of tumors. Tannins posses antibacterial, antitumor, besides antiviral schemes. Alkaloids are strong antibacterial performs<sup>23</sup>.

#### Quantitative Phytochemical Analysis of Different Extarcts of Leaves and Tuber Roots of *E. condylocarpa*

Figure (6) showed that the amount of total condensed tannin in leaves are more than in the root tubers which are (11.24 and 1.67 mg/kg respectively to leaves and roots), total condensed tannin found in vegetative tissues and roots of most of the plant which has a strong antioxidant and

biological activity<sup>24</sup>. While toal phenolic content of leaves and tuber roots by different solvents have a significant amount, showed that in leaves higher in ethanol then in ethyl acetate then in chloroform, and in root tubers more in ethanol then in ethyl acetate and then in chloroform. Phenolic combination are known for their the aromatic ring of the phenolic components with hydroxyl groups, high phenolic celements linked to high phenolic diet to the adverting of cardiovascular, cancer, and infections outstanding to the biologicalan besides pharmacological activites of phenolic compounds<sup>25</sup>.

#### Characterization of Nanoparticles and Plant Extract

There are several analytical techniques like Visible spectroscopy, Fourier transform infrared (FT-IR) spectroscopy, scanning electron microscopy (SEM), and X-ray diffraction (XRD), and Energy dispersive (X-ray) (EDX), which is utilized to describe structure, physical criterion and optical properties of ZnO nanoparticles and plant extract.

#### UV-visible Analysis

UV-vis technique is coeval qualitative and quantitative analysis. It is an initial, fleet and substantial step for causing the proficiency of plant to equip the NPs, and confirmed the

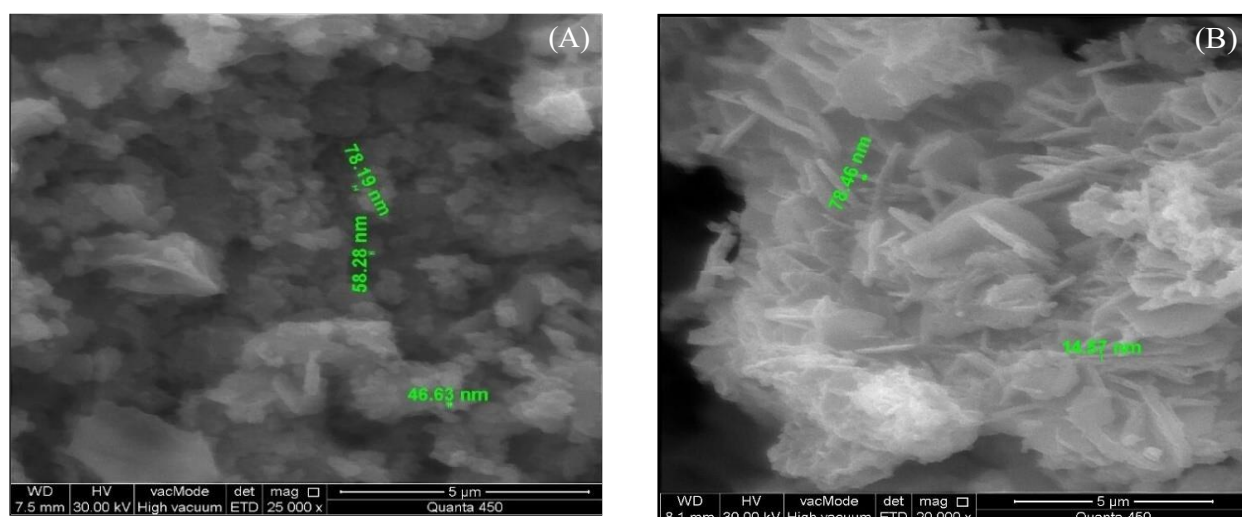


Figure 11: SEM descriptions of ZnO NPs synthesis from (A) leaves, (B) root tubers of *Euphorbia condylocarpa*

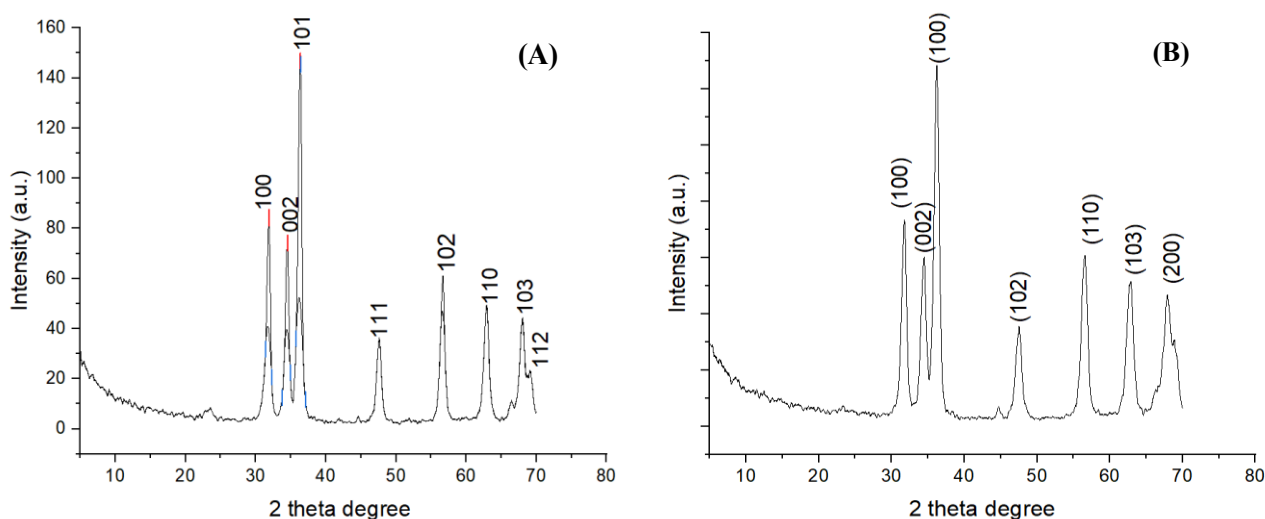


Figure 12: XRD analysis of ZnO NPs from (A) leaves, (B) tuber roots of *E. condylocarpa*



formation of ZnO NPs. The absorbance of the samples were measured using UV-vis spectroscopy across a 200–400 nm provision guidance about the optic possessions of the achieved the NPs<sup>26</sup>. The absorption peak of leaf and root tuber extract of *E. condylocarpa* was observed at 255nm, and 280nm (Figure 7). The absorption peak of leaf and tuber root NPs obeyed at the wavelengths with grade to nearly 285 nm and 265nm within the UV-vis range of 200–800 nm (Figure 8). Similar results have been reported by<sup>27</sup>.

#### Fourier- Transformed Infrared Spectroscopy (FT-IR)

FT-IR Spectro of the extract of plant and ZnO NPs were investigate the pureness, composition, and functional group of biosynthesis of ZnO NPs. The FT-IR spectra of leaves and root tubers of *Euphorbia condylocarpa* offering electrons for the ions that available from zero valent atoms (Figure 9), after that these atoms combined together to form NPs (Figure 10). The location point group 1055, 1058cm<sup>-1</sup> pertain to C-O stretching. In the plant extracts this peak represents the stabilizing agents<sup>28</sup>. The FT-IR analysis of the green synthesized using plant extract revealed the specific changes in related peaks, indicated the prescence of capping agent or bonded to nanoparticles surface<sup>29</sup>.

#### SEM Analysis

The morphological investigation of ZnO NPs which synthesis by green methods was examined though scanning electron microscopy technique (SEM). The SEM descriptions of ZnO NPs of both leaf and tuber root of *E. condylocarpa* are presented in (fig). It can be noticed that most of the ZnO NPs are in nanometer scale with the average diameter of 55 nm, 60nm for root and leaf nanoparticles.

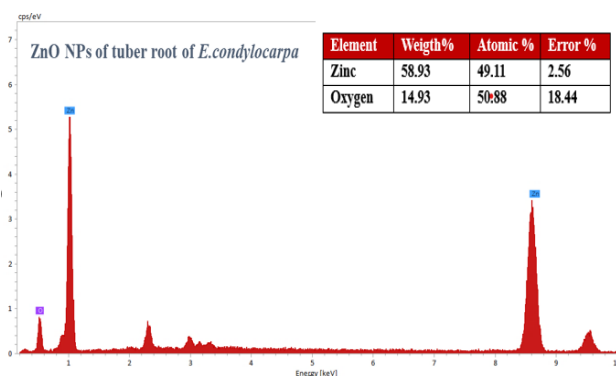


Figure 13: EDX analysis of ZnO NPs synthesis from root tubers of *Euphorbia condylocarpa*

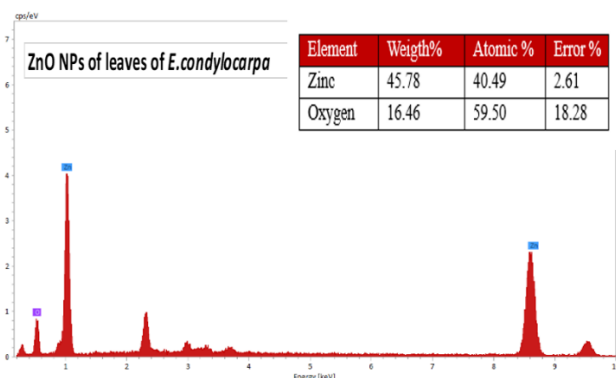


Figure 14: EDX analysis of ZnO NPs synthesis from leaves of *Euphorbia condylocarpa*

#### XRD Analysis

x-ray diffraction analysis of nanoparticles synthesis using plant extract is a new technique for the synthesis of nanoparticles. The XRD is done to analyses the structure and crystalline size of syntheses nanoparticles<sup>30</sup>. X-ray diffraction analysis used to approve the crystalline behavior of the ZnO NPs from *E. condylocarpa* leaf and tuber root extract. The diffraction peak locations 31.57, 34.78, 36.68, 44.71, 47.67, 56.77, 63.06, 67.84 corresponding for 100, 002, 101,111, 102, 110, 103,112 planes respectively for ZnO NPs for leaves. While for ZnO NPs for tuber root, the diffraction peak locations (31.23, 34.65, 36.76, 47.68, 55.98, 62.64, 67.31) corresponding to 100, 002, 101, 11, 102, 110, 103, 112 respectively. Hing diffraction peaks indicated the crystalline nature of the materials. The XRD pattern of ZnO NPs reveals sharp peaks that indicate the purity and crystallinity of green synthesis of nanoparticles<sup>31</sup>. The average of crystalline size nanoparticles purposive employing the Debye- Scherrer formula (Table4)

#### EDX Analysis

The basic construction of ZnO NPs was acquired from EDX analysis. (Fig. 13,14) flaunts the subsistence of chemical elements and compilation in the primed of ZnO NPs from leaves and tuber roots of *E. condylocarpa*. The presence of a huge portion of Zn and O is indicated of ZnO configuration. As slated the atomic prospect of Zn and O are almost equal (ZnO in 1:1 ratio). The high proportion of zinc present.

#### Minimum Inhibition Concentration (MIC) of Plant Extracts and Nanoparticles

MIC value of the extract and nanoparticles to restrain the growth of each of bacteria was resolute by using micro broth dilution methods. The results obtained were recorded in (Table5). The highest antimicrobial activities of leaves plant extracts by different solvents (chloroform, ethyl acetate, and ethanol) against *S. aureus*, *P. aeruginosa*, and *E. coli*, was significantly occurred in ethanol extracts against *E coli* with the MIC values 4.17 mg/ml., while this mentioned extract has no significant effect on *Staphylococcus aureus* and *Pseudomonas aeruginosa* bacteria. When all data compared to ciprofloxacin it shows close of better than antimicrobial reference(antibiotic).

The means in each category of column followed by the same letters are not significantly different at  $p < 0.05$  according to Duncan's Multiple Range test.

#### CONCLUSION

Plants have unique origin of bioactive substance with biological activities and medicinal properties. The use of various solvents to be crucial in the in extraction of bioactive compounds. In vitro antibacterial activity of various solvent extracts of chloroform, ethyl acetate, as well as ethanol of leaves beside root tuber extracts of *E. condylocarpa* and green synthesis nanoparticles have more preventive reaction against tested bacteria. Phytochemical analysis of above extracts showed appear of terpenoids, alkaloids, flavonoids, saponin, also tannin. Further studies on medicinal plants as a source of alternative medicine are necessary because of their bioactive qualities and generally

safe solvents. For incoming projects, the employed along with therapeutic techniques for extraction and desiccation.

### Acknowledgements

We extend our authentic gratitude for university of Salahaddin, college of science for their immeasurable advocate expediting consummate of this research.

### REFERENCES

1. Parvin S, Reza A, Das S, Miah MMU, Karim S. Potential Role and International Trade of Medicinal and Aromatic Plants in the World. *European Journal of Agriculture and Food Sciences*. 2023;5(5):89-99. <https://doi.org/10.24018/ejfood.2023.5.5.701>
2. dev Sharma A, Kaur I, Chuhan A, Kumar S, Singh N. Phytochemical profile, in-vitro antioxidant, anti-diabetic and anti-inflammatory activities of traditionally used *Euphorbia hirta* (L.) growing under wild conditions of Northern Punjab. *Drug Analytical Research*. 2023;7(1):27-40. <https://doi.org/10.22456/2527-2616.132488>
3. Nasrollahzadeh M, Sajadi SM, Rostami-Vartooni A, Khalaj M. Journey on greener pathways: use of *Euphorbia condylocarpa* M. bieb as reductant and stabilizer for green synthesis of Au/Pd bimetallic nanoparticles as reusable catalysts in the Suzuki and Heck coupling reactions in water. *RSC Advances*. 2014;4(82):43477-84. <https://doi.org/10.1039/C4RA07173E>
4. Ghosh S, Bishal A, Ghosh SK, Jana K, Gayen B, Sahu S, et al. Herbal medicines: A potent approach to human diseases, their chief compounds, formulations, present status, and future aspects. *Int J*. 2023;10:442-64. <https://doi.org/10.15379/ijmst.v10i1.2608>
5. Mohammadi A, Arabhosseini A. Antioxidant and Anti-Microbial Properties of *Euphorbia condylocarpa* Roots under Hot-air Drying Conditions. *J Med Plants By prod*. 2024;13(2):405-15. <https://doi.org/10.22034/jmpb.2023.360155.1502>
6. Pahlevani AH, Riina R. Synopsis of *Euphorbia* subgen. *Esula* sect. *Helioscopia* (Euphorbiaceae) in Iran with the description of *Euphorbia mazandaranica* sp. nov. *Nord J Bot*. 2014;32(3):257-78. <https://doi.org/10.1111/njb.01690>
7. C. C. Townsend, Guest E. *Flora of Iraq*. Baghdad 1980. 547 p.
8. Mohammadi S, Asgary V, Shandiz SAS, Heidari E, Jozaghkar H, Cohan RA, et al. Antimicrobial Activity of Methanolic Root Extracts of *Euphorbia Condyllocarpa* against Pathogenic Bacteria. *Adv Studies Biol*. 2015;7(2):55-64. <http://dx.doi.org/10.12988/asb.2015.41049>
9. Ioannou P, Baliou S, Samonis G. Nanotechnology in the Diagnosis and Treatment of Antibiotic-Resistant Infections. *Antibiotics*. 2024;13(2):1-27. <https://doi.org/10.3390/antibiotics13020121>
10. Rodrigues AS, Batista JG, Rodrigues MA, Thihe VC, Minarini LA, Lopes PS, et al. Advances in silver nanoparticles: a comprehensive review on their potential as antimicrobial agents and their mechanisms of action elucidated by proteomics. *Frontiers in Microbiology*. 2024;15:1440065. <https://doi.org/10.3389/fmicb.2024.1440065>
11. Amin ZS, Afzal M, Ahmad J, Ahmed N, Zeshan B, Hashim NHHN, et al. Synthesis, characterization and biological activities of zinc oxide nanoparticles derived from secondary metabolites of *Lentinula edodes*. *Molecules*. 2023;28(8):3532. <https://doi.org/10.3390/molecules28083532>
12. Yang X, Cao X, Chen C, Liao L, Yuan S, Huang S. Green Synthesis of Zinc Oxide Nanoparticles Using Aqueous Extracts of *Hibiscus cannabinus* L.: Wastewater Purification and Antibacterial Activity. *Separations*. 2023;10(9):466. <https://doi.org/10.3390/separations10090466>
13. Farooq A, Khan UA, Ali H, Sathish M, Naqvi SAH, Iqbal S, et al. Green chemistry based synthesis of zinc oxide nanoparticles using plant derivatives of *Calotropis gigantea* (giant milkweed) and its biological applications against various bacterial and fungal pathogens. *Microorganisms*. 2022;10(11):2195. <https://doi.org/10.3390/microorganisms10112195>
14. Ahmad NM, Mohamed AH, Zainal-Abidin N, Nawahwi MZ, Azzeme AM. Effect of optimisation variable and the role of plant extract in the synthesis of nanoparticles using plant-mediated synthesis approaches. *Inorganic Chemistry Communications*. 2024;161:111839. <http://dx.doi.org/10.1016/j.inoche.2023.111839>
15. Chatepa LEC, Mwamatope B, Chikowe I, Masamba KG. Effects of solvent extraction on the phytoconstituents and in vitro antioxidant activity properties of leaf extracts of the two selected medicinal plants from Malawi. *BMC Complementary Medicine and Therapies*. 2024;24(1):317. <https://doi.org/10.1186/s12906-024-04619-7>
16. Sundrarajan M, Ambika S, Bharathi K. Plant-extract mediated synthesis of ZnO nanoparticles using *Pongamia pinnata* and their activity against pathogenic bacteria. *Advanced powder technology*. 2015;26(5):1294-9. <https://doi.org/10.1016/j.appt.2015.07.001>
17. Ayoub S, Karim, Halala R, Qadr, Sakar A, Saeed, Khudhur NS. Determination of the Effects of Biogenic Metal Nanoparticles Concentrations on Soil Properties, *Vicia faba* Growth and the Toxicity on Black Aphid Karbala International Journal of Modern Science (KIJOMS) 2025;11:282-92. <https://doi.org/10.33640/2405-609X.3402>
18. Rahman JK, Jaff DMA, Basar N, Kuthi NA, Yaqoubi R. Phytochemical Screening, Isolation of Coumarins and Examining Bioactivity of *Prangos Platychlaena* Boiss. Plant in Iraq. *Applied Ecology and Environmental Research*. 11(2):2425-43. [https://doi.org/10.15666/AEER/1802\\_24252443](https://doi.org/10.15666/AEER/1802_24252443)

19. Savithramma N, MLR, KR, devi aPS. Antimicrobial activity of Silver Nanoparticles synthesized by using Medicinal Plants. international jurnal of Chem-Tech Research 2011;3(3):1394-402
20. Dahanayake JM, Perera PK, Galappatty P, Perera HDSM, Arawwawala LDAM. Comparative phytochemical analysis and antioxidant activities of Tamalakyadi decoction with its modified dosage forms. Evidence-Based Complementary and Alternative Medicine. 2019;2019(1):6037137.<https://doi.org/10.1155/2019/6037137>
21. Song W, Qin S-T, Fang F-X, Gao Z-J, Liang D-D, Liu L-L, et al. Isolation and purification of condensed tannin from the leaves and branches of *Prunus cerasifera* and its structure and bioactivities. Applied biochemistry and biotechnology. 2018;185(2):464-75.<https://doi.org/10.1007/s12010-017-2635-9>
22. Engeda Dessalegn MM, Hiwot Gebremeskel, Nigatu Tuasha. Determination of total phenolic and flavonoid contents, antioxidant and antibacterial potential of the bark extracts of *Syzygium guineense* (Wild.) DC. Dessalegn et al BMC Complementary Medicine and Therapies. 2025;35(25):1-10.<https://doi.org/10.1186/s12906-025-04788-z>
23. Adil M, Filimban FZ, Ambrin, Quddoos A, Sher AA, Naseer M. Phytochemical screening, HPLC analysis, antimicrobial and antioxidant effect of *Euphorbia parviflora* L.(Euphorbiaceae Juss.). Scientific Reports. 2024;14(1):5627.<https://doi.org/10.1038/s41598-024-55905-w>
24. Westley R, Ma D, Hawkins BJ, Constabel CP. Tissue and cellular localization of condensed tannins in poplar roots and potential association with nitrogen uptake. Frontiers in Plant Science. 2024;15:1388549.<https://doi.org/10.3389/fpls.2024.1388549>
25. Belew AA, Hanan GGMW, Meshesha DS, Akele ML. Evaluation of total phenolic, flavonoid contents, antioxidant and antibacterial activity of leaf extracts from *Rhus vulgaris*. Discover Plants. 2025;2(1):1-15.<https://doi.org/10.1007/s44372-025-00222-3>
26. Bekele SG, Ganta DD, Endashaw M. Green synthesis and characterization of zinc oxide nanoparticles using *Monoon longifolium* leave extract for biological applications. Discover Chemistry. 2024;1(1):5.<https://doi.org/10.1007/s44371-024-00007-9>
27. Dönmez S. Green synthesis and characterization of zinc oxide nanoparticles by using RHODODENDRON ponticum L. leaf extract. Turkish Journal of Health Science and Life. 2021;4(1):54-7
28. Azeez A. Barzinjy BSH. Green synthesis and characterization of Ag nanoparticles using fresh and dry *Portulaca Oleracea* leaf extracts: Enhancing light reflectivity properties of ITO glass. micro nano letters 2024;1-13.<https://ietresearch.onlinelibrary.wiley.com/doi/10.1049/mna2.12198>
29. Albarakaty FM, Alzaban MI, Alharbi NK, Bagrwan FS, Abd El-Aziz AR, Mahmoud MA. Zinc oxide nanoparticles, biosynthesis, characterization and their potent photocatalytic degradation, and antioxidant activities. Journal of King Saud University-Science. 2023;35(1):102434
30. Kumar M, Ranjan R, Dandapat S, Srivastava R, Sinha MP. XRD analysis for characterization of green nanoparticles: a mini review. Global Journal of Pharmacy and Pharmaceutical Sciences. 2022;10(1):555779.<http://dx.doi.org/10.19080/GJPPS.2022.10.555779>
31. Mardani-Talaei M, Razmjou J, Ajdari A, Serrão JE, Vivekanandhan P. Green synthesis of zinc oxide nanoparticles from *Sargassum ilicifolium* to enhance tomato resistance against *Tuta absoluta*. Scientific Reports. 2025;15(1):13596.<https://doi.org/10.1038/s41598-025-97535-w>