



Sustainable synthesis of ZnO nanoparticles from *Brickellia cavanillesii* extract with enhanced photocatalytic and antimicrobial properties

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Abstract

Zinc oxide (ZnO) semiconductor nanoparticles (NPs) were synthesized via a green chemistry approach using a phytochemical-rich natural extract derived from *Brickellia cavanillesii* in an environmentally friendly approach. Multiple techniques were used to study the properties of the obtained materials, including Scanning Electron Microscopy (SEM) to investigate surface morphology. At the same time, Transmission Electron Microscopy (TEM) was utilized to measure morphological characteristics and estimate the average particle size of the obtained materials; the analysis results suggest a direct influence of the extract and its concentration on particle size. The synthesized NPs presented high crystallinity and a zincite hexagonal shape, as analyzed employing X-ray diffraction (XRD). The photocatalytic performance of the NPs was systematically analyzed by monitoring the photodegradation of a series of organic dye pollutants under ultraviolet (UV) irradiation. The selected dyes: Congo red, methylene blue, malachite green, rhodamine B, and methyl orange, served as representative models for both anionic and cationic contaminants commonly found in industrial wastewater. The results demonstrated a remarkable capability of the materials to remove these pollutants from aqueous solutions, achieving complete degradation (100%) of Congo red and methylene blue. Furthermore, the synthesized NPs were able to eliminate at least 81% of malachite green, rhodamine B, and methyl orange, highlighting their strong potential for application in wastewater treatment and environmental remediation. Finally, the antimicrobial activity of NPs against multiple bacteria (*Bacillus subtilis*, *Staphylococcus aureus*, *Klebsiella pneumoniae*, and *Escherichia coli*) was analyzed, demonstrating a bacteriostatic effect on the bacteria.

Keywords Antimicrobial · *Brickellia cavallinesii* · Environmental remediation · Organic dyes · ZnO nanoparticles

Introduction

Nanomaterials are a diverse and rapidly evolving materials, characterized by at least one dimension measuring less than 100 nm (nm) [1]. These materials can take many structures, including nanoparticles (NPs), nanoflowers, nanosheets, and nanotubes, among many other nanostructures. These morphologies determine the properties and performance of their applications [2]. The growing interest in nanomaterials stems from their wide-ranging applications and unique physicochemical properties. They have enabled significant advances in fields such as sustainable agriculture, drug delivery, clinical therapeutics, diagnostics, tissue engineering, biosensing, and electronics [3]. These benefits arise from their high surface energy, large contact area, and enhanced properties compared to their bulk counterparts [4].

NPs can be synthesized using a wide range of physical and chemical methods. Physical methods include thermal decomposition, pulsed laser deposition, and chemical vapor deposition [5], while chemical methods involve hydrolysis, sol–gel, solvothermal, pyrolysis, and hydrothermal synthesis [6]. Generally, these conventional synthesis processes require a high-energy consumption, emission of toxic chemicals, reliance on complex equipment, harsh synthesis conditions, and the need for vacuum, elevated temperatures, and high pressure [7, 8]. Additionally, they pose significant environmental and health risks due to the use of hazardous solvents and the generation of harmful byproducts [9, 10]. Due to the numerous disadvantages of conventional synthesis methods, green methodologies are emerging as an option to replace environmentally harmful methods [11, 12]. Green NP synthesis is a simple, low-cost, and easily reproducible process that uses natural sources, including vitamins, bacteria, fungi, algae, and plant extracts obtained from various parts of plants such as roots, stems, leaves, flowers, fruits, or seeds, where phytochemicals like alkaloids, flavonoids, polyphenols, terpenoids, proteins, and sugars act as reducing, stabilizing, and capping agents [13]. The aim is to avoid toxic reagents and reduce energy demands by using milder conditions with a lower environmental impact [14, 15].

Numerous previous studies have emphasized the increasing importance of green synthesis methods to obtain metal oxide NPs, particularly through the use of plant extracts as eco-friendly stabilizing agents. This sustainable and environmentally conscious approach has been applied to reduce the use of hazardous chemicals typically involved in conventional synthesis processes. Plant-mediated synthesis has successfully been employed to fabricate a wide range of metal oxide NPs, including copper (Cu) [16], gold (Au) [17], silver (Ag) [18], nickel (Ni) [19], and zinc (Zn), among others [20]. In particular, zinc oxide (ZnO) is an n-type semiconductor that has attracted significant interest owing to its remarkable physicochemical properties, including a ~ 3.37 eV band gap and a comparatively high exciton binding energy of about 60 meV. The intrinsic properties of ZnO make it a multifunctional material with wide applications in fields such as agriculture, medicine, semiconductors, food, electronics, and environmental remediation, including wound healing, sunscreens, anti-cancer therapies, drug

delivery, antimicrobial agents, catalysis, and sensors [21–25]. Its broad utility is due to its excellent optical and electronic characteristics, biocompatibility, low toxicity, and chemical stability, making it attractive for both research and industrial use [26].

A diverse array of plant extracts has been successfully employed to synthesize ZnO NPs employing a green methodology, showcasing the versatility and effectiveness of this method. Notable examples include: *Thymbra Spicata* L [27], *Phoenix roebelenii* [28], *Calligonum comosum* [29], and *Fumaria officinalis* [30], among other plant-based extracts. *Brickellia cavanillesii* (*B. cavanillesii*), also referred to as “prodigiosa,” is a native medicinal plant from Mexico and the southern U.S.A., belonging to the Asteraceae family, known for its use in the treatment of diabetes, hypertension, anxiety, diarrhea, among other conditions, in addition to presenting antioxidant properties [31, 32]. The medicinal properties of *B. cavanillesii* are attributed to the molecules in its phytochemical composition, which include acacetin, kaempferol, quercetin, genkwanin, among multiple bioactive compounds [33]. The rich phytochemical profile of *B. cavanillesii* positions it as a promising candidate for the green synthesis of nanoparticles. These naturally occurring metabolites can serve as both reducing and stabilizing agents, facilitating the formation of nanoparticles under mild, environmentally benign conditions without the need for toxic reagents or high-energy inputs.

Semiconductor ZnO NPs were obtained through an eco-friendly methodology using the extract of *B. cavallinesii* at different concentrations. The obtained materials were studied using various characterization techniques. Subsequently, their ability to eliminate pollutants in wastewater was analyzed by the degradation of Congo red (CR), methylene blue (MB), malachite green (MG), rhodamine B (RB), and methyl orange (MO). The antimicrobial performance of the NPs was assessed by examining their effect on two Gram-positive bacteria: *Bacillus subtilis* (*B. subtilis*) and *Staphylococcus aureus* (*S. aureus*), and two Gram-negative bacteria: *Klebsiella pneumoniae* (*K. pneumoniae*), and *Escherichia coli* (*E. coli*).

Materials and methods

Materials

The synthesis was developed employing *B. cavanillesii* (Cass.) A. Gray leaves obtained from local botany, which is a medicinal plant used to treat hypertension, diabetes mellitus or diarrhea, due to its varied phytochemical composition. As a zinc precursor, zinc nitrate hexahydrate was used and purchased from Sigma-Aldrich with a purity of 99%. The five different colorants used to test the photocatalytic activity were also obtained from Sigma-Aldrich: MB with 99% purity; RB with 96% purity; MO with 98% purity; CR with 90% purity; and MG with 90% purity. Hinton–Muller Agar (HMA) and Lysogeny–Broth (LB) media were purchased from BD Bioxon to perform the antimicrobial test. The Microbiology Laboratory of the University of Guanajuato provided clinical bacterial isolates.

Preparation of *B. cavanillesii* extracts

The extracts of *Brickellia cavanillesii* were obtained using a hot aqueous extraction method. For the 1%, 2%, and 4% synthesis preparations, 0.5 g, 1 g, and 2 g of finely powdered plant material, respectively, were mixed with 50 mL of deionized water in a 100 mL beaker. The mixtures were subjected to continuous magnetic stirring for 2 h at room temperature to promote the diffusion and solubilization of phytochemicals into the aqueous phase. After stirring, the suspensions were heated to 60 °C and maintained at that temperature for 1 h. Finally, the extracts were vacuum filtered to remove solid residues and obtain the clear plant extract.

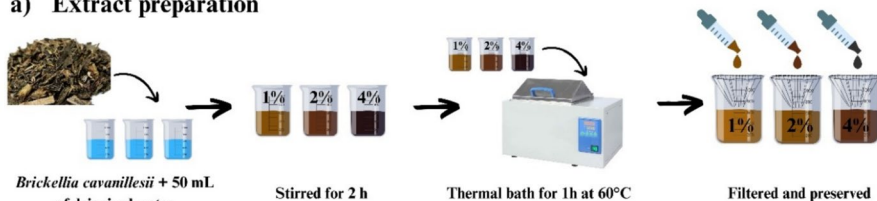
Biosynthesis of ZnO NPs employing *B. cavanillesii* extract

To obtain ZnO NPs, 2 g of the zinc precursor was poured into each filtered solution and stirred for 1 h in the absence of light. Afterward, the resulting solution was placed in a thermal treatment and maintained at a constant temperature of 60 °C in a 12 h lapse. Subsequently, they were calcinated at 400 °C for 1 h. The same procedure was applied to all three solutions. The calcinated NPs were labeled as BC-1% for 1% extract, BC-2% for 2% extract, and BC-4% for 4% extract. The process of obtaining *B. cavallinesii* extracts and synthesizing ZnO NPs is shown in Fig. 1.

Characterization

The particle size distribution and crystallinity were analyzed employing Transmission Electron Microscopy (TEM) and selected area diffraction (SAED), using a JEOL JEM-2100 electronic microscope. The surface morphology and topographical

a) Extract preparation



b) Green synthesis of ZnO NPs

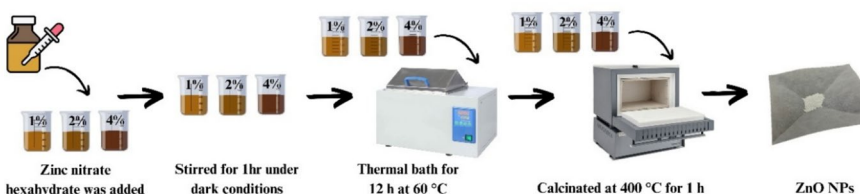


Fig. 1 Extract preparation (a) and green synthesis of ZnO NPs (b)

features of the samples were investigated using Scanning Electron Microscopy (SEM), while the elemental composition was determined through Energy Dispersive X-ray Spectroscopy (EDS). Both analyses were conducted with a JEOL JSM-6310LV scanning electron microscope, providing high-resolution imaging and accurate elemental mapping to evaluate the distribution of various elements within the material. The crystallinity of the materials was studied employing X-ray diffraction (XRD), using a Bruker D2-phase diffractometer; a 2θ scanning angle between 10° and 80° was applied with Cu $K\alpha$ radiation, which provided information about the phase purity of the synthesized nanoparticles. UV–Vis spectroscopy employed a PerkinElmer spectrophotometer in the 800–200 nm range. The identification and characterization of the functional groups present in the syntheses were studied by Fourier Transform Infrared (FTIR) spectroscopy; the analysis was performed with a PerkinElmer spectrophotometer, operating within the spectral range of $2500\text{--}350\text{ cm}^{-1}$.

Photocatalytic assays

Five different dye suspensions were prepared to analyze the ability to eliminate organic pollutants in wastewater of the ZnO NPs. Initially, 50 mg of the catalyst was incorporated to 50 mL of the dye solution at a 15 ppm concentration and set to stir in dark conditions for 30 min. Afterward, the photocatalyst-dye suspension was placed into a UV light reactor with a 10 W UV lamp. Aliquots of 1 mL were taken every 10 min for the first 60 min. Finally, the samples were taken every 30 min until 180 min.

Antibacterial disk diffusion test

Antibacterial effect of ZnO NPs was evaluated against four clinical isolates of bacteria: two Gram-positive (*S. aureus* and *B. subtilis*), and two Gram-negative bacteria (*K. pneumoniae* and *E. coli*). Each clinical isolate bacterium was cultivated in LB broth at a temperature of 37°C . The growth kinetics of the cultures were analyzed by measuring the optical density (OD) employing a UV–Vis spectrophotometer at 600 nm at regular intervals until an OD_{600} value of approximately 0.5 was achieved. Upon reaching the desired OD, 200 μL of the bacterial culture was uniformly spread onto the surface of pre-prepared HMA Petri dishes using a sterile spreader to ensure consistent bacterial coverage. Subsequently, sterile filter paper disks (6 mm in diameter) were aseptically placed onto the agar surface. Each disk was impregnated with 15 μL of the NPs suspensions prepared at varying concentrations (BC-1%, BC-2%, and BC-4% NPs at 1, 2, and 4 mg/mL for each NPs). As a positive control, 10 μL penicillin/streptomycin (GIBCO, 15,140–122) over a paper disk was employed, and 15 μL of water was used as a negative control. After an 18 h incubation period, the inhibition halo size was measured using Image J software, taking the width of the Petri dish as a reference [34]. Kruskal–Wallis with the post hoc Duns test was employed for statistical analysis, employing OriginPro software.

Antibacterial assay using LB liquid medium

The impact of the synthesized ZnO NPs on the growth kinetics of bacterial strains was evaluated. Each strain was cultured in LB medium containing 4 mg/mL of BC-1%, BC-2%, or BC-4% NPs in Erlenmeyer flasks and incubated at 37 °C for 16 h. Growth was monitored by measuring the optical density (OD) at 600 nm every 2 h. At the start of the experiment, an aliquot of each culture was cooled to 4 °C and used as a blank for subsequent measurements. A culture without nanoparticles served as the control. The kinetics of bacterial growth with and without NP treatment are expressed as the mean, with error bars representing the standard deviation ($n=3$). All assays were performed in triplicate to ensure reproducibility.

Results and discussion

Characterization

TEM and SAED

TEM micrographs along with the corresponding histograms of the particle sizes of BC-1%, BC-2%, and BC-4% are illustrated in Fig. 2a, b, and c, respectively. For the BC-1% synthesis, a large aggregation of NPs and sizes in the 10–75 nm range with an average of 38.701 nm can be observed. On the other hand, for the BC-2% synthesis, a slight decrease in the agglomeration of the NPs can be observed, resulting in more distinct and well-separated NPs. These particles fall within a size range from 15 to 50 nm, and 29.14 nm of average size; a considerable reduction in the particle

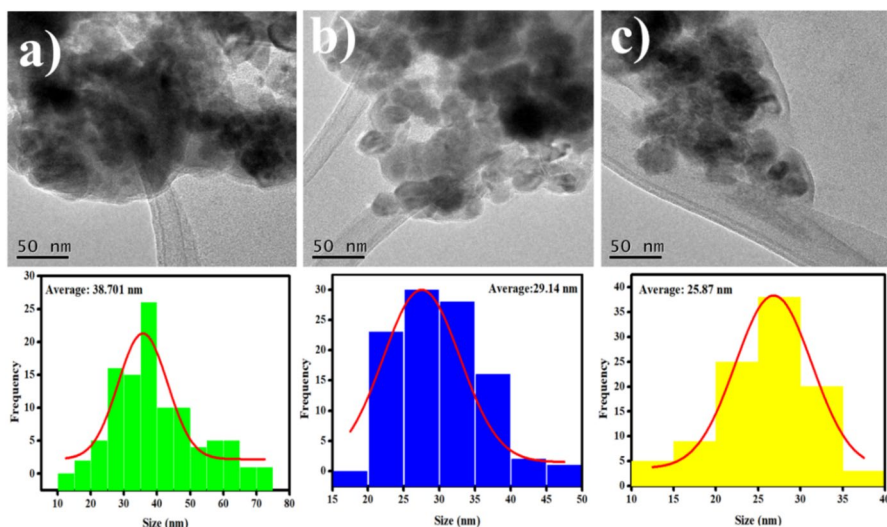


Fig. 2 TEM micrographs and associated size distribution histograms **a** BC-1%, **b** BC-2%, and **c** BC-4%

size compared to BC-1% is also highlighted. For BC-4%, a further reduction in the particle size is observed compared to the rest of the syntheses, with the NPs exhibiting an average size of 25.87 nm. The particles range in size from 10 to 40 nm, showing a smaller distribution. This reduction in the particle size can be attributed to the higher concentration of *B. cavallinesii* extract, which likely provides more organic molecules, including phenolic acids or flavonoids that effectively prevent excessive agglomeration. Increasing the percentage of the extract used affects the particle size, demonstrating the influence of the extract on the properties of the NPs [35]. Figure 3 shows the images obtained by SAED, the planes (101), (002), (101), (102), and (110) were found, which correspond to a hexagonal structure of ZnO [36, 37]. Additionally, the interplanar distance was calculated from HR-TEM micrographs; the interplanar distances were 0.257 nm, 0.262 nm, and 0.265 nm for the syntheses with 1%, 2%, 4%, concentrations, respectively, which correspond to the (101) plane [38].

SEM and elemental analysis

Figure 4a, b, and c shows the SEM micrographs and the elemental composition obtained by EDX for the BC-1%, BC-2%, and BC-4% syntheses, respectively. High particle agglomeration can be observed for all syntheses; however, for the BC-2% and BC-4% syntheses, a more significant presence of isolated particles, less particle agglomeration, and a quasi-spherical morphology can be observed, compared to the BC-1% synthesis, in which the highest agglomeration is observed [39]. The presence of isolated particles is due to the effect of the *B. cavallinesii* extract

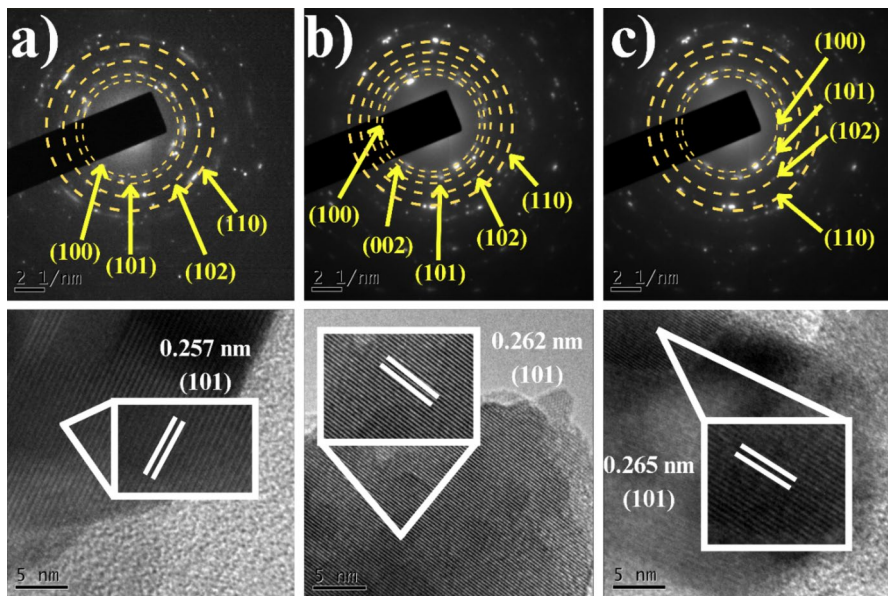


Fig. 3 SAED patterns and HR-TEM micrographs of **a** BC-1%, **b** BC-2%, and **c** BC-4%

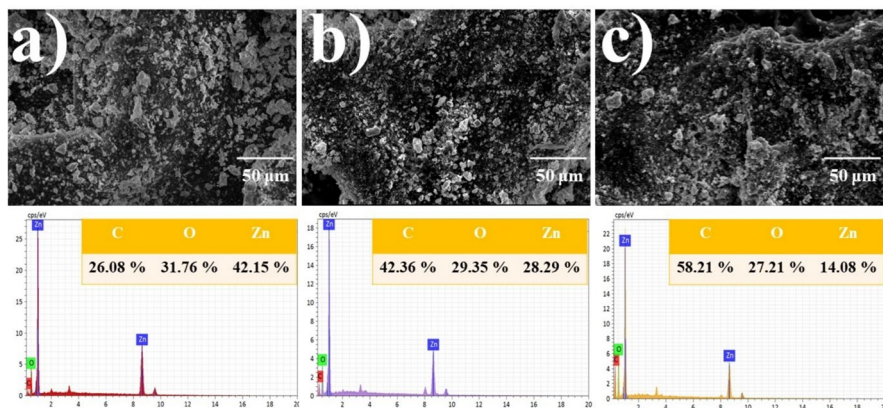


Fig. 4 SEM micrographs and elemental composition of the synthesized ZnO semiconductor employing *B. cavallinesii* extract, **a** BC-1%, **b** BC-2%, and **c** BC-4%

molecules on the synthesis of ZnO NPs. The elemental composition of the syntheses was obtained from the spectra obtained by EDX. The presence of three elements, oxygen (O), carbon (C), and zinc (Zn), with their relative concentrations varying according to the different synthesis conditions employed [40]. Carbon, likely originating from the organic molecules in the *B. cavallinesii* extract, appears in varying amounts depending on the concentration of the extract used in the synthesis, reaching 26.08%, 31.76%, and 42.14% for percentages 1, 2, and 4, respectively [41]. As the carbon increases, the presence of O and Zn is modified, with a considerable reduction in the presence of Zn for the BC-4% synthesis.

XRD

Figure 5 illustrates the XRD pattern of the obtained NPS. It is shown the characteristic ZnO peaks, 31.73° , 34.42° , 36.22° , 47.51° , 56.53° , 62.83° , 66.30° , 67.89° , 72.56° , 76.89° , 81.36° and 89.54° which are the planes (100), (002), (101), (102), (110), (103), (200), (112), (201), (004) and (202), respectively, these results agree with the crystallographic card No. 96-900-4182 [42]. The results are in agreement with a well-stabilized hexagonal zincite structure of ZnO, which is confirmed by the diffraction patterns obtained from the synthesized NPs. The diffractograms show no other signals, demonstrating the formation of a single ZnO phase. Sharp, well-defined peaks indicate the high crystallinity of the materials obtained [43]. The results are consistent with previous publications on synthesizing ZnO nanomaterials using natural extracts as stabilizers. The Debye–Scherrer equation was employed to estimate the average crystallite size of the materials, derived from the XRD patterns (Eq. 1) [44]:

$$D = \frac{\lambda * K}{\beta * \cos(\theta)} \quad (1)$$

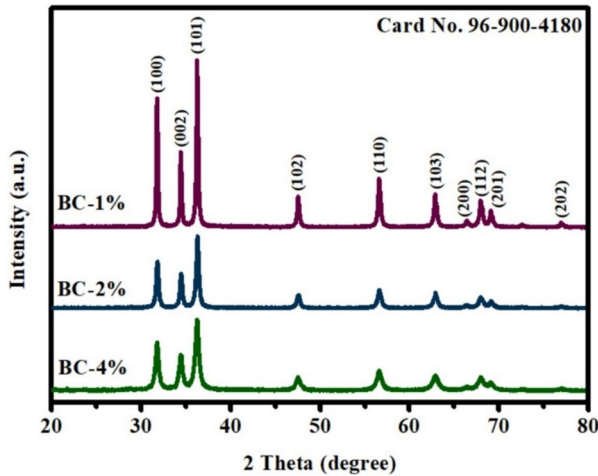


Fig. 5 XRD pattern of the synthesized ZnO NPs

The crystallite size is represented by D , λ stands for the X-ray wavelength, K is the proportionality constant, the full width at half maximum (FWHM) is represented by β , and θ stands for the Bragg's. The calculated crystallite sizes were 32.67 nm, 22.14 nm, and 15.18 nm for BC-1%, BC-2%, and BC-4%, respectively. In the diffractogram presented in Fig. 5, it can be observed that by increasing the percentage of *B. cavallinesii* used in the synthesis of the NPs there is a considerable decrease in the signal especially when comparing BC-1% with BC-2% and BC-4%, impacting the crystallite size which decreases considerably as the percentage of extract increases, highlighting the significant impact of the extract and its concentration on the structural properties of the material. As detailed in Table 1, the lattice parameters corresponding to the hexagonal structure were obtained (Eq. 2) where the interplanar spacing is represented by d ; the Miller indices are represented by h , k , and l ; and the lattice constants are represented by a and c [45].

$$\frac{1}{d^2} = \frac{4}{3} \left(\frac{h^2 + hk + l^2}{a^2} \right) + \frac{l^2}{c^2} \quad (2)$$

Table 1 Lattice parameters and strain of the synthesized ZnO NPs employing *B. cavallinesii* extract

Synthesis	Lattice constants		Strain ($\times 10^{-3}$)	Dislocation density	Crystallinity degree (%)
	a=b	c			
BC-1%	3.2622	5.2040	3.56	9.3×10^{-4}	84.18
BC-2%	3.243	5.1924	5.25	2.204×10^{-3}	76.77
BC-4%	3.2474	5.2186	7.67	4.33×10^{-3}	86.61

As presented in Table 1, the micro-strain was calculated, employing Eq. (3) where ε is the micro-strain. As the crystal size decreases, the strain increases [46].

$$\varepsilon = \frac{\beta}{4 \tan(\theta)} \quad (3)$$

Furthermore, dislocation density was calculated using the Williamson–Smallman relation (Eq. 4), where δ represents the dislocation density, and D is the previously calculated crystallite size [47]. Additionally, the crystallinity degree was obtained employing Eq. 5, where X_c is the crystallinity degree, A_c stands for the area under the crystalline peaks, and A_a is the area under the amorphous peaks, as presented in Table 1 [48].

$$\delta = \frac{1}{D^2} \quad (4)$$

$$X_c = \frac{A_c}{A_c + A_a} * 100 \quad (5)$$

UV–Vis and band gap calculation

Figure 6 shows the absorbance spectrum obtained and the band gap calculation of the NPs using a natural extract of *B. cavallinesii*. The maximum absorbance point of each synthesis can be observed due to the surface plasmon resonance (SPR), located at 374.04 nm, 375.45 nm, and 375.79 nm. A slight red shift can be observed for BC-1%, BC-2%, and BC-4%. The differences between the syntheses are due to the amount of extract molecules present during the synthesis process [49, 50].

The band gap was calculated employing the TAUC model [51]:

$$\alpha h\nu = A(h\nu - E_{bg})^{1/2} \quad (6)$$

where α is the absorption coefficient, h represents Planck's constant, ν corresponds to photon frequency, A is a constant, and E_{bg} signifies the optical energy gap. The calculated values were 3.31 eV, 3.29 eV, and 3.06 eV for BC-1%, BC-2%, and BC-4%, respectively. The red shift observed due to the influence of the extract impacts the band gap; at a higher concentration of extract and a more significant

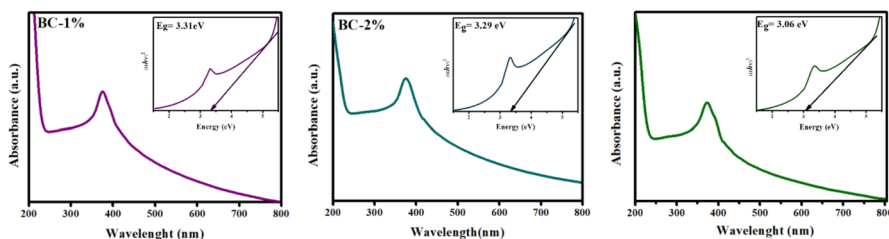


Fig. 6 UV–Vis spectra and band gap calculation of the obtained ZnO NPs employing *B. cavallinesii*

presence of organic molecules during the synthesis process, the band gap decreases, demonstrating a direct influence between the extract and the optical properties of the NPs [52].

FTIR

The functional groups present in the synthesized NPs obtained using *B. cavallinesii* extract were studied using FTIR spectroscopy; the generated spectra are shown in Fig. 7. The syntheses were analyzed from 2500 to 400 cm^{-1} . The spectra present signals located in the fingerprint region with signals at 1375 cm^{-1} and 1113 cm^{-1} , assigned as C–N vibrations characteristic of aliphatic amine groups [49]; 1030 cm^{-1} , related to the C–O stretching vibrations characteristic of phenolic functional groups [53]; and 851 cm^{-1} , which belongs to the –CH stretching [54] of the organic molecules of *B. cavallinesii*. A strong signal located at 651 cm^{-1} , this peak belongs to the Zn–O stretching vibration, which serves as confirmation of the successful synthesis of the ZnO bond [20].

Degradation of organic dyes

Congo red

The photocatalytic performance of the semiconductor NPs synthesized using *B. cavallinesii* extract was studied by analyzing the degradation efficiency of five different organic dyes under UV irradiation. This assessment aimed to determine the effectiveness of the biosynthesized nanoparticles in facilitating the breakdown of complex dye molecules. Figure 8a shows the degradation of CR over time; a similar behavior can be observed for the three syntheses reaching a degradation of 96.71%,

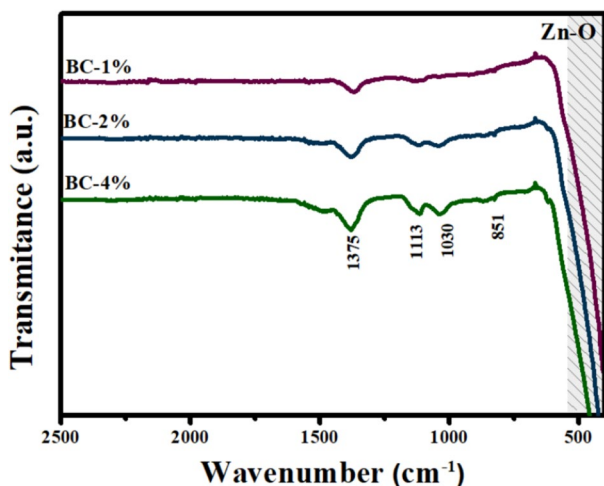


Fig. 7 FTIR spectra of the NPs synthesized employing *B. cavallinesii*

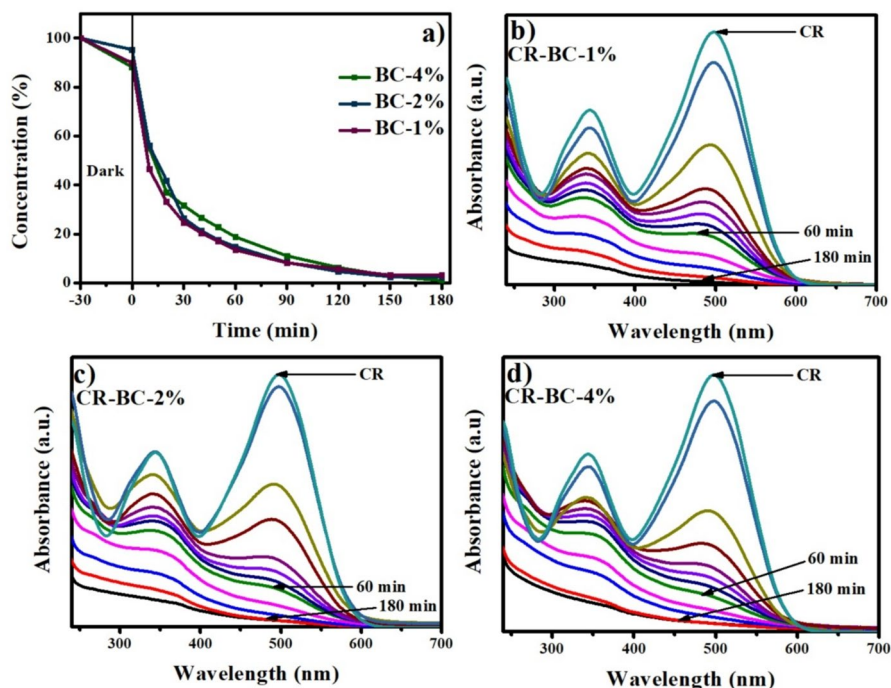


Fig. 8 **a** Photocatalytic degradation of CR using ZnO synthesized NPs under UV light, UV–VIS spectrum of CR degradation using **b** BC-4%, **c** BC-2%, and **d** BC-1%

97.42% and 100% for BC-1%, BC-2% and BC-4%, respectively, in a period of 180 min. Figure 8b, c, and d presents the absorbance spectra of CR degradation obtained from UV–Vis spectroscopy. The decrease in the signal can be observed in all degradations, which present a maximum absorbance point at 498 nm. A degradation of 86.47%, 85.16%, and 81.19% is highlighted for 1%, 2%, and 4%, respectively, in a 60-min lapse. Despite presenting similar behavior, the BC-4% synthesis shows the greatest degradation of the contaminant; this may be due to its properties, which include having the smallest band gap, resulting in less energy required during the photocatalysis process. These results can be compared with Brishti et al., results, where ZnO NPs were synthesized using *Epipremnum aureum* leaves and obtained a 57.6% efficiency [55].

Methylene blue

The degradation of MB is presented in Fig. 9a, where a similar degradation can be seen for BC-1% and BC-2%, with a slightly faster degradation for BC-4%, achieving a degradation of 87.13% in 60 min compared to BC-1% and BC-4%, which reached 82.18 and 81.39, respectively. Figure 9 b, c, and d shows the degradation spectra obtained employing UV–Vis spectroscopy, with a maximum absorption length at 662 nm. The synthesized NPs were able to eliminate 96.6%, 96.1%, and 100% for

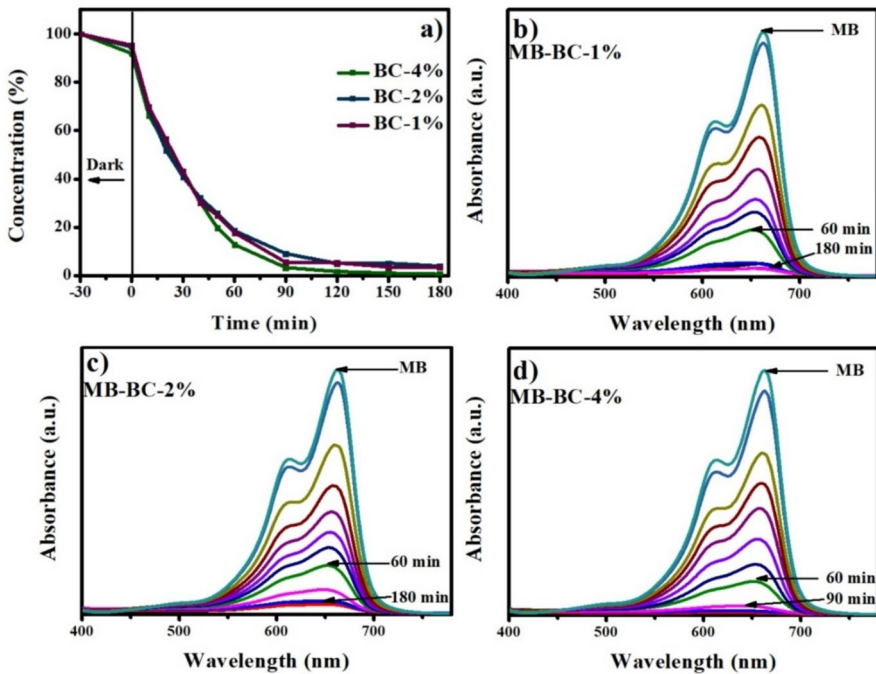


Fig. 9 **a** Photocatalytic degradation of MB using ZnO synthesized NPs under UV light, UV–VIS spectrum of MB degradation using **b** BC-4%, **c** BC-2%, and **d** BC-1%

the 1%, 2%, and 4% syntheses, respectively. As in the photodegradation of CR, a better degradation is highlighted using BC-4%, highlighting properties such as a smaller band gap compared to the rest of the synthesis, a smaller crystallite size of 15.18 nm and a smaller average particle size of 25.87 nm compared to BC-1% and BC-2% which have a particle size of 38.701 nm, and 29.14 nm, respectively. These results can be compared to those reported by Alharthi et al., who synthesized ZnO NPs using an extract derived from *Salvadora persica* leaves. In their study, the ZnO NPs demonstrated a photocatalytic degradation 75% after 150 min of exposure. This comparison highlights the influence of different green synthesis approaches and plant extracts on the overall photocatalytic performance of ZnO-based materials [56].

Malachite green

MG photodegradation under UV light using ZnO NPs obtained from *B. cavalline-sii* is presented in Fig. 10a. At time zero, there is a decrease in the contaminant, and at time 10, there is an increase. This is attributed to the significant adsorption of the contaminant onto surface of the material, which takes place during the stirring period when the photocatalyst and dye are exposed to dark conditions. Subsequently, the photodegradation of MG under UV light occurred without any similar variation. Similar behavior can be seen for the three syntheses, with the BC-4%

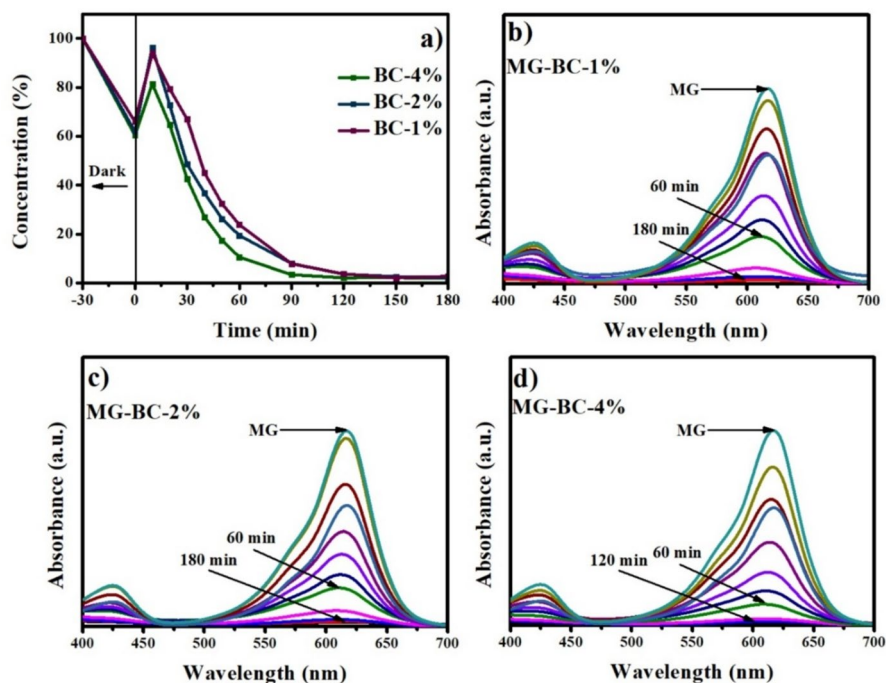


Fig. 10 **a** Photocatalytic degradation of MG using ZnO synthesized NPs under UV light, UV–VIS spectrum of MG degradation using **b** BC-4%, **c** BC-2%, and **d** BC-1%

NPs demonstrating the highest degradation efficiency compared to the rest of the samples. After 60 min of UV exposure, BC-4% achieved an 89.39% degradation. Throughout the experiment, the final degradation percentages for BC-1%, BC-2%, and BC-4% were 97.27%, 97.80%, and 98.14%, respectively. The results highlight the superior photocatalytic performance of BC-4%, which likely benefits from a smaller particle size and smaller band gap value. The obtained results are comparable and superior to previously reported works, demonstrating the great potential of the ZnO NPs obtained in the elimination of organic pollutants such as MG in environmental remediation [57].

Rhodamine B

Rhodamine B (RB) is a difficult-to-degrade dye used in textile industries. It is considered neurotoxic, associated with carcinogenic effects, and multiple human health problems [58]. The photodegradation of RB under UV light was studied using ZnO NPs synthesized as presented in Fig. 11a. It shows lower degradation than the rest of the analyzed contaminants, with similar behavior for BC-1% and BC-4%. Figure 11b, c, and d illustrates the degradation spectra obtained through UV–Vis, showing the changes in absorbance of the RB over time. Initially, the RB dye exhibits a prominent absorption peak at 552 nm, which is characteristic of its properties. As the photodegradation progresses, a progressive decline in the

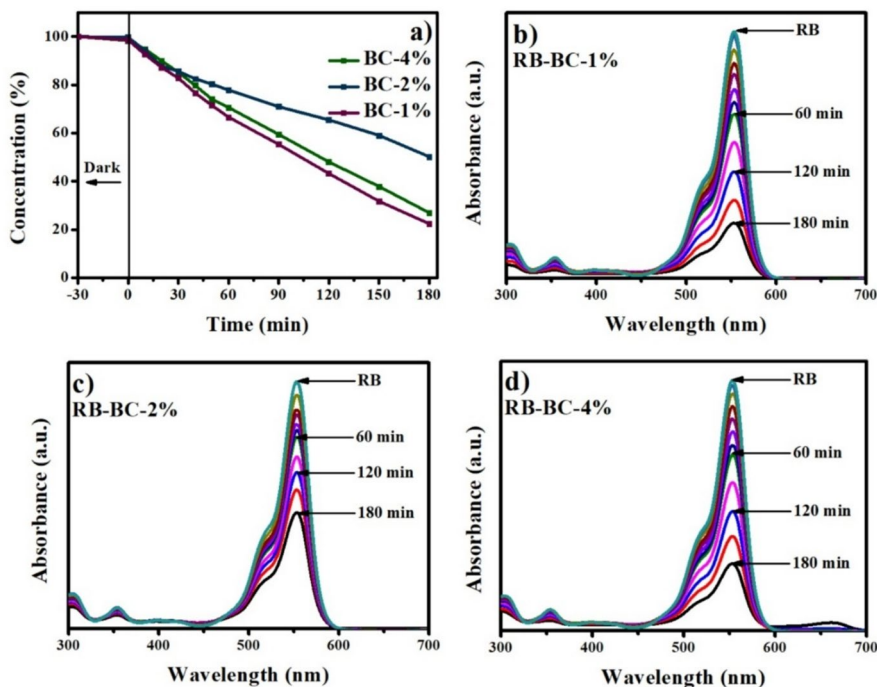


Fig. 11 **a** Photocatalytic degradation of RB using ZnO synthesized NPs under UV light, UV–VIS spectrum of RB degradation using **b** BC-4%, **c** BC-2%, and **d** BC-1%

intensity of the dye's characteristic absorption peak is observed. This attenuation in absorbance indicates the degradation of the dye molecules, signifying the cleavage of chromophore structures under UV light irradiation. At the end of the experiment, the degradation percentages of the dye were 77.56%, 48.79%, and 73.04% for BC-1%, BC-2%, and BC-4%, respectively.

Methyl orange

Finally, the degradation of MO was analyzed as presented in Fig. 12 where the three syntheses present a similar behavior over time with slight differences between them, on the other hand, the absorbance spectra of the degradations analyzed by UV–Vis spectroscopy are illustrated in Fig. 12b, c, and d where the maximum absorbance wavelength is at 462 nm, the decrease in the signal presented indicates the photodegradation of the contaminant where BC-4% stands out as in the rest of the analyzed contaminants achieving a degradation of 52.01% in 60 min. The syntheses eliminated the contaminant by 93.45%, 96.05%, and 91.37%, for BC-1%, BC-2%, and BC-4%, respectively. A comparison of photocatalysis studies for the degradation of organic dyes using ZnO NPs obtained by green synthesis is presented in Table 2.

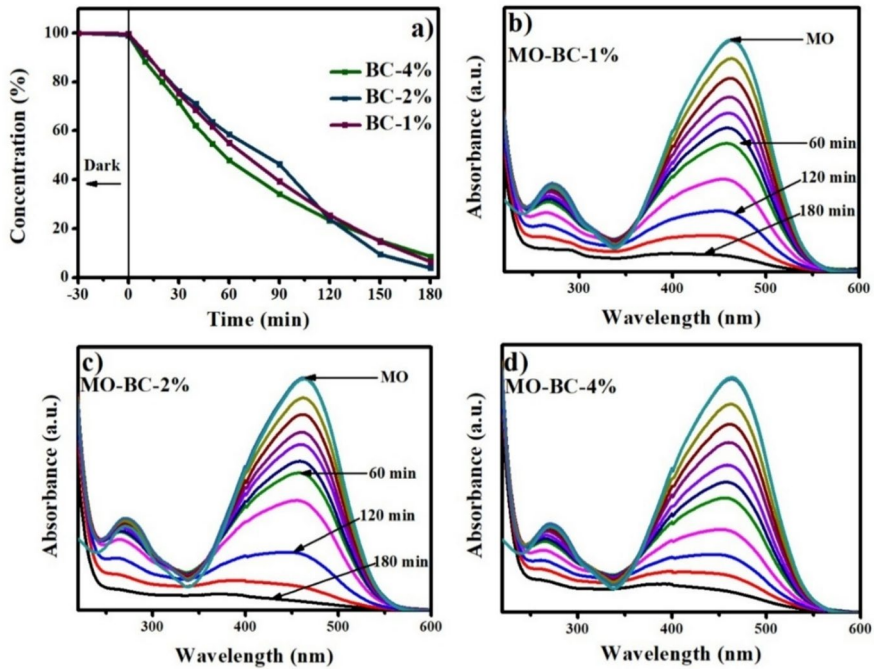


Fig. 12 a Photocatalytic degradation of MO using ZnO synthesized NPs under UV light, UV–VIS spectrum of MO degradation using BC-4% (b), BC-2% (c), and BC-1% (d)

Table 2 Comparison of photodegradations of organic dyes using ZnO NPs obtained by green synthesis

Extract	Dye	Catalyst concentration	Percentage of degradation (%)	Time of degradation (min)	References
<i>Brickellia cavanillesii</i>	CR	15 ppm	100	180	Present work
<i>Epipremnum aureum</i>	CR	10 ppm	57.6	100	[55]
<i>Acacia concinna</i>	CR	25 ppm	92	180	[59]
<i>Brickellia cavanillesii</i>	MB	15 ppm	100	180	Present work
<i>Scutellaria baicalensis</i>	MB	0.05 mg/mL	98.6	210	[60]
<i>Salvadora persica</i>	MB	20 ppm	82	150	[56]
<i>Brickellia cavanillesii</i>	MG	15 ppm	98.14	180	Present work
<i>Ruellia tuberosa</i>	MG	10 ppm	92	150	[61]
<i>Brickellia cavanillesii</i>	RB	15 ppm	77.56	180	Present work
<i>Alchemilla vulgaris</i>	RB	10 ppm	75	120	[62]
<i>Sechium edule</i>	RB	20 ppm	95	75	[63]
<i>Brickellia cavanillesii</i>	MO	15 ppm	96.05	180	Present work
<i>Calotropis procera</i>	MO	20 ppm	81	100	[64]
<i>Moringa oleifera</i>	MO	20 ppm	70	180	[65]

Photocatalytic mechanism

Figure 13 illustrates a proposed mechanism for the photodegradation of organic dyes using ZnO NPs in an aqueous medium. ZnO NPs can generate an electron–hole pair upon receiving energy from UV or visible light with sufficient energy to overcome the band gap. The process begins with the absorption of light energy in the surface of the photocatalyst (Eq. 7), the electrons in the valence band (VB) are excited and promoted to an empty conduction band (CB), leading to the generation of an electron–hole pair (e^-/h^+). The photogenerated holes in the VB interact with the H_2O to produce $OH^\cdot$ (Eq. 8), while interacting with OH^- to produce $OH^\cdot$ (Eq. 9). Then, the electrons in the CB react with the present O_2 to generate superoxide radicals $O_2^{\cdot-}$ (Eq. 10). The produced superoxide molecules are highly reactive and interact with H^+ ions to generate hydrogen peroxide H_2O_2 (Eq. 11 and Eq. 12) [53]. H_2O_2 interacts with $O_2^{\cdot-}$ to produce hydroxyl radicals (Eq. 13). These molecules are known as reactive oxygen species (ROS), which are highly oxidizing molecules that react with the pollutants, eliminating them into less polluting molecules, leading to the mineralization of the contaminant by producing H_2O , CO_2 and, in some cases, remaining products of the reaction (Eq. 14) [66].

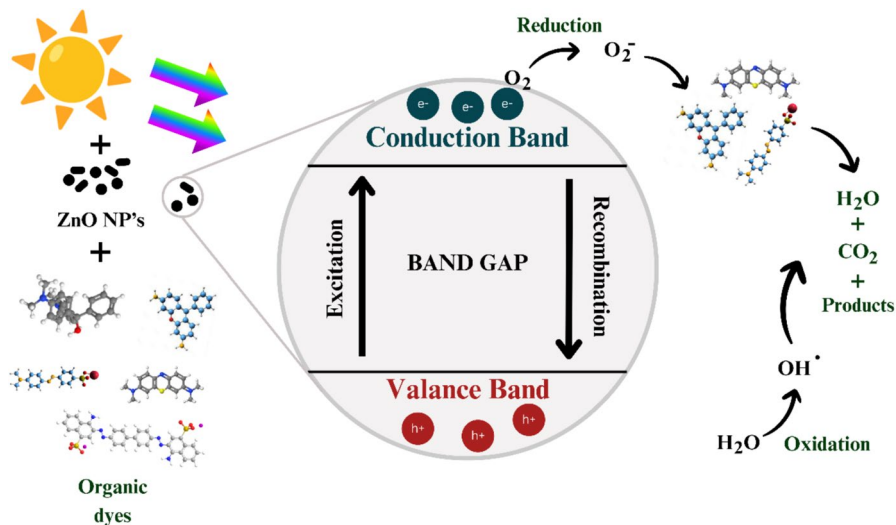
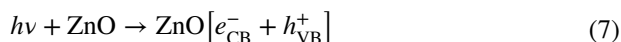
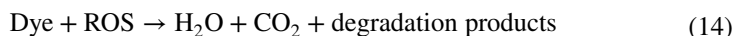
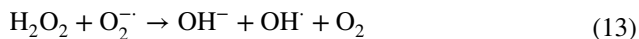


Fig. 13 Proposal of the mechanism of photodegradation of ZnO NPs



Antimicrobial activity of the NPs

Antibiogram of ZnO NPs

The evaluation of antibacterial activity using a disk diffusion test shows no growth inhibition in any of the bacteria tested (Fig. 14). ZnO NPs have been described as a nanomaterial with antibacterial effects [67]. However, our results indicate that ZnO NPs prepared by green synthesis, employing *B. cavanillesii*, do not exhibit

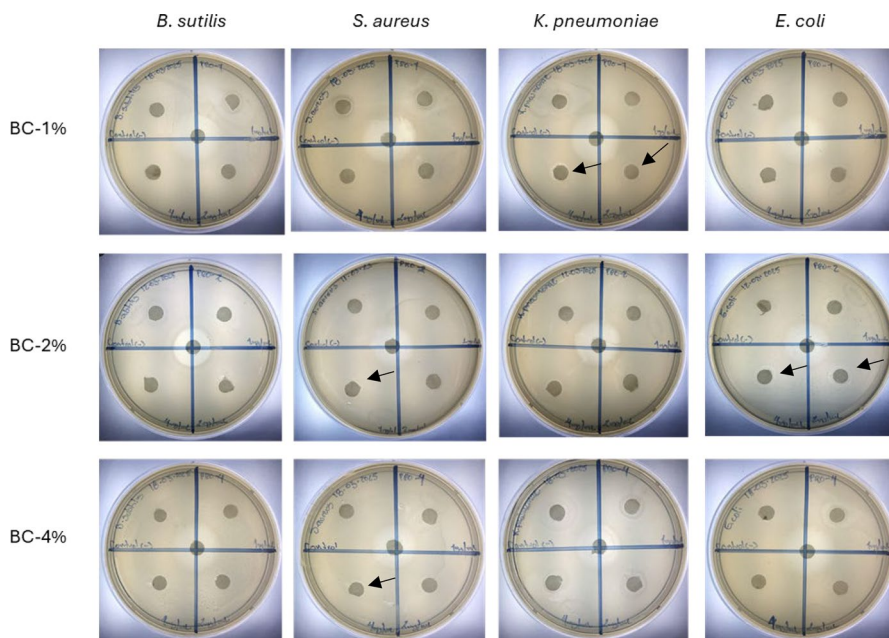


Fig. 14 Antibiogram assay of ZnO NPs. ZnO NPs synthesized using *B. cavanillesii* extract do not induce antibacterial activity

this effect using a disk diffusion assay. The absence of the inhibition halo (Fig. 14) could be due to interactions between NPs and the presence of organic remnants from *B. cavanillesii* extract. Polyphenols from plants can affect nanoparticle size, which directly relates to the antimicrobial effect [68, 69]. It has been reported that ZnO NPs produce ROS, including hydrogen peroxide, which are generated through direct interaction between the NPs and the aqueous medium [70, 71]. Additionally, the size of ZnO NPs plays an essential role in their interaction with bacterial strains; smaller NPs (18–22 nm) promote better interaction due to the increased contact surface between bacteria and nanomaterials [69]. In our case, the ZnO NPs range in size from 25 to 39 nm. Another factor that can influence the antimicrobial effect is the diffusion of the tested NPs and the thickness of the agar used [71, 72]. However, a faded halo was observed at some ZnO NPs concentrations tested on Petri dishes (black arrows, Fig. 14), suggesting a possible bacteriostatic effect meaning that ZnO NPs may inhibit bacterial growth without killing the bacteria. To explore this possibility, antimicrobial activity was evaluated using LB-liquid medium.

Effect of NPs on the kinetic growth of bacterial strains

Another way to evaluate the antimicrobial effect is by monitoring the kinetic growth of bacterial cultures in LB-liquid medium. In this approach, bacteria were grown in LB medium supplemented with 4 mg/ml of ZnO NPs. Our results show a decrease in bacterial growth in all cases (Fig. 15), suggesting a bacteriostatic effect of the ZnO NPs synthesized using *B. cavanillesii* extract. Additionally, we can indicate that the remnants of *B. cavanillesii* extract may impede the proper diffusion of ZnO NPs in the disk-diffusion assay on the Petri dishes containing the LB-agar medium. Since the primary antimicrobial mechanism of ZnO NPs involves the generation of ROS and hydrogen peroxide by direct interaction with aqueous medium [70], the NPs suspension in LB medium may enhance this interaction, leading to increased ROS and greater contact between NPs and bacterial cells, resulting in a bacteriostatic effect. A relevant observation is that Gram-positive bacteria presented a more significant decrease in growth than Gram-negative strains (Fig. 15). This difference can be explained by the outer structure of Gram-negative bacteria, which have different layers that confer protection against the external environment [69, 73]. Besides, Gram-negative bacteria have a thick peptidoglycan layer that provides additional protection [74].

Conclusion

In this study, ZnO semiconductor NPs were obtained following a green methodology employing *B. cavallinesii* extract. Multiple characterization techniques demonstrated the impact of the extract concentration on the material characteristic; the particle size analyzed by TEM is considerably reduced by increasing the percentage of the extract, obtaining particle sizes less than 20 nm. On the other hand, a hexagonal zincite phase was found by XRD, and the influence of the extract on the crystallite size was related, which is reduced by increasing the percentage of the extract.

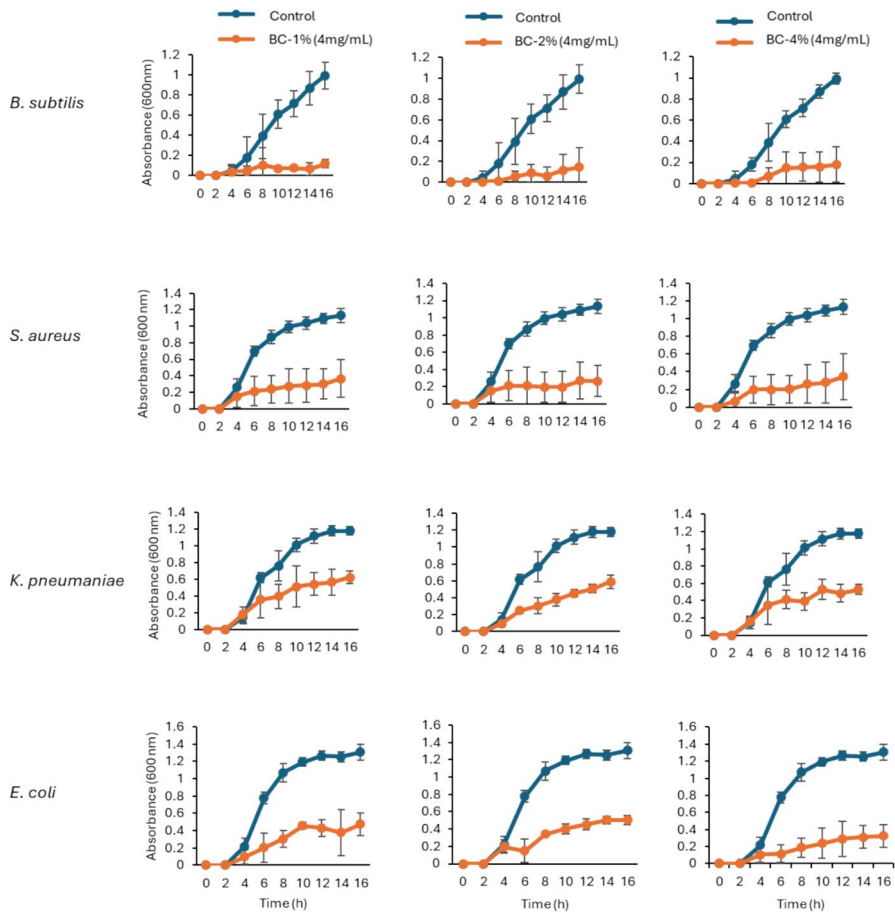


Fig. 15 Effect of synthesized ZnO NPs employing *B. cavallinesii* extract on the kinetic growth profiles of bacterial strains

The variations in the properties of the materials are due to the phytochemicals of the extract, which function as stabilizing agents. The obtained NPs demonstrated remarkable efficiency in the degradation of multiple organic dyes under UV light, highlighting the results in the degradation of CR, MB, and MG, by degrading 100%, 100%, and 98.14%, respectively, demonstrating the potential of NPs for the treatment of contaminated water. Additionally, the results of the antimicrobial activity of the NPs obtained employing the disk diffusion method demonstrated a possible bacteriostatic effect, demonstrating the inhibition of bacterial growth without killing the bacteria. Notably, the inhibitory effect exhibited greater potency against Gram-positive bacterial strains compared to their Gram-negative counterparts, likely due to differences in cell wall structure and permeability. These findings underscore the potential of the synthesized NPs as effective bacteriostatic agents, particularly for applications in the biomedical field.

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Authors contribution A. Villegas-Fuentes: Investigation, formal analysis and experimental. D.K. Meza-Martinez: Formal analysis and methodology. L.M. Olmos-Ortiz: Supervision, writing review and methodology. A.R. Vilchis-Nestor: Writing review and investigation. P.A. Luque: Conceptualization, resources, and editing.

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Data availability No datasets were generated or analysed during the current study.

Declarations

Conflict of interest The authors declare no conflict of interest.

Ethical approval This study does not involve experiments on human tissues.

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