

PHOTOCATALYTIC ACTIVITY INDUCED BY ZnO NANOPARTICLES BIOSYNTHESIZED BY *TURBINARIA DECURRENS* EXTRACT

M. Ramya Anjalin¹, D. Shanmuga Priya^{1*}, T. Akkini Devi¹, S. Kalaiarasi¹, C. Vedhi²

¹Research Scholar (Reg. No.: 22212012032003),

^{1,1*}PG & Research Department of Chemistry, A.P.C. Mahalaxmi College for Women, Thoothukudi, Affiliated to Manonmaniam Sundaranar University, Abishekapatti, Tirunelveli-627012, Tamil Nadu, India.

²PG & Research Department of Chemistry, V.O. Chidambaram College, Thoothukudi, Tamil Nadu, India.

Corresponding Author: dshanmugapriyaresearch@gmail.com

ABSTRACT

Turbinaria decurrens, a brown algae from the *Sargassaceae* family, was gathered from the Thoothukudi coastal area and employed its ethanol extract for the biosynthesis of ZnO nanoparticles. UV–Visible spectroscopy (UV–Vis Spectroscopy) displayed a prominent absorption peak at 356 nm, verifying ZnO nanoparticles formation. Fourier Transform Infrared Spectroscopy (FT-IR) analysis identified functional groups that stabilize the nanoparticles and confirmed the Zn–O bond. X-ray Diffraction (XRD) patterns exhibited sharp peaks matching the hexagonal wurtzite crystal structure of ZnO. High-resolution transmission electron microscopy (HR-TEM) and Scanning electron microscopy (SEM) images revealed rod-like and flake-like structures of morphology, while energy dispersive X-ray analysis (EDAX) verified Zn and O as the primary elements. The photocatalytic activity of the ZnO nanoparticles was tested via methylene blue dye degradation under light exposure, showing a progressive reduction in the 660 nm absorption peak and attaining 66% degradation in 80 minutes. Kinetic studies revealed adherence to both pseudo-zero-order and pseudo-first-order models, with superior fit to the zero-order model. Furthermore, the nanoparticles demonstrated antibacterial effects against Gram-positive and Gram-negative bacteria and antioxidant properties in the 2,2-Diphenyl-1-picrylhydrazyl (DPPH) scavenging assay. These findings indicate that *Turbinaria decurrens*-derived ZnO nanoparticles are promising photocatalysts and eco-friendly materials for environmental and biomedical purposes.

Keywords: *Turbinaria decurrens*, ZnO nanoparticles, methylene blue, photocatalytic activity, HR-TEM, DPPH assay

How to cite this article: M. Ramya Anjalin¹ D. Shanmuga Priya^{1*} T. Akkini Devi¹ S. Kalaiarasi¹ C. Vedhi². PHOTOCATALYTIC ACTIVITY INDUCED BY ZnO NANOPARTICLES BIOSYNTHESIZED BY *TURBINARIA DECURRENS* EXTRACT. Int J Drug Deliv Technol. 2026;16(85s):325-339. DOI: 10.25258/ijddt.16.85s.29.

1. INTRODUCTION

Nanotechnology involves the development, synthesis, characterization, and application of materials and devices through precise control of their size and shape at the nanoscale. Nanomaterials exhibit distinct physicochemical properties compared to their bulk counterparts, which are intrinsically linked to their dimensions and morphology [1]. Nanoparticles represent the smallest category of microscopic or ultrafine particles, ranging from 1 to 100 nm in size [2]. Researchers have found that particle size influences the physicochemical traits of materials, highlighting the importance of these nanostructures. Nanoparticles currently hold a prominent position among various nanostructures due to widespread interest [3, 4].

However, their application is not always entirely safe owing to the precursors involved. Several techniques exist for synthesizing ZnO nanostructures, including solvothermal [5], sol-gel [6], reduction [7] and co-precipitation methods [8]. Among these, co-

precipitation stands out as the preferred approach for ZnO nanoparticles production because it is straightforward, cost-effective, eco-friendly, suitable for large-scale manufacturing, and utilizes precursors like NaOH solutions that enable low-temperature processing. Biosynthesis of ZnO nanoparticles has also been documented using similar principles [9].

Marine algae are primarily categorized into three groups based on pigmentation: brown, green, and red algae. Brown algae from the *Sargassaceae* family are prevalent in tropical and subtropical marine habitats. The genus *Turbinaria* commonly adheres to rocky coastal substrates and is renowned for its abundance of bioactive compounds, such as fucoxanthin, phenolics, and polysaccharides [10]. *Turbinaria decurrens*, notable brown seaweed, contains antioxidant, antimicrobial, and pharmacologically potent metabolites that support nanoparticles synthesis and stabilization. *Turbinaria decurrens* is particularly rich in phenolic compounds, flavonoids,

fucoxanthin, sulfated polysaccharides, and other bioactive metabolites that act as natural reducing and capping agents during nanoparticles synthesis. These phytochemicals facilitate the formation of stable ZnO nanoparticles while minimizing the use of hazardous chemicals. Moreover, *Turbinaria decurrens* is abundantly available along the Gulf of Mannar coast, making it a sustainable, renewable, and cost-effective marine resource for green nanomaterial synthesis. Compared with many terrestrial plant extracts, *Turbinaria decurrens* offers the additional advantage of abundant marine polysaccharides and sulfated compounds, which improve nanoparticles stabilization and provide a sustainable marine biomass for large-scale synthesis [11].

Methylene blue, a synthetic dye widely used in textiles, paper, pharmaceuticals, and printing, poses significant environmental and health risks when released into water bodies. Dye pollutants in wastewater impair light penetration, disrupt photosynthesis in aquatic life, and can induce toxic or carcinogenic effects [12]. Traditional wastewater treatments often fail to fully eliminate these dyes. Consequently, photocatalytic degradation with semiconductor nanoparticles has become a promising, sustainable solution [13]. ZnO nanoparticles are ideal photocatalysts owing to their potent oxidative capacity, high photosensitivity, and chemical stability. Upon light exposure, they produce reactive oxygen species like hydroxyl radicals and superoxide ions, effectively breaking down methylene blue into non-toxic byproducts [14].

In recent years, ZnO nanoparticles have garnered substantial interest for their exceptional biological attributes, especially antibacterial and antioxidant effects [15]. They demonstrate robust antimicrobial action against Gram-positive and Gram-negative bacteria via mechanisms including reactive oxygen species production, cell membrane damage, and metabolic disruption. The DPPH assay is a standard method to assess the radical-scavenging potential of nanomaterials by evaluating their ability to donate hydrogen or electrons to stable free radicals [16].

Unlike previous algae-mediated ZnO nanoparticles, the present work combines comprehensive structural characterization with photocatalytic, antibacterial, antioxidant and electrochemical investigations in a single study. Furthermore, the use of *Turbinaria decurrens*, which has been rarely explored for ZnO biosynthesis, provides additional novelty. This study investigates the biosynthesis of ZnO nanoparticles employing ethanol extract from the brown seaweed *Turbinaria decurrens*. The nanoparticles were analyzed using UV-Vis spectroscopy, FT-IR, XRD, HR-TEM, SEM and EDAX. Cyclic voltammetry was conducted to

study their electrochemical properties. Additionally, photocatalytic performance was tested via methylene blue degradation under light, alongside antibacterial assays against selected strains and DPPH-based antioxidant evaluation, underscoring their potential environmental and biomedical applications.

2. MATERIALS AND METHODS

2.1 Materials

All chemicals used in this study, including zinc sulphate heptahydrate ($\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$), sodium hydroxide (NaOH), methanol, and 2,2-diphenyl-1-picrylhydrazyl (DPPH), were of analytical reagent grade, purchased from Merck, India, and used without further purification. Ethanol (99.9%) was used as the extraction solvent. *Turbinaria decurrens* (Figure 1) was collected from the Thoothukudi coast, Tamil Nadu, India. The collected seaweed was washed, shade-dried and powdered for extract preparation. Double-distilled water was used throughout the study. Methylene blue was used as the model dye for the photocatalytic degradation experiments.



Figure 1. *Turbinaria decurrens*

2.2 Preparation of seaweed extract

Turbinaria decurrens was collected from Thoothukudi coastal waters. Samples were cleaned with seawater, then tap water, to eliminate epiphytes and debris, followed by a distilled water rinse and air-dried in darkness for 3 days [17]. Dried material was ground into coarse powder using a mixer and stored airtight. For extraction, 5 g of the powdered seaweed was mixed with 100 mL of ethanol (99.9%) and heated in a water bath at 60°C for 30 min. The extract was then cooled, filtered through Whatman No. 1 filter paper, and used for the biosynthesis of ZnO nanoparticles.

2.3 Synthesis of Zinc Oxide Nanoparticles

Zinc oxide nanoparticles were synthesized via a precipitation route. 1 M zinc sulphate heptahydrate ($\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$) solution was prepared by dissolving in 50 mL of double-distilled water. 20 ml of *Turbinaria decurrens* extract was added under continuous

stirring using a magnetic stirrer for 1 h. Subsequently, 2 M NaOH solution was added dropwise under constant stirring until the formation of a white precipitate was observed. The reaction mixture was agitated for 2 h, centrifuged at 3500 rpm for 15 min (REMI R-8C centrifuge), dried at 60°C for 24 h, and calcined at 400°C for 2 h. The resulting white ZnO nanoparticles underwent characterization [18]. Figure 2 illustrates the biosynthesis schematic representation of the ZnO nanoparticles using *Turbinaria decurrens* extract.

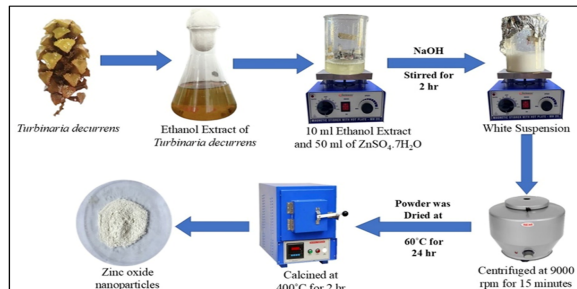


Figure 2. Biosynthesis of ZnO nanoparticles using *Turbinaria decurrens* extract

2.4 Photocatalytic measurement

The photocatalytic activity of *Turbinaria decurrens* using ZnO nanoparticles was evaluated by methylene blue degradation under sunlight, monitored by using UV-Vis spectroscopy. Photocatalytic experiments were performed under natural sunlight. The light intensity was not measured because a calibrated light intensity metre was unavailable. To ensure experimental consistency, all photocatalytic experiments were carried out under identical environmental conditions using the same location, time period, and experimental setup. In a standard run, 0.2 g of ZnO nanoparticles was dispersed in 100 mL of a 1 ppm methylene blue dye solution (pH 7), stirred in the dark for 1 h to establish adsorption-desorption equilibrium before irradiation. During irradiation, 10 mL aliquots were withdrawn at 10 min intervals, and their absorbance was measured using a UV-Vis spectrophotometer [19].

2.5 Antibacterial Activity

The antibacterial activity of the synthesized ZnO nanoparticles was evaluated using the agar disc diffusion method [20]. Sterile 6 mm paper discs containing different concentrations of the sample were placed on agar plates inoculated with the test bacterial strains. Ciprofloxacin was used as the positive control. The plates were incubated at 37°C for 24 h and the antibacterial activity was determined by measuring the diameter of the inhibition zones (mm).

2.6 Antioxidant Activity via DPPH Assay

The antioxidant activity of ZnO nanoparticles was determined using the DPPH free radical scavenging

assay [21, 22]. Varying concentrations were mixed with 3 mL of DPPH methanol solution, incubated in the dark for 30 min at room temperature, and absorbance was read at 517 nm. Ascorbic acid served as a standard. Scavenging percentage calculated as:

DPPH radical scavenging effect

$$= \frac{(A_0 - A_1)}{A_0} \times 100 \% \text{ inhibition}$$

Where A_0 is control absorbance, A_1 is sample/reference absorbance.

2.7 Characterization of ZnO Nanoparticles

UV-Vis spectral analysis was performed on JASCOV-530, and FT-IR analysis was recorded using a Nicolet Si5 spectrometer (model P-4600) using ATR and KBr pellets. X-ray diffraction (XRD) was performed using an X'Pert Powder X'Celerator diffractometer with a $\text{CuK}\alpha$ monochromatic filter. TEM analysis was performed by using a PHILIPS CM 200 instrument operated at an operating voltage of 200 kV and a resolution of 2.4 Å. Cyclic voltammetry (CV) was measured by electrochemical workstation model 650c. The Scanning Electron Microscope (SEM) and Energy Dispersive X-ray (EDAX) analyses were performed using a JEOL JSM-6700F field emission scanning electron microscope.

3. RESULTS AND DISCUSSION

3.1 Optical, Functional, and Structural Analysis

UV-Vis spectroscopy assessed optical properties of *Turbinaria decurrens* extract-synthesized ZnO nanoparticles (Figure 3a). A sharp peak at 356 nm indicated ZnO's band-gap transition in UV. This absorption peak confirms the successful formation of ZnO nanoparticles and is in good agreement with previous studies, which have reported characteristic UV absorption bands for green-synthesized ZnO nanoparticles in the range of 355–360 nm using different biological sources [23].

$$(\alpha h\nu)^2 = A (h\nu - E_g)$$

----- (1)

Where α is the absorption coefficient. $h\nu$ is the photon energy, and E_g is the direct band gap energy of ZnO nanoparticles using *Turbinaria decurrens*. The Tauc plot of $(\alpha h\nu)^2$ vs $h\nu$ is shown in Figure 3b. By extrapolating the linear portion of the curve to the photon energy axis, the optical band gap energy of the synthesized ZnO nanoparticles was determined to be 3.51 eV. This value is slightly higher than that of bulk ZnO, which may be attributed to the quantum confinement effect associated with the nanoscale particle size. Similar band gap values have been reported in previous studies on ZnO nanostructures, where variations in the optical band gap were attributed to differences in particle size, crystal structure, morphology, and synthesis conditions [24]. This wide band gap property makes ZnO

nanoparticles suitable for applications in photocatalysis.

The FT-IR spectra of the *Turbinaria decurrens* extract and the biosynthesized ZnO nanoparticles were examined to identify the functional groups involved in the reduction and stabilization of the nanoparticles. A detailed comparison of the spectra from the *Turbinaria decurrens* ethanol extract (Figure 3c) and the ZnO nanoparticles (Figure 3d) reveals key differences attributable to the synthesis process. In the extract spectrum, a broad band at approximately 3327 cm⁻¹ corresponds to O-H stretching from phenolic and alcoholic groups. Peaks at 2973 cm⁻¹ and 2888 cm⁻¹ indicate C-H and O-H stretching, respectively, while absorptions at 1381 cm⁻¹, 1087 cm⁻¹, and 1046 cm⁻¹ are assigned to C=O, C-O, and C-O-O stretching vibrations. Additionally, a band at 880 cm⁻¹ signifies aromatic C-H bending. These features confirm the presence of phytochemicals, including flavonoids, phenols, and terpenoids, which facilitate the reduction and capping of ZnO nanoparticles.

In the FT-IR spectrum of the ZnO nanoparticles, the O-H stretching band shifted to 3395 cm⁻¹ [25], while the bands at 2918 cm⁻¹ and 2850 cm⁻¹ were assigned to C-H stretching and O-H stretching vibrations of carboxylic acids, respectively [26, 27]. The absorption bands at 1384 cm⁻¹ and 1072 cm⁻¹ correspond to C=O asymmetric stretching and C-O stretching vibrations, respectively [28], whereas the peaks at 1041 cm⁻¹ and 832 cm⁻¹ are attributed to C-O-O symmetric stretching and aromatic C-H bending vibrations. The characteristic absorption bands observed in the 400–600 cm⁻¹ region, particularly at 607 cm⁻¹ and 465 cm⁻¹, are assigned to Zn-O stretching vibrations, confirming the successful formation of crystalline ZnO nanoparticles [29].

The structural characteristics of the ZnO nanoparticles synthesized using *Turbinaria decurrens* extract were investigated by X-ray diffraction (XRD) (Figure 3e). XRD analysis was performed using Cu K α radiation. The Cu mentioned in the XRD analysis refers to the Cu K α X-ray radiation source (λ = 1.5406 Å) used in the diffractometer and does not indicate the presence of copper in the synthesized ZnO nanoparticles.

The average crystallite size was calculated using the Debye-Scherrer equation:

$$D = k\lambda/\beta \cos \theta \quad \text{----- (2)}$$

Where, k is the Scherer's constant, λ is the wavelength of the X-ray and β and θ are the half-width of the peak and half of the Bragg angle, respectively.

The XRD pattern of the synthesized ZnO nanoparticles exhibits diffraction peaks at 2θ values

of 31.59°, 34.27°, 36.11°, 47.86°, 56.42°, and 62.69°, indexing to the hexagonal wurtzite structure in accordance with Joint Committee on Powder Diffraction Standards data (JCPDS card No. 36-1451) [30]. In addition to the characteristic ZnO diffraction peaks, a few crystallite diffraction peaks of low intensity were observed. These crystalline peaks may be attributed to residual phytochemical species or trace secondary phases formed during the biosynthesis process. However, their very low intensity indicates that the ZnO phase remains dominant, confirming the successful formation of crystalline ZnO nanoparticles. Peak broadening further indicates the nanoscale dimensions of the synthesized particles [31]. The calculated average crystallite size is approximately 46.17 nm, consistent with the nanocrystalline nature of the material. Furthermore, dislocation density (δ) and microstrain (ϵ) were assessed using established theoretical formulas.

$$\delta = \frac{1}{D^2} \quad \text{----- (3)}$$

$$\epsilon = \frac{\beta}{(4 \tan \theta)} \quad \text{----- (4)}$$

Where δ = Dislocation density (m⁻²), D = Average crystallite size, ϵ = Micro-strain, β = FWHM of the diffraction peak (in radians), θ = Bragg's angle. The calculated dislocation density was 0.018988 m⁻², suggesting a comparatively low level of structural defects within the ZnO crystal lattice. Similarly, the micro-strain value was determined to be 0.011144, indicating the presence of a small degree of lattice distortion caused by the nanoscale particle dimensions, as presented in Table 1. The relatively low values of dislocation density and micro-strain demonstrate the good crystalline quality of the synthesized ZnO nanoparticles, which is further supported by the presence of sharp and well-defined diffraction peaks observed in the XRD pattern.

The lattice parameters of the synthesized ZnO nanoparticles were calculated from the XRD data. ZnO possesses a hexagonal wurtzite crystal structure, in which the lattice parameters along the basal plane are equal ($a = b \neq c$) [30]. To determine the ZnO hexagonal structure, lattice constants a and c were calculated by using the formula in the equation.

$$\frac{1}{d^2} = \frac{4}{3} \cdot \frac{h^2 + hk + k^2}{a^2} + \frac{1}{c^2} \quad \text{----- (5)}$$

The lattice constants were found to be $a = b = 3.25$ Å and $c = 5.20$ Å. These values are in close agreement with standard values reported for ZnO, confirming the high crystallinity and phase purity of the prepared nanoparticles. The obtained results

confirm that the *Turbinaria decurrens* extract acts as an efficient reducing and stabilizing agent in the biosynthesis process, leading to the formation of highly crystallite ZnO nanoparticles with a well-defined hexagonal wurtzite structure. Previous studies report that plant phytochemicals reduce zinc ions and stabilize the nanoparticles, leading to the formation of ZnO with a stable wurtzite structure, which supports the present findings [32].

Table 1. XRD peak positions, Miller indices, and crystallographic parameters of biosynthesized ZnO nanoparticles synthesized using *Turbinaria decurrens* extract

2θ	FWHM	FWHM	Miller indices	D	D (nm)	Dislocation density (δ) m ⁻²	Micror-strain (ε)
H	HM (rad)						
31.5991	0.40115	0.006995	100	0.401	20.5912	0.024283	0.006184
34.2759	0.25329	0.004415	002	0.253	32.8502	0.015220	0.003581
36.1150	0.39970	0.006970	101	0.399	20.9175	0.023906	0.005347
47.8647	0.05427	0.000945	102	0.054	160.4325	0.003117	0.000532
56.4266	0.44547	0.007767	110	0.445	20.2514	0.024691	0.003619
62.6999	0.42271	0.007371	103	0.422	22.0174	0.022711	0.003026
Average					46.17	0.018988	0.011144

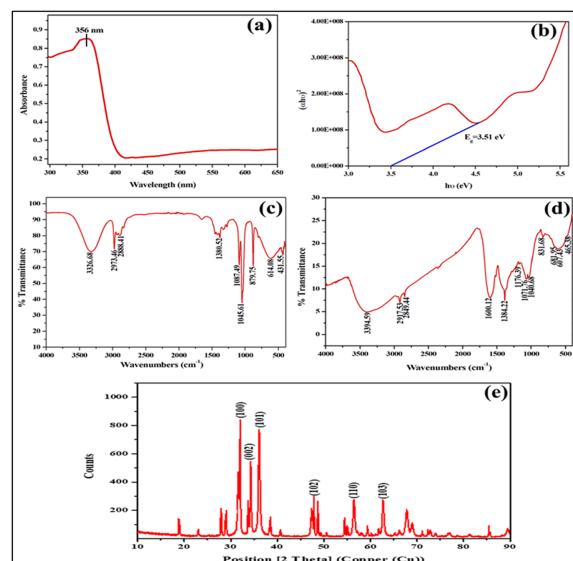


Figure 3. Characterization of synthesized ZnO nanoparticles using *Turbinaria decurrens* extract: (a) UV-Vis absorption spectrum of ZnO nanoparticles (b) Tauc plot used to determine the optical band gap energy of ZnO nanoparticles (c) FT-IR spectrum of *Turbinaria decurrens* extract (d) FT-IR spectrum of the synthesized ZnO nanoparticles indicating the characteristic functional groups and Zn-O stretching vibration and (e) X-ray diffraction (XRD) pattern showing the crystalline hexagonal wurtzite structure of the biosynthesized ZnO nanoparticles

3.2 Morphological Analysis

The morphological and structural characteristics of the synthesized ZnO nanoparticles using *Turbinaria decurrens* extract were analyzed using HR-TEM, as shown in Figure 4a,4b. The particles exhibit a rod-like shape. Clear lattice fringes with a spacing of 0.21 nm corresponding to the (101) plane confirm their crystalline nature. These results are consistent with previous reports on biosynthesized ZnO nanoparticles [33]. The SAED pattern (Figure 4c) shows concentric rings and diffraction spots, indicating the polycrystalline structure of ZnO. The observed lattice fringes and SAED pattern correspond to the (100), (002), and (101) planes of hexagonal ZnO, which are in good agreement with XRD results (JCPDS card No. 36-1451), confirming the crystallinity. The average particle diameter was found to be 25 nm, as supported by the particle size distribution histogram shown in Figure 4d. The histogram revealed that the majority of nanoparticles were distributed between 20 and 25 nm, with a relatively narrow size distribution, which is consistent with earlier reports on bio-synthesized ZnO [34].

The surface morphology of the synthesized ZnO nanoparticles using *Turbinaria decurrens* extract was

analyzed using scanning electron microscopy (SEM) analysis and the images at different magnifications as shown in Figure 4e and 4f. The SEM images of ZnO nanoparticles using *Turbinaria decurrens* extract show flake-like morphology with irregularly shaped edges.

Although HR-TEM images show rod-like ZnO nanoparticles, SEM images exhibit aggregated nanoflake-like structures. This difference is mainly due to the different imaging techniques and sample preparation methods. HR-TEM observes individual nanoparticles at high resolution, whereas SEM mainly shows the surface morphology of dried nanoparticles aggregates. During drying, the rod-like nanoparticles aggregate because of interparticle interactions, resulting in a flake-like appearance. Therefore, both HR-TEM and SEM observations are consistent and represent the same ZnO nanoparticles at different levels of aggregation.

EDAX analysis confirms the elemental composition of the synthesized ZnO nanoparticles using *Turbinaria decurrens* extract shown in Figure 4g. The EDAX examination verified that the prepared sample had a Zn and O chemical composition. The Zn peak appeared at 1 keV and O peaks appeared at 0.5 keV. The quantitative data indicated that Zn was present at a relatively higher percentage compared to O, confirming the successful synthesis of ZnO nanoparticles using *Turbinaria decurrens* extract.

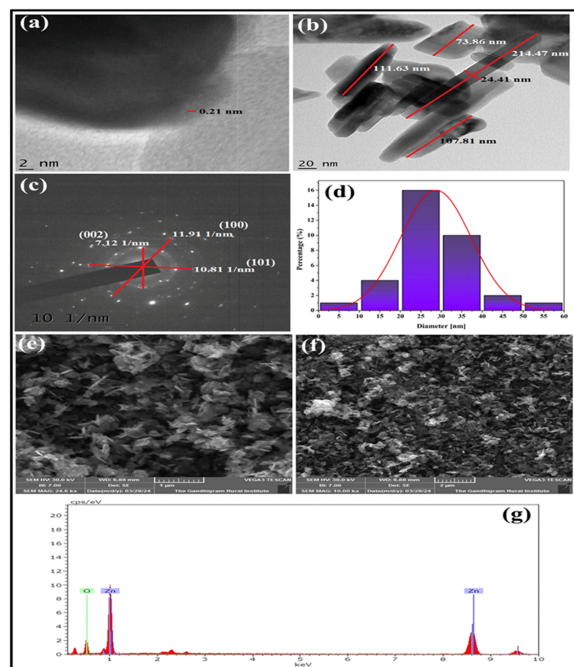


Figure 4. Morphological and elemental characterization of ZnO nanoparticles synthesized using *Turbinaria decurrens* extract: (a, b) High-resolution transmission electron microscopy (HR-

TEM) images of the synthesized ZnO nanoparticles (c) SAED pattern of the biosynthesized ZnO nanoparticles showing indexed diffraction rings corresponding to the (100), (002), and (101) crystallographic planes of hexagonal wurtzite ZnO (JCPDS Card No. 36-1451); (d) Particle size distribution histogram derived from the HR-TEM images; (e, f) SEM images of the ZnO nanoparticles at different magnifications and (g) EDAX spectrum of the synthesized ZnO nanoparticles

3.3 Cyclic Voltammetry Analysis

The electrochemical behavior of ZnO nanoparticles using *Turbinaria decurrens* extract was studied using cyclic voltammetry. A platinum foil was used as a counter electrode, an Ag/AgCl electrode as a reference electrode, and a 0.1 M KOH solution as an electrolyte [35]. Cyclic voltammetry was performed with a potential range from +0.1 V to +0.6 V at scan rates of 10-100 mV/s shown in Figure 5a. The oxidation and reduction peaks appeared around 0.42 V and 0.28 V, respectively. The nanoparticles exhibited good electrochemical activity and fast electron transfer, suggesting potential for sensing and energy applications [36].

Specific Capacitance Studies

The specific capacitance of the synthesized ZnO nanoparticles was calculated from the cyclic voltammetry (CV) curves at different scan rates. The variation of specific capacitance with scan rate is shown in Figure 5b. The specific capacitance (C_{sp}) was calculated using the following equation:

$$C_{sp} = \frac{A}{2mk(V_2 - V_1)}$$

----- (6)

where C_{sp} is the specific capacitance (F/g), A represents the area enclosed by the CV curve, m is the mass of the active material (g), k is the scan rate (mVs^{-1}), and $(V_2 - V_1)$ is the potential window.

From the graph, the maximum specific capacitance of approximately 169.49 F/g was observed at a scan rate of 10 mVs^{-1} , while the capacitance decreased to about 16.94 F/g at 100 mVs^{-1} . The decrease in capacitance with increasing scan rate is mainly attributed to the limited diffusion of electrolyte ions into the inner active sites of the electrode material. At lower scan rates, electrolyte ions have sufficient time to penetrate the electrode surface and participate in redox reactions, resulting in higher charge storage capacity. However, at higher scan rates, only the outer surface of the electrode contributes to the electrochemical process, leading to lower capacitance values. These results indicate that ZnO nanoparticles exhibit good capacitive behavior and can be considered promising materials for electrochemical energy storage applications [37].

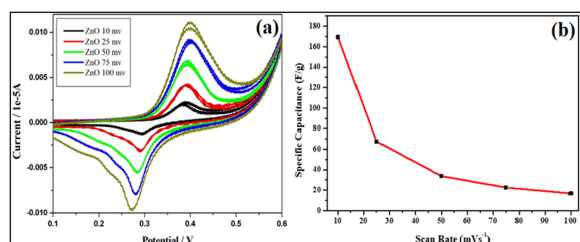


Figure 5. Electrochemical characterization of ZnO nanoparticles synthesized using *Turbinaria decurrens* extract: (a) Cyclic voltammograms recorded at different scan rates, synthesized ZnO nanoparticles and (b) Relationship between scan rate and specific capacitance ($F\ g^{-1}$), illustrating the supercapacitive performance of the ZnO nanoparticles

4. APPLICATIONS

4.1 Photocatalytic Degradation of Methylene Blue Dye Using ZnO Nanoparticles

The photocatalytic breakdown of methylene blue dye was examined via UV-Vis spectroscopy. Figure 6 a, b displays the absorption spectra for the pure methylene blue solution and ZnO nanoparticles produced with *Turbinaria decurrens* extract. The blank sample showed a prominent peak at around 660 nm, typical of the dye's maximum absorption. Absorbance stayed nearly unchanged over 0 to 3 hours of irradiation, demonstrating minimal degradation without a catalyst [38]. This verifies that light alone cannot effectively break down the dye, necessitating a semiconductor photocatalyst [39].

Conversely, adding ZnO nanoparticles to the dye solution led to a steady drop in the 660 nm peak intensity as irradiation time increased from 0 to 80 minutes. This decline in absorbance signifies successful dye degradation driven by the nanoparticles photocatalytic action [40]. The chromophoric groups in the dye are disrupted, causing decolorization and mineralization [41]. Under illumination, ZnO generates electron-hole pairs: electrons combine with oxygen to form superoxide radicals, and holes react with water to produce hydroxyl radicals. These reactive species degrade the dye into intermediates, ultimately yielding CO_2 and H_2O [42].

Thus, the dye solution without nanoparticles exhibits little degradation, while ZnO nanoparticles markedly boost efficiency, confirming their role as potent photocatalysts for removing dye pollutants from water [43].

The photocatalytic degradation efficiency of ZnO nanoparticles was determined as the percentage of dye removal using the equation:

$$\% \text{ degradation} = \frac{(C_0 - C_t)}{C_0} \times 100 \text{ -----7}$$

Here, C_0 is the initial concentration and C_t is the concentration at time t . After 80 minutes of visible light exposure, degradation reached 66% (Figure 6c), showing substantial methylene blue breakdown. To evaluate the photocatalytic performance of the present ZnO nanoparticles, the degradation efficiency was compared with previously reported synthesized ZnO photocatalysts used for methylene blue degradation under different experimental conditions. The comparison demonstrates that the photocatalytic activity of the synthesized ZnO nanoparticles is comparable with several biologically synthesized ZnO photocatalysts reported in the literature.

Table 2. Comparison of photocatalytic degradation efficiency of synthesized ZnO nanoparticles for methylene blue (MB) dye degradation reported in recent studies

Biological Source	Particle Size (nm)	Band Gap (eV)	Photocatalytic degradation efficiency (%)	Irradiation Time (min)	References
<i>Sargassum muticum</i>	15-50	3.28	96	60	[44]
<i>Padina pavonica</i>	35.89	Not reported	79	180	[45]
<i>Ulva Lactuca</i>	10-50	Not reported	90	120	[46]
<i>Secamone emetica</i>	50.19	2.98	93.57	210	[47]
<i>mango flower</i>	30.54	3.13	98.36	70	[48]
<i>E. prostrata</i>	14-58	3.10	95.6	120	[49]

leaves					
<i>Turbinaria decurrens</i>	46.1	3.51	66	80	Present Study

As shown in Table 2, the photocatalytic degradation efficiency of the ZnO nanoparticles synthesized using *Turbinaria decurrens* extract is comparable with those of other biologically synthesized ZnO nanoparticles reported in recent studies. Despite these experimental differences, the present ZnO nanoparticles achieved 66% degradation of methylene blue within 80 min under visible light irradiation, demonstrating their potential as an environmentally friendly photocatalyst for wastewater treatment. Although the photocatalytic degradation efficiency (66%) obtained in the present study is near than that reported for certain biosynthesized ZnO photocatalysts, the degradation efficiency is strongly influenced by several experimental parameters, including catalyst dosage, initial dye concentration, irradiation source, light intensity, crystallite size, particle morphology, and the surface phytochemicals involved in nanoparticle stabilization. In the present study, the phytochemical molecules derived from *Turbinaria decurrens* may partially cover the active surface of the ZnO nanoparticles, thereby reducing the number of accessible photocatalytic active sites. Nevertheless, the biosynthesized ZnO nanoparticles exhibited appreciable photocatalytic activity under environmentally benign synthesis conditions without the use of hazardous reducing or stabilizing agents. Therefore, the present material represents a sustainable and eco-friendly photocatalyst with potential applications in wastewater treatment [50].

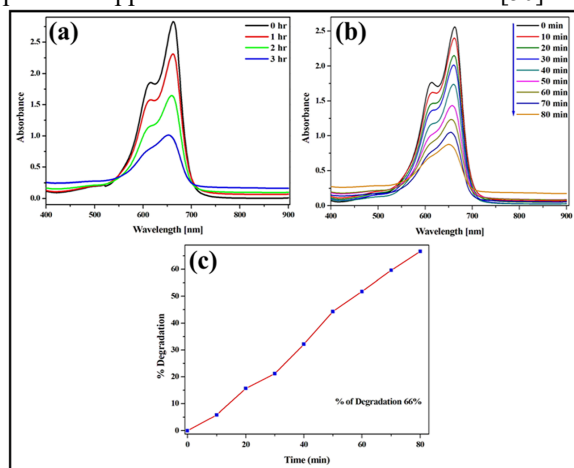
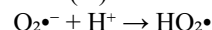
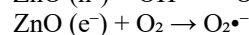
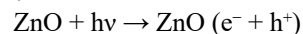


Figure 6. Photocatalytic degradation of methylene blue (MB) using ZnO nanoparticles synthesized

from *Turbinaria decurrens* extract under sunlight irradiation: (a) UV-Vis spectrum of blank methylene blue solution; (b) UV-Vis degradation spectra of methylene blue in the presence of ZnO nanoparticles at different irradiation times; and (c) Photocatalytic degradation efficiency (%) of methylene blue as a function of irradiation time

4.1.1 Mechanism of Photocatalytic Degradation

Under visible light, ZnO nanoparticles absorb photons, exciting electrons from the valence band to the conduction band, creating electron-hole pairs. These pairs reach the nanoparticles surface for redox reactions. Holes react with OH^- or H_2O to form hydroxyl radicals ($\bullet\text{OH}$), and electrons reduce O_2 to superoxide radicals ($\text{O}_2^{\bullet-}$). These oxygen species drive oxidative degradation. The mechanism is depicted below [51], with the full pathway in Figure 7.



In summary, reactive oxygen species, especially $\bullet\text{OH}$, oxidize methylene blue into intermediates and then mineralize it to CO_2 and H_2O . This underscores the excellent photocatalytic performance of ZnO nanoparticles for wastewater treatment and remediation.

Although radical scavenger experiments and photoluminescence (PL) analysis were not performed in the present study, the proposed photocatalytic mechanism is supported by previous reports describing ROS-mediated degradation pathways in biosynthesized ZnO nanoparticles.

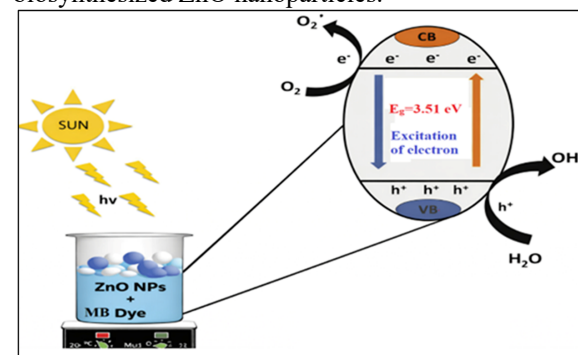


Figure 7. Mechanism for the photocatalytic degradation of methylene blue (MB) dye by ZnO nanoparticles synthesized using *Turbinaria decurrens* extract under visible light irradiation

4.1.2 Kinetic Analysis of Methylene Blue Degradation

The degradation kinetics of methylene blue with *Turbinaria decurrens* extract-synthesized ZnO

nanoparticles were assessed via pseudo-zero-order and pseudo-first-order models. Kinetic plots from data are in Figures 8a, 8b.

Pseudo-Zero-Order Kinetic Model

Degradation followed a pseudo-zero-order model, where rate is concentration-independent [52]:

$$C_0 - C_t = k_0 t \quad \text{----- (8)}$$

C_0 is the initial methylene blue concentration, C_t is at time t , and k_0 is the rate constant. Plotting $C_0 - C_t$ vs. time (Figure 8a) showed linearity, suggesting partial zero-order kinetics. The slope gave $k_0 = 0.0232 \text{ min}^{-1}$. Minor deviations indicate influence of dye concentration, common in heterogeneous photocatalysis due to surface adsorption [53].

Pseudo-First-Order Kinetic Model

This model, suitable for concentration-dependent heterogeneous reactions [54], is

$$\ln\left(\frac{C_0}{C_t}\right) = kt \quad \text{----- (9)}$$

The $\ln(C_0/C_t)$ vs. time plot (Figure 8b) was linear ($R^2 = 0.98768$), with $k = 0.01722 \text{ min}^{-1}$. This fit aligns with the Langmuir-Hinshelwood mechanism, involving dye adsorption followed by ROS-mediated degradation [55, 56].

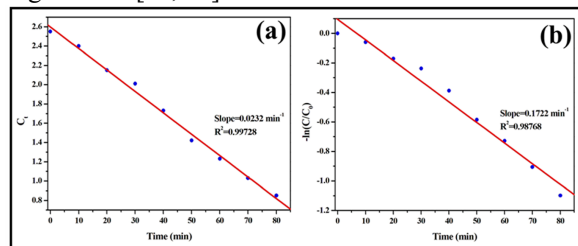


Figure 8. Kinetic analysis of the photocatalytic degradation of methylene blue (MB) using ZnO nanoparticles synthesized from *Turbinaria decurrens* extract: (a) Pseudo-zero-order kinetic plot and (b) pseudo-first-order kinetic plot

4.2 Evaluation of Antibacterial Activity of ZnO nanoparticles

Antibacterial activity of *Turbinaria decurrens* extract and ZnO nanoparticles was tested against five pathogenic bacteria via agar well diffusion. Inhibition zones at various concentrations are in Table 3, with plates in Figure 10a, b. Both showed activity against all strains, consistent with prior reports on biosynthesized ZnO nanoparticles [57].

ZnO nanoparticles outperformed the extract, especially at high concentrations, with 16 mm zones against *Staphylococcus aureus* vs. 7-10 mm for the extract. This enhancement arises from nanoparticles small size, large surface area, cell wall penetration, ROS generation ($\bullet\text{OH}$, H_2O_2), oxidative damage to proteins/lipids/DNA, and Zn^{2+} ion release disrupting metabolism. Activity rises with concentration due to more interactions and ion release [58-60]. Although ethanol was used during the preparation of algae

extract, it was completely removed during the drying and calcination steps. Therefore, the observed antibacterial activity is attributed to the synthesized ZnO NPs rather than residual ethanol. However, inclusion of an ethanol-treated negative control would provide additional validation and will be considered in future studies.

The antibacterial mechanism of biosynthesized ZnO nanoparticles is illustrated schematically in Figure 9. As shown in Figure 9, the antibacterial activity is mediated through a multi-faceted mechanism involving reactive oxygen species (ROS) generation, Zn^{2+} ion release, membrane disruption, oxidative stress, intracellular biomolecular damage, and ultimately bacterial cell death. Upon exposure to bacterial cells, ZnO nanoparticles initially adhere to the negatively charged cell surface through electrostatic interactions, leading to structural alterations in the cell wall and increased membrane permeability. This direct physical interaction facilitates nanoparticles internalization and promotes the dissolution-mediated release of Zn^{2+} ions into the cytoplasmic environment [61].

Subsequently, ZnO nanoparticles function as semiconductor materials that generate electron-hole pairs under ambient or UV illumination, which subsequently drive redox reactions with dissolved oxygen and water molecules to produce ROS, including superoxide anions ($\text{O}_2^{\bullet-}$), hydroxyl radicals ($\bullet\text{OH}$), and hydrogen peroxide (H_2O_2). These highly reactive species induce severe oxidative stress within bacterial cells, resulting in lipid peroxidation of the cell membrane, denaturation of essential proteins, inhibition of respiratory enzymes, and damage to genomic DNA. The released Zn^{2+} ions further potentiate toxicity by binding to thiol ($-\text{SH}$) groups of critical bacterial enzymes, particularly those involved in the electron transport chain, thereby disrupting metabolic processes and amplifying intracellular ROS production [62].

The synergistic effects of membrane damage, ion toxicity, and ROS-mediated oxidative stress culminate in loss of membrane integrity, leakage of intracellular contents (e.g., K^+ ions, proteins and nucleic acids), inhibition of cell division, and ultimately bacterial cell death [63]. This multi-modal mechanism explains the broad-spectrum antibacterial efficacy of ZnO nanoparticles against both Gram-positive and Gram-negative bacterial strains, as observed in the present study.

Overall, the excellent antibacterial performance of biosynthesized ZnO nanoparticles can be attributed to the synergistic action of ROS generation, Zn^{2+} ion release, membrane disruption, oxidative stress, and intracellular biomolecular damage, which collectively result in efficient bacterial cell death. These findings

support the potential application of the synthesized ZnO nanoparticles in antimicrobial and biomedical fields.

Table 3. Antibacterial activity of *Turbinaria decurrens* extract and biosynthesized ZnO nanoparticles against selected pathogenic bacterial strains, expressed as the zone of inhibition (mm)

Bacteria	Inhibition zone in mm											
	Abciprofloxaacin		200 µg/ml		400 µg/ml		600 µg/ml		800 µg/ml		1000 µg/ml	
	T	Z	T	Z	T	Z	T	Z	T	Z	T	Z
	D	n	D	n	D	n	D	n	D	n	D	n
	O	O	O	O	O	O	O	O	O	O	O	O
<i>Escherichia coli</i>	2	2	5	1	5	1	5	1	5	1	7	1
	1	6		0		1		1		1		3
<i>Staphylococcus aureus</i>	2	2	7	7	5	9	6	1	7	1	7	1
	0	5						0		0		6
<i>Bacillus subtilis</i>	2	2	4	-	6	-	6	1	7	1	9	1
	2	8						0		2		3
<i>Bacillus cereus</i>	2	2	8	9	9	9	9	1	9	1	1	1
	4	9						0		0	0	2
<i>Pseudomonas aeruginosa</i>	2	3	6	1	7	1	7	1	8	1	9	1
	1	0		0		0		0		1		4

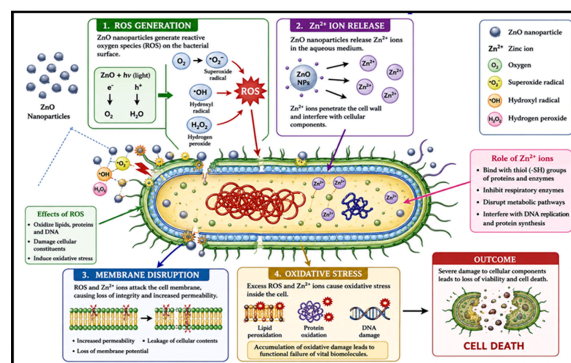


Figure 9. Schematic illustration of the antibacterial mechanism of biosynthesized ZnO nanoparticles, showing bacterial cell adhesion, ROS generation, Zn²⁺ ion release, membrane disruption, oxidative stress, intracellular biomolecule damage, leakage of cellular contents, and bacterial cell death

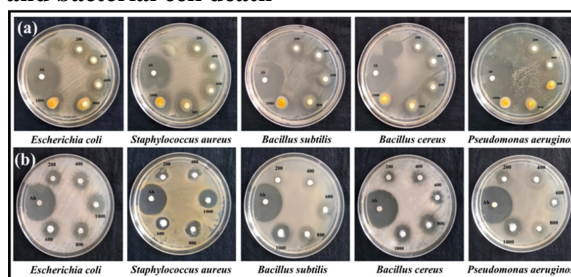


Figure 10. Antibacterial activity against selected pathogenic bacterial strains determined by the agar well diffusion method: (a) *Turbinaria decurrens* extract and (b) ZnO nanoparticles synthesized using *Turbinaria decurrens* extract

4.3 Assessment of the Antioxidant Potential of ZnO nanoparticles by DPPH Radical Scavenging Assay

The antioxidant properties of the *Turbinaria decurrens* aqueous extract and bio-synthesized ZnO nanoparticles were assessed via the DPPH free radical scavenging assay (Figure 11a), with results shown in Table 4. Inhibition rates rose progressively with concentration, reflecting dose-dependent antioxidant effects. At 25 µg/mL, the extract displayed 7.16 % inhibition, the nanoparticles 10.96 % and the standard ascorbic acid 13.18 %. Such concentration-responsive antioxidant behavior aligns with prior reports on marine algae-mediated nanoparticles synthesis [64].

From 25 to 125 µg/mL, antioxidant performance markedly improved across all samples. At 125 µg/mL, the extract reached 50.42 % inhibition; nanoparticles, 60.28 % and ascorbic acid, the highest at 66.48 %. These findings highlight the superior radical-scavenging potential of the ZnO nanoparticles over the raw extract. Literature corroborates that biosynthesized metal oxide nanoparticles from

marine algae benefit from synergistic bioactive compounds in the algal extract, enhancing their antioxidant prowess [65].

The IC_{50} value (Figure 11b), denoting the concentration for 50% radical inhibition, gauges antioxidant potency. Here, the extract had an IC_{50} of 91.76 $\mu\text{g/mL}$, nanoparticles 80.54 $\mu\text{g/mL}$, and ascorbic acid 78.18 $\mu\text{g/mL}$. Lower IC_{50} signifies greater efficacy, confirming the nanoparticles boosted scavenging ability versus the extract, consistent with studies attributing this to nanoscale characteristics [66].

The nanoparticles superior activity stems from phytochemicals like phenolics, flavonoids, and terpenoids in *Turbinaria decurrens*, which stabilize particles during synthesis and donate electrons or hydrogens to quench reactive oxygen species. Marine macroalgae are renowned for abundant antioxidants that shield against oxidative damage [67].

Additionally, the nanoparticles nanoscale dimensions and high surface area facilitate stronger radical interactions via elevated surface-to-volume ratios, promoting efficient electron transfer. Recent studies affirm that bio-derived ZnO nanoparticles in antioxidant and biomedical roles, positioning them for therapeutic use [68]. Thus, these *Turbinaria decurrens*-derived ZnO nanoparticles offer substantial antioxidant promise for biomedical applications.

Table 4. DPPH free radical scavenging activity of *Turbinaria decurrens* aqueous extract and biosynthesized ZnO nanoparticles at different concentrations, compared with the standard antioxidant (ascorbic acid)

Concentration $\mu\text{g/mL}$	TD aqueous extract (%)	ZnO (%)	Ascorbic acid (%)
25	7.16	10.96	13.18
50	15.28	22.30	28.26
75	28.36	35.46	39.24
100	35.18	47.24	59.16
125	50.42	60.28	66.48
IC_{50}	91.76	80.54	78.18

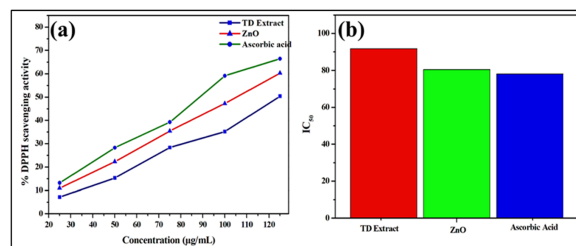


Figure 11. DPPH free radical scavenging activity of *Turbinaria decurrens* aqueous extract and biosynthesized ZnO nanoparticles: (a) concentration-dependent scavenging activity compared with ascorbic acid and (b) comparison of IC_{50} values of the aqueous extract, ZnO nanoparticles and ascorbic acid

CONCLUSION

This study successfully biosynthesized ZnO nanoparticles using *Turbinaria decurrens* brown algal extract. Comprehensive characterization via UV-Vis, FT-IR, XRD, TEM, SEM, and EDAX confirmed their optical, functional, crystalline, morphological, and compositional attributes, with a suitable bandgap indicating photocatalytic promise. The nanoparticles efficiently degraded methylene blue dye and displayed strong electrochemical performance in cyclic voltammetry. They also outperformed the crude extract in antibacterial and antioxidant assays. This green, economical method yields multifunctional ZnO nanoparticles for environmental cleanup, antibacterials, and biomedicine. Future work could optimize synthesis, refine sizes, and probe advanced catalytic biomedical uses. The present study demonstrates that *Turbinaria decurrens* using ZnO nanoparticles represents a sustainable multifunctional nanomaterial with promising photocatalytic, antibacterial, antioxidant, and electrochemical properties.

Acknowledgements

The authors sincerely acknowledge the PG and Research Department of Chemistry, A.P.C. Mahalaxmi College for Women, Thoothukudi, Tamil Nadu, India, for providing laboratory support for the synthesis of the nanoparticles. The authors are extremely grateful to the PG & Research Department of Chemistry, V.O. Chidambaram College, Thoothukudi, Tamil Nadu, India, for providing the Jasco V-530 UV-Vis spectrophotometer and FT-IR spectra. We are thankful to the secretary of A.P.C. Mahalaxmi College for Women, Thoothukudi, Tamil Nadu, India.

Disclosure statement

The authors declare no conflicts of interest regarding the publication of this paper.

Data availability statement

The authors confirm that the data supporting the findings of this study are available within the article.

Additional information

Funding: The authors declare that no funding was received from any organization or agency in support of this research.

Author Contributions

M. R. - Writing – original draft, D. S. - Methodology, T. A. - Formal analysis, S. K. - Reviewing original draft, C.V. - Supervision, Visualization. All the authors contributed to the manuscript and approved the final version.

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