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Synthesis, characterization, antimicrobial and electrochemical activities of zinc oxide nanoparticles obtained from sarcopoterium spinosum (L.) spach leaf extract

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Abstract

In this study, we introduce a green and simple method for the synthesis of ZnONPs utilizing *Sarcopoterium spinosum* (L.) Spach leaf extract as a reducing agent for the first time. Zinc oxide nanoparticles (ZnONPs) were synthesized with a simple, non-toxic, non-expensive and eco-friendly method. In this method, the leaf extract of *S. spinosum* was used as a reducing agent. Moreover, the effect of pH and calcination on the size and shape of ZnONPs were also investigated. The particle size, morphology and the electrochemical activity of the synthesized nanoparticles were characterized by using the scanning electron microscope, the UV/VIS spectroscopy, the X-ray powder diffraction, the Zeta sizer, the energy-dispersive X-ray spectroscopy and Cyclic voltammetry (CV) techniques. The synthesized ZnONPs were evaluated due to antibacterial activity against Gram-positive and Gram-negative bacteria strains and yeast. The obtained results showed that the synthesized ZnONPs show different antibacterial activity on Gram-positive and Gram-negative bacteria strains and antifungal activity against fungal strains. The reason for these different antibacterial and antifungal effects of the obtained nanoparticles is that they have various morphologies and particle sizes. Further, the cost-effective ZnONPs, obtained via biosynthesis, showed a good electroactive behavior and thus, they can be suggested as possible nominees for electrochemical applications.

1. Introduction

Nanotechnology is the creation and manipulation of materials on an atomic or molecular level, and the use of these materials at the nano level for various purposes [1]. Nanotechnology gets great attention as an important research area with its enormous applications in science, engineering, medicine and pharmacy. Nanoparticles evoke materials with a size ranging from 1 to 100 nm. The nanostructured materials are generally synthesized by physical and chemical reduction methods. These conventional methods are costly and need extra stabilizing agents because of unwanted agglomerations of nanoparticles.

In recent years, nanoparticles synthesized via biomolecules, including DNA, protein, enzyme and plant extract have a great attention. Among the biomolecules mentioned, plant extracts have more attention in nanoparticle synthesis because they offer the following advantages: (i) plant extracts are very cheap, stable, and provide large scale production, (ii) they have a low risk of contamination and (iii) they can be easily prepared [2, 3].

Zinc oxide is an important electronic, photonic and optic material because of its wide direct band range of 3.37 eV at room temperature. Syntheses of ZnONPs have been achieved using various techniques including the hydrothermal synthesis [4–6], the sol-gel method [7, 8], the sonochemical method [9, 10], laser ablation [11, 12]

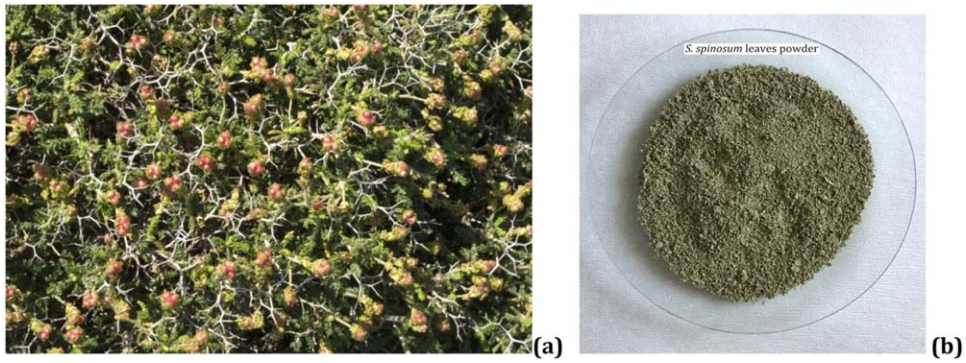

Figure 1. Picture of (a) *S. spinosum* plant and (b) leaves powder.

and the electrochemical method [13]. The research results in literature showed that the ZnO nanoparticle has an antimicrobial agent against Gram-positive and Gram-negative bacterial strains [14]. ZnONPs have also various applications in semiconductors, piezoelectric devices, solar cells, gas sensors and cosmetic materials.

S. spinosum belongs to the family Rosaceae, a characteristic species of the Mediterranean region [15, 16]. *S. spinosum,* also known as Thourny burnet, has been used in traditional and herbal medicine in the Middle East to treat digestive problems, diabetes and cancer [17]. It is known that *S. spinosum* roots have been used for the treatment of diabetes by people in the Middle East (Beddouin or Arab) for a long time.

Nanoparticles can be synthesized by physical, chemical and biological methods. The main methods for nanoparticle productions are chemical and physical approaches that are often costly and potentially harmful to the environment. The biological approach has been actively pursued in recent years as an alternative, efficient, inexpensive, and environmentally safe method for producing nanoparticles with specified properties. Plant extracts are good stabilizers and reducing agents for nanoparticle syntheses. The use of biological organisms such as bacteria, fungi and plant extracts could be an alternative to chemical and physical methods for the production of nanoparticles in an ecofriendly manner [18–21]. During the past years, it has been demonstrated that many biological organisms, including plants [22], fungi, such as yeast and molds [23, 24] and bacteria [25] can transform metal ions into metal nanoparticle formations.

According to literature review, we could not find any synthesis, characterization or application of the *S. spinosum* leaf extract as reducing agent. In this article, we obtained a green and simple method for the synthesis of ZnONPs utilizing *S. spinosum* leaf extract. The structure, morphology and electrochemical activity of the synthesized ZnONPs were investigated by standard characterization techniques. As well, mechanisms of the formation of the ZnONPs by means of plant materials were introduced. Moreover, the antimicrobial activity of the nanostructured materials was evaluated by using the modification microdilution broth method.

2. Experimental

2.1. Materials

Zinc acetate dihydrate, ethylene glycol, sodium hydroxide, hydrogen chloride were purchased from Merck. *S. spinosum* leaves were collected at Mersin University, Ciftlikkoy Campus (Collection stations: Mersin University, Ciftlikkoy Campus, maquis degraded areas, 100–130 m) in June 2016. The voucher specimens are deposited in the Biology Herbarium, at Mersin University, Mersin, Turkey.

2.2. Preparation of plant extract

S. spinosum leaves were washed three times with double distilled water to remove dust particles and then dried for one week. The dried leaves were powdered in a home mixer grinder (figure 1). The extract used for the reduction of Zn^{2+} ions to zinc oxide nanoparticles was prepared by putting 5 g dried powdered leaves in a flask with 150 mL distilled water. The mixture was then heated to 60 °C for 120 min, using a magnetic stirrer. The extract was cooled to room temperature and was centrifuged at 14 000 rpm for 20 min. The extract was stored in a refrigerator in order to be used in further experiments.

2.3. Green synthesis of ZnONPs

For the synthesis of nanoparticles, three separate sets of 50 mL of *S. spinosum* leaf extract was taken and heated to 60 °C, using a stirrer-heater. The initial pH of the leaf extract was around 5.8. 0.1 M HCl or 0.1 M NaOH was added to each solution until the pH values of 4, 7 and 10 were obtained, respectively. Zinc acetate dihydrate

(5.5 g) was dissolved with 50 mL distilled water. The prepared zinc acetate dihydrate solution was then added dropwise to the *S. spinosum* leaf extract solutions at a temperature of 60 °C. In each case, the reaction mixture was heated to 60 °C under continuous stirring for two hours and then left for cooling to room temperature. A pale-brown precipitate was obtained through centrifugation and washed with double-distilled water. The ZnONPs were then collected in a ceramic crucible and heated in an air heated furnace at 100, 200, 300, 400 and 500 °C for 150 min. The color of ZnONPs were dark for 100, 200 and 300 °C and light-yellow powder for 400 and 500 °C, respectively. The obtained products were powdered and packed for characterization purposes.

2.4. Characterization techniques and instrumentations

The biosynthesized ZnONPs were characterized by using the UV–vis spectrophotometer (Shimadzu 1800 spectrophotometer), the X-ray powder diffraction (XRD) (Rigaku diffractometer with CuKα, $\lambda = 1.5406$ Å) and the Cyclic voltammetry (CV) (CHI 660E electrochemical workstation). The glassy carbon electrode (ca. 3 mm) was used as a working electrode, a platin wire as a counter electrode and Ag/AgCl as a reference electrode. The morphology and size of the nanomaterials were examined by the scanning electron microscopy (SEM) (Zeiss), the energy dispersive x-ray analysis (EDX) and the dynamic light scattering (DLS) analysis.

2.5. Electrochemical properties

The electrochemical investigation of biosynthesized ZnONPs was carried out by the cyclic voltammetry. The electrochemical activity of the ZnONPs were investigated in a three-electrode system on the glassy carbon working electrode (GCE). The platinum wire and Ag/AgCl (1 M KCl) were used as a counter and reference electrode, respectively. The cyclic voltammetry measurements were performed at the potential range of +1.0 and −1.0 V and at the scan rate range of 25 and 125 mV s^{-1} in 0.1 M NaOH supporting electrolyte.

2.6. Preparation of GC/ZnO electrodes

Before modification, the surface of glassy carbon electrode (GCE) was polished using 1.00, 0.30 and 0.05 mm alumina slurry and then ultrasonicated in 1:1 (*v:v*) nitric acid, acetone and deionized water for 5 min, respectively. The polished glassy carbon electrode was modified by the drop casting method. Briefly, 5 mg of nanoparticle was dissolved in 1 mL deionized water, magnetically stirred for 24 h and then ultrasonicated for 30 min to get homogeneous paste. This paste (5 μL) was carefully cast on the freshly polished GCE. A small beaker was covered over the electrode and the solvent was evaporated at room temperature [26].

2.7. Antimicrobial activities

In vitro, the antimicrobial activities of the ZnONPs were tested against Gram-positive and Gram-negative bacteria, and fungi. The antimicrobial susceptibility test was evaluated using the modification microdilution broth method [27–29]. Six reference bacterial strains and two fungal strains were used: *Escherichia coli* (ATCC 25922), *Pseudomonas aeruginosa* (ATCC 27853), *Staphylococcus aureus* (ATCC 25925), *Streptococcus pneumoniae* (ATCC 10353), *Bacillus subtilis* (ATCC 6633), *Enterococcus faecalis* (ATCC 29212), *Candida glabrata* (ATCC 4322), and *Candida albicans* (ATCC 90028). The fungal and bacterial cell inoculum was prepared from the stock culture grown in Tryptic Soy Agar (TSA, Merck, Darmstadt, Germany) at 28 °C for 24 h and Mueller-Hinton Agar (MHA, Merck, Darmstadt, Germany) 37 °C for 24 h, respectively. The cell density adjusts to match the turbidity of a Mac Farland 0.5. Antibacterial and antifungal activity tests were performed in Mueller-Hinton broth (Merck, Darmstadt, Germany) and Triptic soy broth (Merck, Darmstadt, Germany), respectively. Dilutions of ZnONPs and standard drugs in the test medium were prepared serial dilutions; 500, 250, 125,… μg/mL concentration with Mueller-Hinton broth and Triptic soy broth. Then 5 μL of cell suspension was added to each tube, except the last one, which acted as a control well. Only 5 μL of fungal and bacterial suspension were added in another control tube without chemicals and used as a control for growing. All plates were incubated at 28 °C (for fungi) and at 37 °C (for bacteria) for 24 h. Minimum inhibitory concentration (MIC) values were recorded on the lowest concentrations of the compounds, which had no visible turbidity for bacteria and fungi. Ampicillin and fluconazole were used as reference drugs in antibacterial and antifungal activity tests, respectively. The results were obtained visually and by measuring optical density for 24 h.

3. Results and discussion

3.1. UV–vis spectroscopy

The green synthesis of ZnONPs were monitored by UV–vis spectrophotometer scanning in range of 800 to 300 nm wavelenght. With the adding of *S. spinosum* leaf extract to the Zn^{2+} aqueous solution, a color change from brown to dark brown was observed and this result confirmed the formation of ZnONPs. This was also verified with the observation of an absorption band at about 370 nm. The UV–vis spectra of ZnONPs were synthesized using *S. spinosum* leaf extracts with various calcination temperatures and pH media, given in

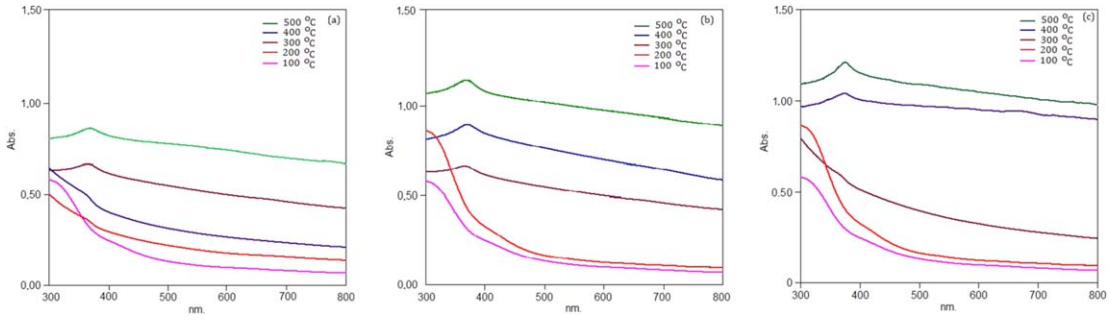

Figure 2. UV-Visible spectra of ZnONPs prepared with various calcination temperature and pH of *S. spinosum* leaf extract. (a) pH = 4, (b) pH = 7 and (c) pH = 10.

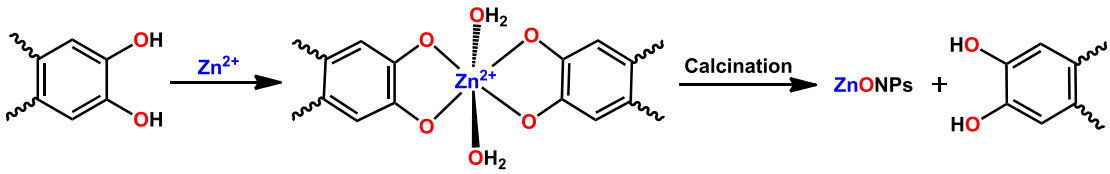

Figure 3. Possible mechanism of formation ZnONPs by using *S. spinosum* leaf extracts.

figure 2. The ZnONPs formation was not observed at calcination temperatures of 100 and 200 °C in all pH media. The spectra reveal an absorption peak at 367 nm after 300 °C that can be assigned as the formation of ZnONPs. As seen in figure 2, the formation of ZnONPs changes depending on pH. As the pH value increases from 4 to 10 the absorption peak intensity also increases. As a result, we observed that by the increase of the formation of $Zn(OH)_2$ precipitation in the basic medium, the rate of ZnONPs production increases [30]. Furthermore, the enhanced formation of ZnONPs in neutral and basic medium may be due to the ionization of phenolic groups present in the leaf extracts. The calcination temperature is another important parameter that plays a significant role in the bio-reduction of nanoparticle formation. In this study, it was found that the formation of ZnONPs increases with increasing temperature. When the UV–vis results are considered, the formation of nanoparticles was found to be best in the pH = 10 media and at temperatures of 300, 400 and 500 °C, respectively. Therefore, only the EDX, XRD and DLS characterizations of zinc oxide nanoparticle graphics in pH = 10 media and at 300, 400 and 500 °C were given in the following part of this study.

In recent years, various plant extracts were used extensively in the green synthesis of metal and metal oxide nanoparticles. Various plant constituents, including terpenoids, polyphenols, alkaloids, phenolic acids play an important role in the biosynthesis of nanoparticles. Flavonoids are polyphenolic compounds and contain various functional groups, capable of nanoparticle formation [31–34]. According to previous reports, catechin, epicatechin, hyperoside and isoquercetin were detected in *S. spinosum* extract [17, 34]. Checking literature, we found that these main components acted as a reducing and/or capping agents for the synthesis of the nanoparticles [27–29]. The formation mechanism of the green synthesis of nanoparticles has not been confirmed up to now. It is estimated that phenolic components, present in the plants, act as ligation agents which allow a complex formation between Zn^{2+} and polyphenolic groups [31–33]. A probable formation mechanism of ZnONPs via *S. spinosum* leaf extracts can be summarized as follows (figure 3) [35–37].

3.2. X-ray diffraction analysis

Temperature is an important factor affecting the formation of nanoparticles in plant extracts. The crystalline nature of the biosynthesized ZnONPs by *S. spinosum* leaf extract was carried out by the x-ray diffraction analysis. Figure 4 shows the XRD pattern samples at different temperatures. As seen in figure 4, there are no peaks in the XRD patterns of the sample which was treated at temperature 100 and 200 °C. This is because of the amorphous nature of the synthesized ZnONPs. As the temperature increased from 300 to 500 °C, the crystallinity of ZnONPs also increased. Depending on this, the intensity of the biosynthesized ZnONPs peak increases with increasing temperature [38]. Generally, temperature elevation increases the reaction rate and efficiency of nanoparticle synthesis. Furthermore, crystal particles are formed much more frequently at high temperatures. It is assumed that elevating the temperature increases the nucleation rate [18].

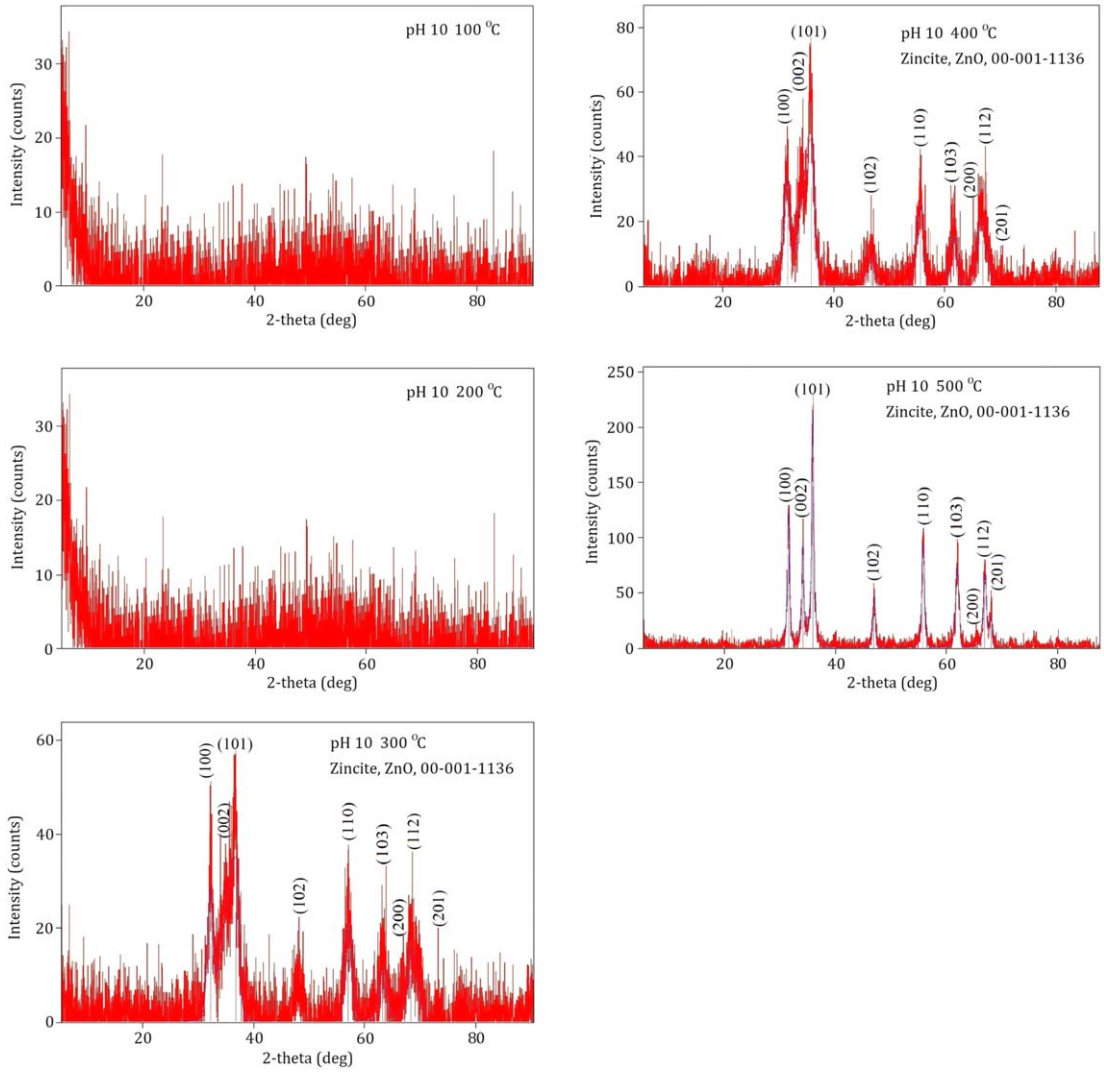

Figure 4. XRD pattern of ZnO prepared at different calcination temperature (pH = 10).

The XRD patterns of ZnONPs synthesized using *S. spinosum* leaf extracts at temperatures of 300, 400 and 500 °C match well with the ICDD card number of 00-001-1136. All the peaks of (100), (002), (101), (102), (110), (103), (200), (112) and (201) can be well indexed to the hexagonal Wurtzite structure. Strong intensity and narrow width of ZnO diffraction peaks indicate that the resulting product was crystalline in nature.

3.3. Scanning electron microscopy analysis (SEM)

Further analysis was carried out using SEM, EDX and the dynamic light scattering analysis to identify the shape, chemical composition and size of the prepared nanoparticles. It is known that the shape and size of the nanoparticles depends on parameters like temperature, pH media and etc [39]. SEM analysis showed that biosynthesized ZnONPs were spherical, triangle pyramide, rectengular, rod and plate in shape (figure 5). The size of the ZnONPs varies between 26.39–115.00 nm, depending on the shape of the nanoparticles. The rod-like nanoparticles were observed at 400 and 500 °C and in all examined pH media. The triangle pyramid, rectangular and plate shape nanoparticles occurred at 300 °C and in all pH media.

EDX analyses were carried out to confirm the presence of ZnONPs and to determine the elemental composition of the ZnONPs. EDX spectra verified the presence of zinc and oxygen elements in the synthesized nanostructure (figure 6).

The Dynamic Light Scattering (DLS) is widely used to measure the size distribution of the synthesized ZnONPs. According to the particle size distribution, all synthesized nanoparticle sizes range from 26.39 to 115.00 nm. The particle size of the synthesized ZnO varies depending on the temperature and the pH media. Results showed that the synthesized nanoparticle sizes decrease with pH and temperature. According to the particle size distribution, we observed the smallest nanoparticle size at pH value of 10 and temperature of 500 °C.

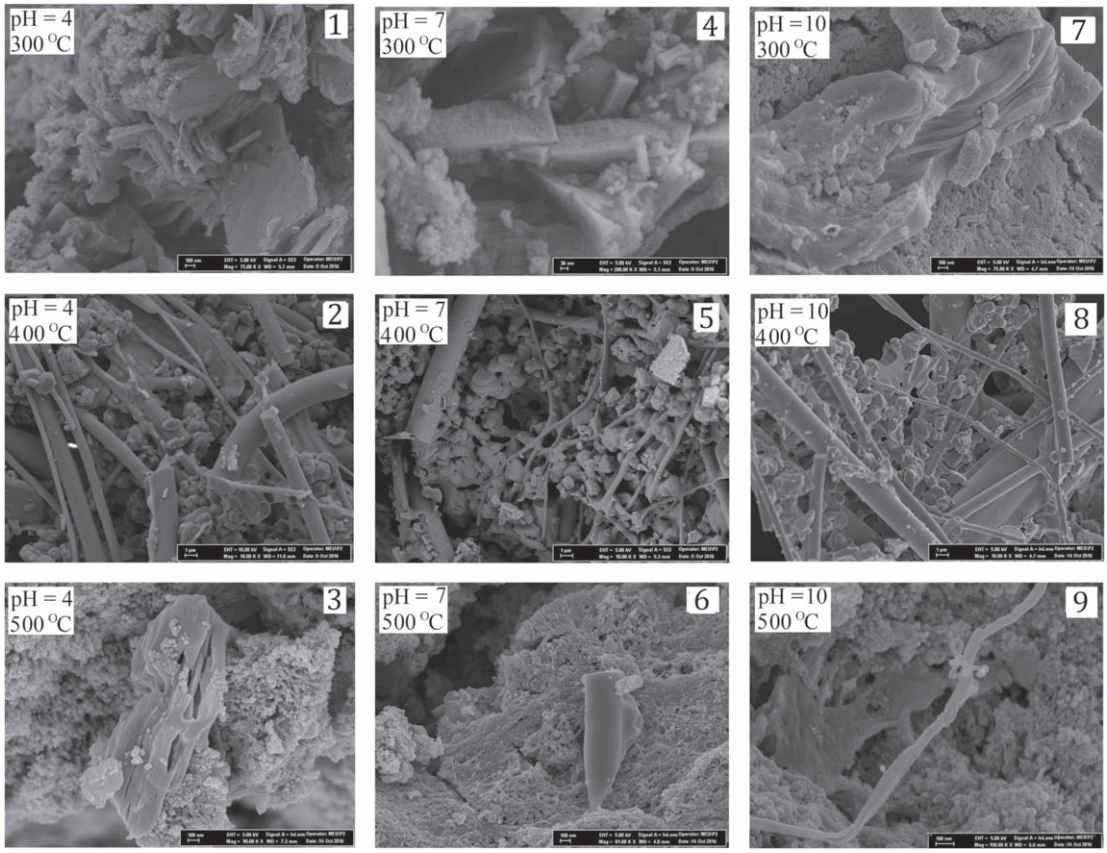

Figure 5. SEM microphotographs of ZnONPs synthesized using *S. spinosum* leaf extract at different calcination temperature and pH.

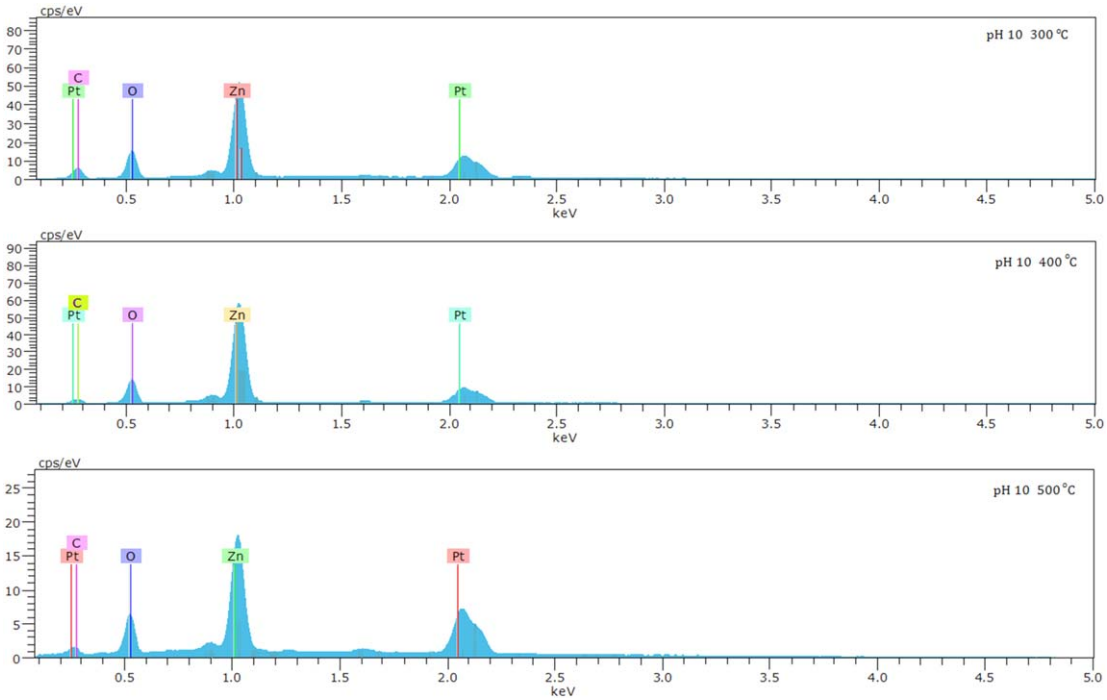

Figure 6. EDX profile of ZnONPs synthesized using *S. spinosum* leaf extract at different calcination temperature.

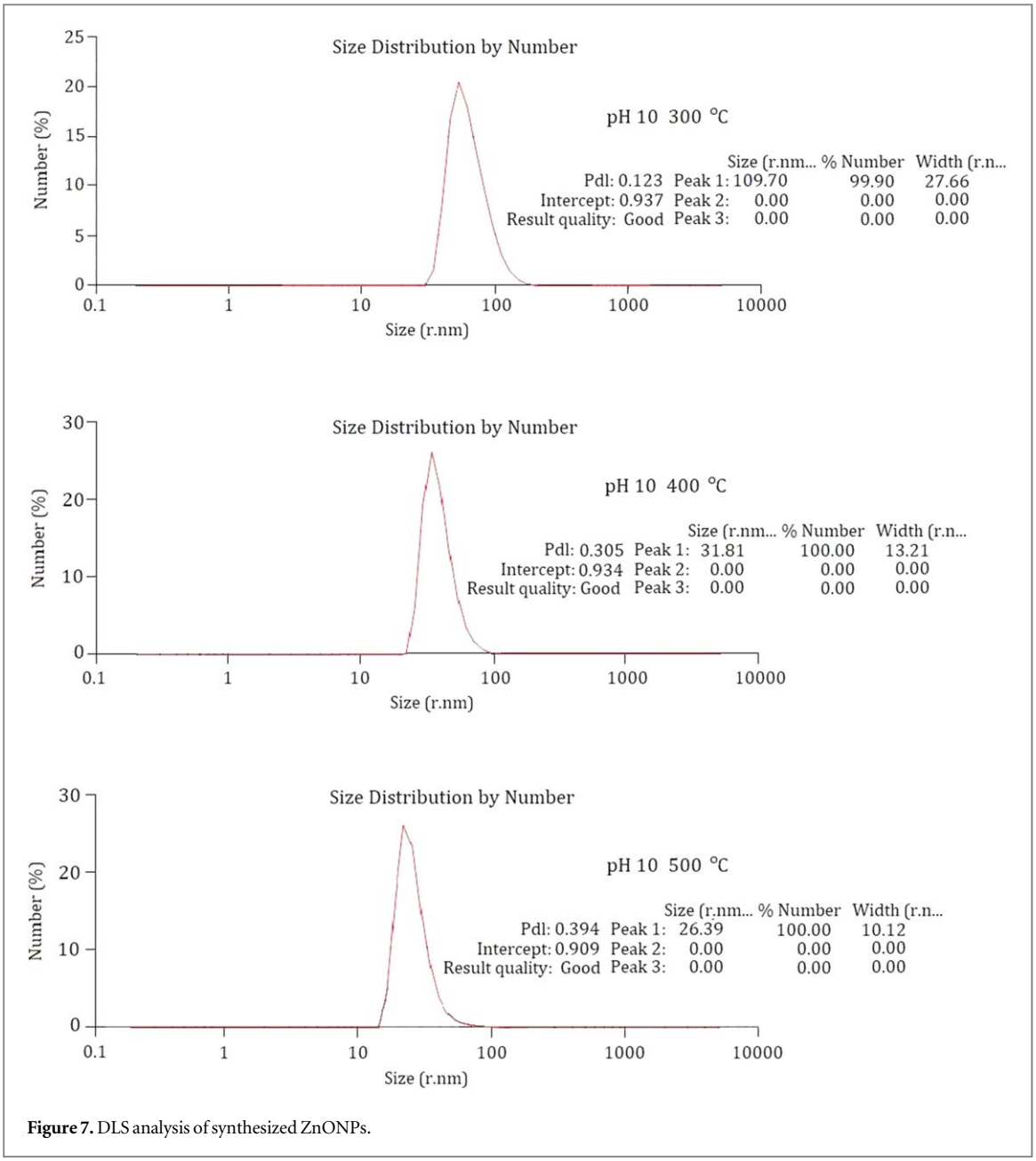

Figure 7. DLS analysis of synthesized ZnONPs.

Table 1. Particle size distribution of the synthesized ZnONPs (nm) at different pH media and calcination temperature.

	Temperature (°C)		
pH	300	400	500
4	115.00	78.21	58.77
7	113.40	63.47	38.73
10	109.70	31.81	26.39

On the other hand, the largest nanoparticle size was at pH value of 4 and temperature of 300 °C (table 1). Figure 7 shows the particle size distribution of ZnONPs that synthesized at pH = 10 media and 300, 400 and 500 °C.

3.4. Electrochemical properties of ZnONPs

The electrochemical properties of the synthesized ZnONPs were investigated by the cyclic voltammetry. The electrochemistry of the bare GCE and ZnONPs modified GCE in 0.1 M NaOH versus Ag/AgCl reference electrode are shown in figure 8. In the present study, three modified electrodes were constructed with ZnONPs,

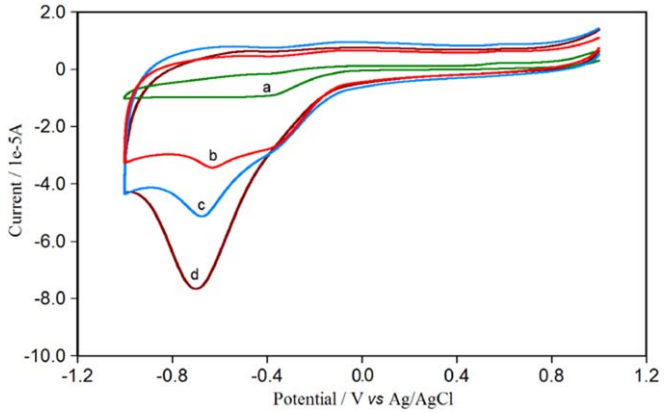

Figure 8. Cyclic voltammograms of (a) bare GCE, (b) ZnO_300/GCE, (c) ZnO_400/GCE and (d) ZnO_500/GCE in 0.1 M NaOH supporting electrolyte at scan rate of 100 mV s^{-1}.

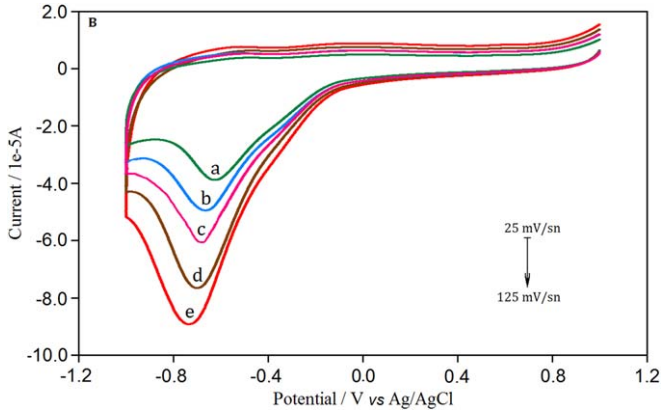

Figure 9. Cyclic voltammograms of ZnO_500/GCE in 0.1 M NaOH supporting electrolyte at variable scan rates, (a) 25, (b) 50, (c) 75, (d) 100 and (e) 125 mV s^{-1}.

synthesized at different temperatures of 300, 400 and 500 °C, specified as ZnO_300/GCE, ZnO_400/GCE and ZnO_500/GCE, respectively, at pH = 10 medium. Based on figure 8, it can be seen that there is no apparent peak on the unmodified electrode. When GCE modified with ZnONPs, a significant reduction current was observed at all modified electrodes and ZnO_500/GCE exhibited greater reduction current response compared to ZnO_300/GCE and ZnO_400/GCE. The apparent reduction peak in figures 8(b), (c) and (d), observed at about −650 mV, indicated the reduction of ZnO into metalic Zn. Based on observed results, synthesized ZnONPs showed good electrochemical behavior and can herewith be recommended as electrocatalysts for electrochemical application.

For the purpose of evaluating the electrochemical behavior of the ZnONPs, the scan rate effect was investigated. The electrochemical activity of the ZnO nanoparticle was assessed by monitoring the change in the reduction current during cathodic scan (figure 9). As seen in figure 9, as the scan rate increases the reduction peak current increases linearly, which indicates that the electron transfer is under diffusion control [26].

3.5. Antimicrobial activity investigation

The previous literature studies pointed out that there has been a great attention to the production of nanomaterials, which exhibit antimicrobial activity to remove the risk of infectious diseases [30]. It has been known for a long time that ZnO shows the best antimicrobial activity among metal oxide nanoparticles, therefore it is employed in antibacterial creams, ointments and lotions. The most interesting part is that ZnONPs affects insignificantly human cells [40].

Many studies have reported that toxicity is significantly affected by various morphologies and particle sizes of ZnONPs [41–46].

In the present study, antimicrobial activities of ZnONPs were tested for *in-vitro* antibacterial activities against Gram-positive and Gram-negative strains and antifungal activity using a modification microdilution

Table 2. MIC values (μg mL^{-1}) of the ZnONPs tested against the Gram-positive, Gram-negative bacteria and fungi [a].

pH	Temp. (°C)	*S. aureus*	*S. Pneumonia*	*B. subtilis*	*E. faecalis*	*E. coli*	*P. eruginosa*	*C. albicans*	*C. glabrata*
4	300	125	125	125	125	125	125	**15.625**	**62.5**
7	300	125	125	125	125	125	125	**31.25**	**62.5**
10	300	125	125	**31.25**	125	125	125	**15.625**	**31.25**
4	400	250	250	125	125	125	125	125	125
7	400	250	125	125	125	125	125	125	125
10	400	125	125	**62.5**	125	125	125	**62.5**	**62.5**
4	500	250	125	125	125	125	125	125	125
7	500	125	125	125	125	250	125	125	125
10	500	125	**62.5**	**62.5**	**62.5**	**62.5**	125	**31.25**	**31.25**
Fluconazole		—	—	—	—	—	—	*	*
Ampicillin		*	*	*	*	**31.25**	**31.25**	—	—

[a] '-' Non-effective; '*' Effective in all concentrations used.

method. According to antimicrobial studies, all synthesized nanoparticles exhibit antimicrobial activity Gram-negative and Gram-positive bacteria and their MIC values range from 15.62–250 μg mL^{-1}. Considering the antibacterial results, nanoparticles obtained at high pH media and at highest temperature were generally found to be more effective against Gram-negative and gram-positive bacteria (table 2). However, it was determined that the nanoparticles obtained at pH = 10 media and 300, 400 and 500 °C exhibited antibacterial activity only against *B. subtilis*. It was found that nanoparticles obtained at 300 °C and in all pH media were effective against fungus. In addition, the nanoparticles obtained at pH = 10 media and 400–500 °C exhibited also high fungal effect. The reason for these different antibacterial effects of the obtained nanoparticles is that they have various morphologies and particle sizes. The estimated values of MIC's against different bacteria strains and fungal strains mentioned in table 2.

The antimicrobial activities of the synthesized nanoparticles vary depending on the formation of reactive oxygen species (ROS), particle size and concentration, the nanoparticle surface charge, the surface area and the dissolution of the nanoparticles. Synthesized nanoparticles showed different growth inhibition on examined bacterial and yeast species. Synthesized nanoparticles were generally more effective against *C. albicans* and *C. glabrata* species. Depending on the strains (species), the alkalinity of the medium is thought to increase the dissolution of the nanoparticles and hence the toxicity. It is anticipated that the absence of antibacterial effects ZnONPs against some Gram-positive and Gram-negative bacterial strains, which may result from low concentration. In this study, ZnONPs were obtained using *S. spinosum* leaf extract and antimicrobial effects were determined for the first time. Since there is no direct study on *S. spinosum* species, ZnONPs and their antimicrobial effects have been compared with previous studies, which used the leaf extracts of different species.

ZnONPs were synthesized using *Calotropis procera* (Aiton) W.T. Aiton extract and its antimicrobial effect on Gram-positive and Gram-negative bacteria was investigated by Poovizhi *et al* [47]. Poovizhi *et al* reported that the MIC value of the synthesized ZnONPs was measured as 50 μg/mL^{-1} against *S. aureus*, 25 μg mL^{-1} against *P. aeruginosa* and 12.5 μg mL^{-1} against *E. coli* [47]. In our study, lower MIC values were considered to be due to high ZnO concentration. In another study, ZnONPs were synthesized by using *Emblica officinalis* Gaertn extracts and their antimicrobial activity was investigated by Anbukkarasi *et al* [48]. The MIC value of the synthesized ZnONPs was measured as 62.50 μg mL^{-1} against *B. subtilis*, 31.25 μg mL^{-1} against *E. coli*. The MIC value versus *B. subtilis* was found as 62.50 μg mL^{-1} for pH =10, 400 °C, pH = 10, 500 °C and 31.25 μg mL^{-1} for pH = 10, 300 °C in our work. In a study conducted by Aleaghil *et al*, the MIC value of ZnONPs against *S. aureus* was measured as 625 μg mL^{-1} [49]. Whereas, in our study, the MIC values against *S. aureus* of all the samples synthesized at different temperatures and pH values were measured between 125 and 250 μg mL^{-1}. ZnONPs were synthesized and the MIC value against *P. aeruginosa* was measured as 15.625 μg mL^{-1} by Hoseinzadeh *et al* [50]. Compared with our study, we found that ZnONPs synthesized by *S. spinosum* showed more effective antimicrobial activity against *P. aeruginosa* and lower MIC values were obtained. The MIC value of all synthesized ZnONPs versus *P. aeruginosa* was measured as 125 μg mL^{-1}. In the work performed by Saadat *et al*, the MIC value of the synthesized nanoparticles against *P. aeruginosa* was found to be 300 μg mL^{-1} [51]. In our study, we obtained lower MIC values and ZnO nanoparticles showed more antimicrobial activity against *P. aeruginosa*.

4. Conclusions

ZnONPs were synthesized by green synthesis using *S. spinosum* leaf extract and zinc acetate dihydrate. The effects of different pH (4, 7 and 10) media and different temperatures (100, 200, 300, 400 and 500 °C) on the

formation of ZnONPs have been investigated. According to the results, no nanoparticle formation was observed at 100 and 200 °C. ZnO nanoparticle formation began to occur after 300 °C. The formation of nanoparticles was directly proportional to the increasing temperature. According to the results of SEM and XRD, triangle pyramid, rectangular, rod and plate structures were observed at high temperatures and also high crystallization occurred at 400 and 500 °C at all three pH media. Considering the antimicrobial results of nanoparticles, the study revealed that they are evaluated and the MIC values ranged from 15.625 to 250 μg mL^{-1}. As a result, it was determined that synthesized nanoparticles were more effective against *Candida* species. The electrochemical investigation showed that ZnONPs have a good electrochemical activity in alkaline media, hence ZnONPs synthesized via *S. spinosum* can be considered as candidate electro catalysts in electrochemical applications.

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Conflict of interest

The authors declare that there is no conflict of interest regarding the publication of this paper.

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