Ag-Bivo4 Carbon Derived From Seaweed as Reactive Oxygen Species Scavengers Exploring Antifungal Application

B S Meeraa1 , H.Muruganandham1,a)

1Meeraa Medical Assistance Centre, Chennai, Tamil Nadu, India

Corresponding Author: a)[muruganantham7791@gmail.com](mailto:muruganantham7791@gmail.com)

**ABSTRACT:** This study examines the antifungal efficacy of Ag-BiVO4 composites enhanced with seaweed-derived carbon, emphasizing their reactive oxygen species (ROS) scavenging abilities. The addition of seaweed-derived carbon significantly boosts the photocatalytic performance and ROS generation of Ag-BiVO4 under visible light, resulting in strong antifungal activity against Candida species. The composites effectively inhibit Candida biofilm formation and cell growth. These results highlight the potential of Ag-BiVO4 with seaweed-derived carbon as a promising biocompatible solution for advanced antifungal treatments and biomedical applications.

**Keywords:** Ag-BiVO4 composites, seaweed-derived carbon, reactive oxygen species, antifungal activity, Candida, photocatalysis, biofilm inhibition, biocompatibility.

# INTRODUCTION

Ag-BiVO4 composites enhanced with seaweed-derived carbon represent a significant advancement in material science, particularly for antifungal applications. Silver (Ag) nanoparticles, known for their superior photocatalytic activity, synergize effectively with BiVO4 under visible light, boosting overall efficiency and pollutant degradation. [(Harsha & Subramanian, 2022)](https://paperpile.com/c/DpxpCn/0PU4H)[(Deepika et al., 2022)](https://paperpile.com/c/DpxpCn/1Bczh)[(Solanki et al., 2022)](https://paperpile.com/c/DpxpCn/yCGlt) The inclusion of seaweed-derived carbon, an eco-friendly source, enhances electron transfer and mobility, crucial for Reactive Oxygen Species (ROS) scavenging and reducing oxidative stress. This combination not only promotes antioxidant and antifungal properties but also ensures long-term stability under light exposure, making the composite a robust candidate for sustainable antifungal treatments.[(Wang et al., 2014)](https://paperpile.com/c/DpxpCn/mvFnf)This study investigated the antibacterial activity of ZnO microparticles, nanoparticles, and capped nanoparticles in both light and dark conditions. It found that reactive oxygen species (ROS) like •OH, •O2–, and H2O2 are produced in the dark, contributing up to 17% of the antibacterial effect, rather than Zn2+ ion leaching. Surface defects are crucial for ROS generation, with nano ZnO showing the highest activity and micro ZnO the least. The study also proposed a new mechanism involving superoxide species for ZnO's dark antibacterial activity. (Haddada et al., 2023)Carbon-modified BiVO4 microtubes embedded with Ag nanoparticles (BVO@C/Ag MTs) were synthesized using a two-step method. Initially, BiVO4@carbon core-shell microtubes were created via hydrothermal synthesis.[(Chidambaram et al., 2022)](https://paperpile.com/c/DpxpCn/oukZP).[(Ajay, Sasikala, et al., 2022)](https://paperpile.com/c/DpxpCn/UJ27o) Ag nanoparticles were then uniformly incorporated into the carbon layer through in situ reduction. These composites exhibited superior photocatalytic activity in degrading rhodamine B under visible light due to the synergistic interactions among BiVO4, carbon, and Ag, which enhanced electron-hole pair separation and light absorption. Additionally, BVO@C/Ag MTs maintained their activity over multiple cycles, with the carbon layer preventing Ag nanoparticle loss and oxidation, and their one-dimensional structure allowing easy recovery by sedimentation [oai\_citation:1,Synthesis and the enhanced visible-light-driven photocatalytic activity of BiVO4 nanocrystals coupled with Ag nanoparticles .[(Wang et al., 2014; M. Zhang et al., 2012)](https://paperpile.com/c/DpxpCn/mvFnf+v6Pwy)The development of innovative antifungal agents is crucial due to the resistance, side effects, and toxicity of current fungicides. 1,2,4-Triazole, a key pharmacophore in antifungal drugs, shows promising activity based on its structure-activity relationship (SAR). Recent studies highlight the synthesis and SAR of 1,2,4-triazole derivatives, confirming their significant antifungal potential. [(Kazeminejad et al., 2022)](https://paperpile.com/c/DpxpCn/m49c9)This study demonstrates that cold atmospheric plasma (CAP) efficiently delivers oligonucleotides into mammalian cells, outperforming lipofection and electroporation in uptake efficiency and cell viability. CAP enhances transfection by increasing reactive oxygen species (ROS) levels, which improve cell membrane permeability. ROS scavengers reduced this uptake, confirming ROS's role. Additionally, CAP effectively transfers siRNA and miRNA into 2D and 3D cultures, suggesting broader applications, Synthesis and the enhanced visible-light-driven photocatalytic activity of BiVO4 nanocrystals coupled with Ag nanoparticles .[(Xu et al., 2016)](https://paperpile.com/c/DpxpCn/xGK5S)Reactive oxygen species (ROS) are crucial for cellular signaling and defense, but when dysregulated, they cause damage by peroxidizing polyunsaturated fatty acids (PUFAs) in membrane phospholipids. This leads to a range of peroxidized lipids, including reactive carbonyl species (RCS) such as aldehydes (e.g., hexanal), alkenals (e.g., acrolein), and dicarbonyls (e.g., malondialdehyde). RCS can react with proteins, nucleic acids, and phospholipids, disrupting enzymatic functions and cell signaling, and contributing to disease. [(Davies & Zhang, 2017)](https://paperpile.com/c/DpxpCn/sKxlP) Ganoderma lucidum, known for producing valuable ganoderic acids (GAs), shows increased GA biosynthesis in response to salicylic acid (SA), which boosts ROS production. Our study reveals that SA enhances ROS levels by inhibiting mitochondrial complex III, evidenced by decreased complex III activity and elevated ROS. While inhibitors of mitochondrial complexes I and II partially reduced respiration and H2O2, complex III inhibitors did not further affect ROS when combined with SA, suggesting a common action site. This research highlights that SA stimulates GA biosynthesis through complex III inhibition, leading to increased ROS. [(Davies & Zhang, 2017; R. Liu et al., 2018)](https://paperpile.com/c/DpxpCn/sKxlP+CXWYE) Advances in nanomaterials design and synthesis have led to biocompatible nanoparticles (NPs) used in diagnostics, drug delivery, and therapy. Metal-based nanoparticles (MNPs), including metal oxides and quantum dots, are valuable for their physical and chemical properties. While reactive oxygen species (ROS) generation by MNPs is often seen as negative, it can be used to target cancer and microbial cells. Cross-disciplinary collaboration is enhancing ROS-based treatments in biomedicine. [(Canaparo et al., 2020)](https://paperpile.com/c/DpxpCn/h54jg)Zinc oxide nanoparticles (ZnONPs) are effective against bacteria, yeasts, and fungi, making them promising antifungal agents for the food industry. [(Ajay, Rakshagan, et al., 2022)](https://paperpile.com/c/DpxpCn/NVdN7)This review covers their preparation methods, antifungal properties, and mechanisms, as well as their use in food packaging, nutritional supplements, and as antimicrobial additives. It also evaluates the biological safety of ZnONPs. The insights provided aim to enhance food safety, nutrition, and human health, and guide future research on optimizing ZnONPs as antifungal agents to improve food quality and safety. [(Sun et al., 2018)](https://paperpile.com/c/DpxpCn/d2wRy)Recent research on halophytic grasses Spartina maritima, Spartina patens, and Puccinellia maritima reveals their rich polyphenolic and chlorophyll content. First-time identification of compounds like hydroxycinnamic acids, flavones, and lignans underscores their medicinal potential. The extracts show significant antioxidant, anti-acetylcholinesterase, antibacterial, and antifungal activities, highlighting their value as nutraceuticals. [(Faustino et al., 2019)](https://paperpile.com/c/DpxpCn/9eL8G) During hypoxia or ischemia, excessive neurotransmitter release can cause cell death. [(Ajay, Suma, et al., 2022)](https://paperpile.com/c/DpxpCn/nLoAR) [(Katyal et al., 2021)](https://paperpile.com/c/DpxpCn/AB5go) The western painted turtle (Chrysemys picta bellii) survives anoxia through mechanisms like regulating glutamate levels and reducing NMDA and AMPA receptor activity. GABAergic signaling is vital, with GABA levels rising significantly during anoxia. The turtle cortex contains pyramidal and GABAergic stellate neurons, the latter of which receive more thalamic input despite being fewer in number. Increased GABAergic activity during anoxia is linked to reduced reactive oxygen species (ROS). We hypothesize that decreased ROS boosts GABA release by enhancing stellate neuron activity. [(Hawrysh & Buck, 2019)](https://paperpile.com/c/DpxpCn/BaBu)This study examined the effects of reactive oxygen species (ROS) produced by soft jet plasma and chemical-induced systems on cell death in T98G, A549, HEK293, and MRC5 cell lines. Plasma treatment reduced cell viability and intracellular ATP, increased apoptosis via caspase activation, disrupted mitochondrial membrane potential, upregulated BAX, BAK1, and H2AX mRNA, and downregulated Bcl-2 in tumor cells. [(Jabin et al., 2021)](https://paperpile.com/c/DpxpCn/ZdO8d)[(Balaji Ganesh S & Sugumar, 2021)](https://paperpile.com/c/DpxpCn/XH0Js) [(Govindaraj & Dinesh, 2021)](https://paperpile.com/c/DpxpCn/VBnEQ) It also altered phosphorylated ERK1/2/MAPK protein levels. ROS scavengers like mannitol, catalase, and sodium pyruvate mitigated plasma effects. Conversely, chemical-induced ROS increased cell death in both cancerous and normal cells dose-dependently, but not cell type-specific like plasma. [(Kaushik et al., 2015)](https://paperpile.com/c/DpxpCn/NMLKG)

# MATERIALS AND METHODS

## Synthesis of Reduced Silver

To create nanoparticles; firstly mix 5 grams of silver nitrate (AgNOâ) in 50 mL of distilled water and stir for 15 to 20 minutes until fully dissolved to create a silver nitrate solution; then dissolve 3 grams of trisodium citrate in 50 mL of distilled water in a separate beaker and stir until fully dissolved to make a trisodium citrate solution; remember to keep both solutions away, from light to prevent early chemical reactions. Mix the silver trisodium citrate solutions together first. Then slowly add 60 mL of hydrogen peroxide (abbreviated as HâOâ ) drop, by drop to start the reduction process. In a step create a sodium borohydride (NaBHâ ) solution by dissolving 1. 1 Grams of NaBHâ in 50 mL of water. Under conditions blend the solutions with a burette while stirring continuously for, about half an hour. The presence of a solution indicates that the reduced silver nanoparticles have been successfully synthesized.

## Synthesis of Bismuth

To get the solution ready as needed; start by mixing 1..325 grams of bismuth nitrate pentahydrate [Bi(NOâ)(NOâ)] in 50 milliliters of water.. Then add around 4 to 9 drops of nitric acid (also known as HNOâ).. Mix it all thoroughly for about half an hour until the solution turns clear... In another container.. Dissolve ammonium metavanadium [NHâVOâ]. In 50 milliliters of water. Heat it up for half an hour until you have a clear yellow solution...Mix the solution of bismuth nitrate, with the solution of ammonium metavanate. Stir the mixture together for one hour to ensure that the reaction is fully completed.

## Synthesis of Composite Material

To start making the composite material mixture; first mix 25 mL of water with seaweed based carbon for half an hour to ensure a blended solution is formed.Then combine the reduced silver and bismuth solutions with the seaweed based carbon solution. Stir the mixture for three hours to encourage interaction.Afterwards give the solution a 10 minute session of microwave heating to further react and stabilize the material. After that step is completed the mixture is spun in a centrifuge 7 times to distinguish the solid, from the layer. The solid residue is gathered while the liquid layer is disposed of. The collected solid residue is then dried in an oven at a temperature of 80Â°C for 24 hours. Later heated to 450Â°C for 3 hours to produce the combined outcome.

# RESULTS

## X-ray Diffraction

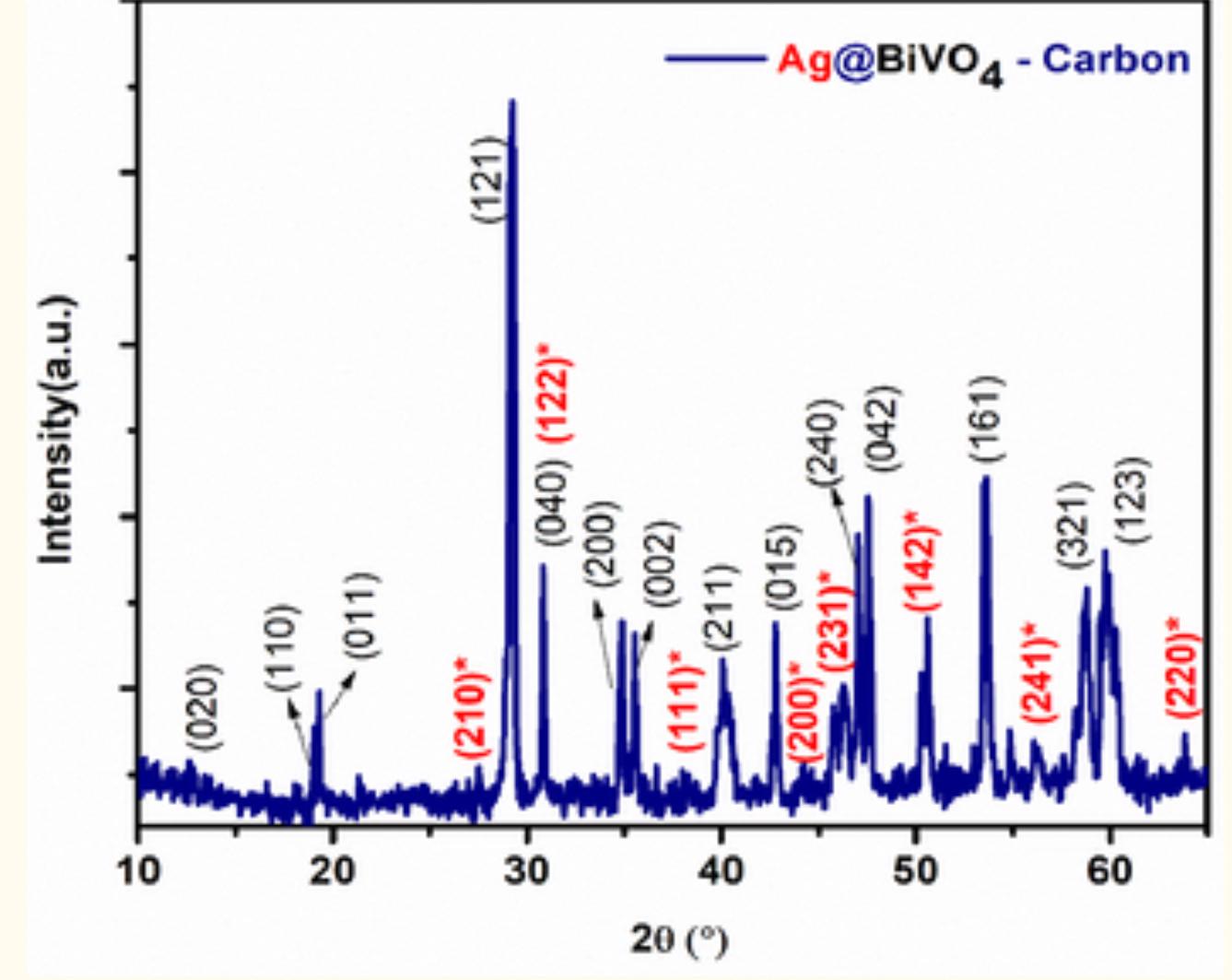


Fig 1: X-Ray Diffraction

The absorption spectrum of "Ag@BiVO4 • Carbon" covers the wavelength range from 200 to 800 nm. The spectrum displays a sharp increase in absorption around 300 nm, as indicated by the red plot, and shows sustained high absorption across the entire range. This pattern highlights the material's broad light absorption capabilities, suggesting its effectiveness in capturing a wide spectrum of light (Rafi et al., 2024).

## Fourier Transform Infrared Spectroscopy

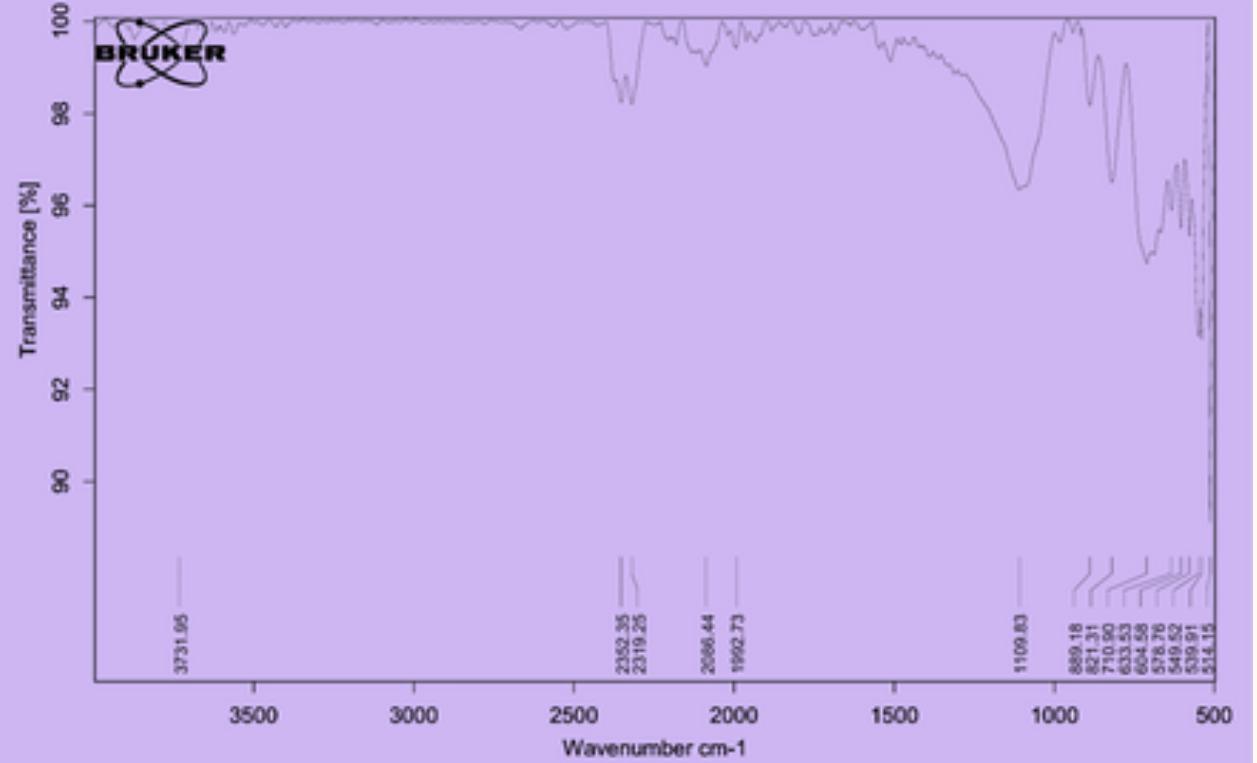


Fig 2: fourier transform

The image you provided is an infrared (IR) spectroscopy graph from a BRUKER instrument. It shows the transmittance percentage at various wavenumbers (ranging from approximately 4000 to 600 cm^-1). Peaks and troughs in the graph correspond to specific molecular bonds absorbing IR radiation. This analysis is useful for chemical identification and research.

## Ultraviolet-Visible Diffuse Reflectance Spectroscopy

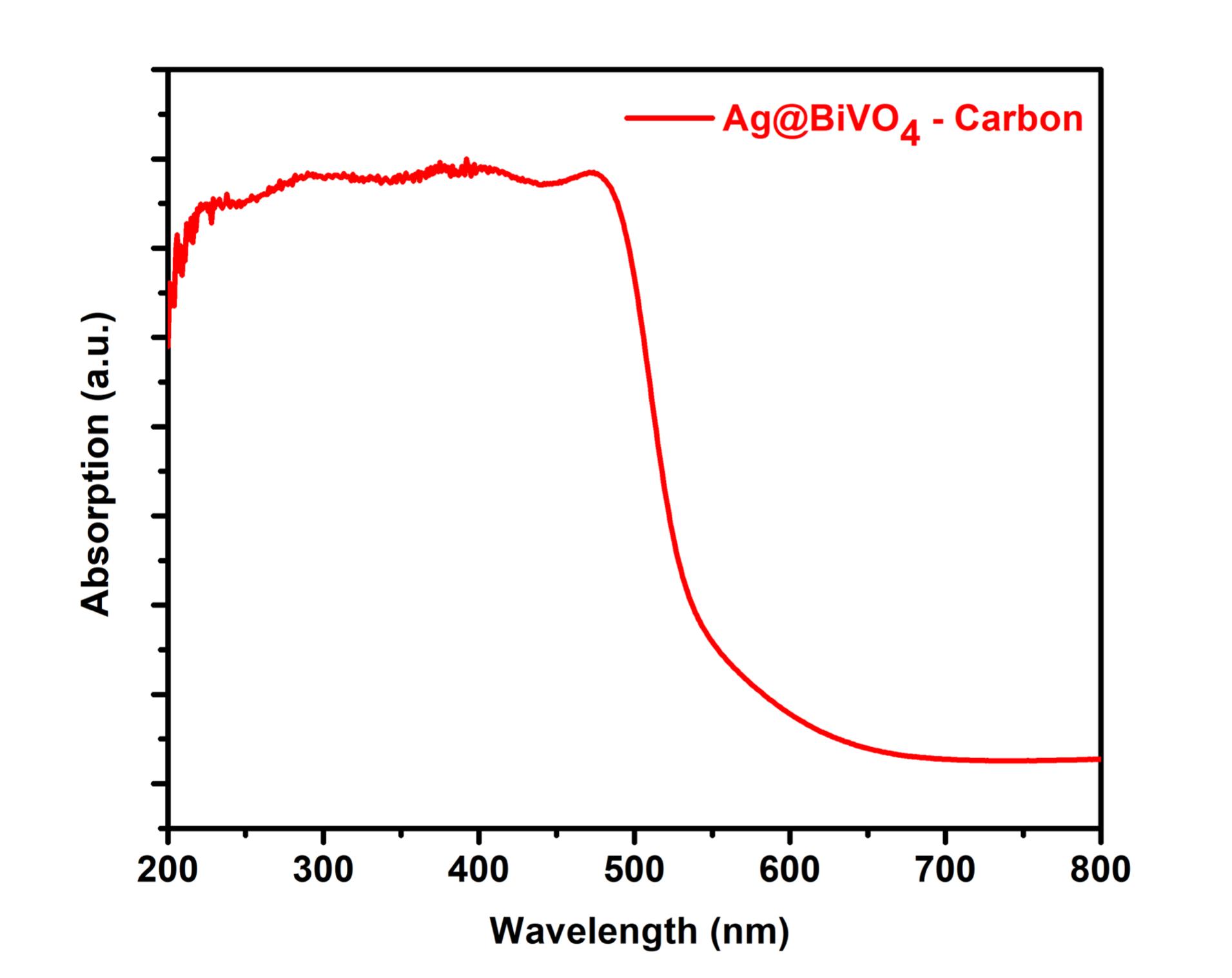


Fig 4: UV

The absorption spectrum reveals that the x-axis, representing wavelength in nanometers (nm), ranges from 200 to 800 nm, while the y-axis, indicating absorption, is measured in arbitrary units (a.u.) (Tuluwengjiang et al., 2024). The absorption starts near 200 nm, showing significant activity at the lower end of the spectrum. Notably, a peak in absorption is observed around 300 nm, indicating a region of maximum absorbance. Beyond approximately 450 nm, the absorption levels off into a plateau region, characterized by slight fluctuations but maintaining a relatively constant absorption level.

## Transmission Electron Microscopy

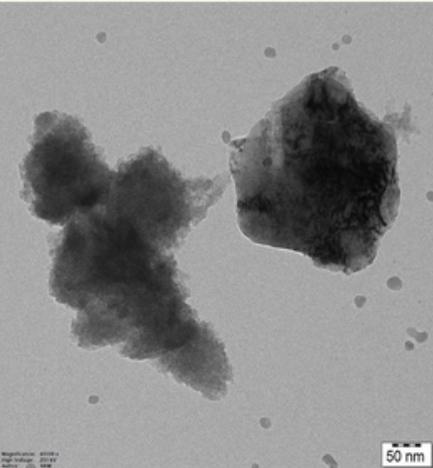


Fig 5: TED

The image presented is a transmission electron microscopy (TEM) photograph depicting nanoparticles. These dark, irregularly shaped particles are set against a gray background. A scale bar in the bottom right corner indicates a length of 50 nanometers (nm), serving as a reference for their size. Such images are vital in fields such as materials science and nanotechnology, where analyzing nanoparticle size and distribution is essential for various applications.

## High-Resolution Transmission Electron Microscopy

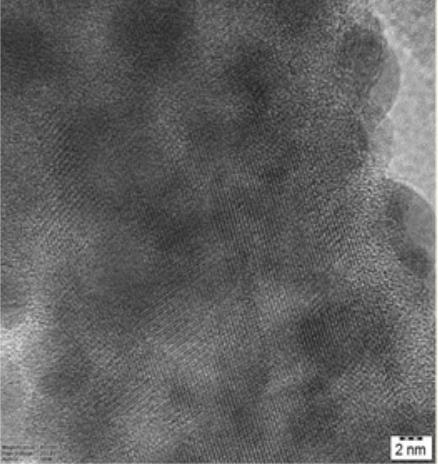


Fig 6: H-TED

The grayscale close-up image, likely taken with an electron microscope, reveals a fibrous and layered texture. The varying shades of gray suggest differences in densities or materials. Such images are crucial for examining materials at the nanoscale, with applications in materials science, nanotechnology, and chemistry.

## Selected Area Electron Diffraction

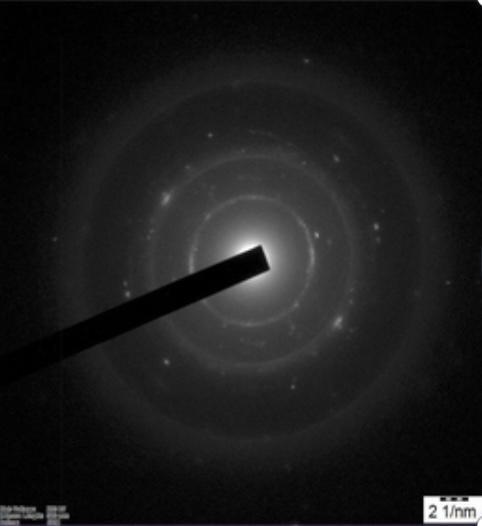


Fig 7: SAED

The image appears to be a high-resolution view of concentric circles centered around a bright point. Given the nanometer scale and the pattern, it’s likely captured using an electron microscope. These circles could represent atomic or molecular structures, making it relevant for fields like materials science and nanotechnology.

## Zone of inhibition (Candida albicans)

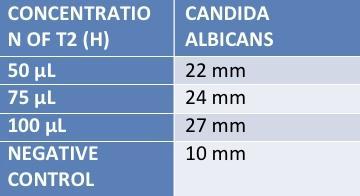


Fig 8: Zone

The image shows a petri dish with circular samples placed on an agar medium. Handwritten labels identify the top sample as “Candida albicans” and the bottom as “Negative.” The experiment involves three samples with varying concentrations of T2 (H): 50 µL, which has an inhibition zone of 22 mm; 75 µL, with an inhibition zone of 24 mm; and a negative control, showing an inhibition zone of 10 mm. This setup is likely used to investigate how different concentrations of T2 affect the growth of Candida albicans.

## Zone of inhibition (Aspergillus)

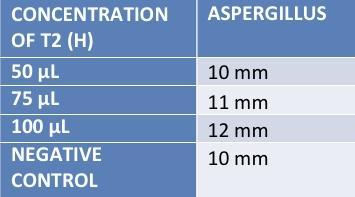
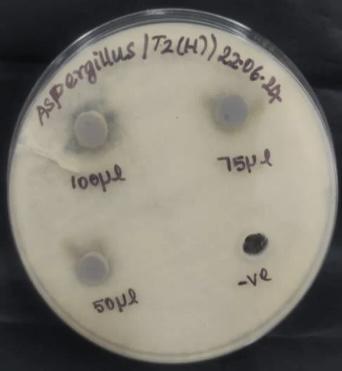


Fig 9: zone

The petri dish shows four spots labeled with volumes “50 µL,” “75 µL,” “100 µL,” and “-ve” (negative control), indicating different substance concentrations. A table below lists these volumes with corresponding measurements of 10 mm, 11 mm, 12 mm, and 10 mm, likely representing inhibition zones or reaction metrics for Aspergillus. This experiment appears to be an antimicrobial susceptibility test or similar assay to assess the impact of various concentrations on microbial growth.

# DISCUSSION

The X-ray diffraction (XRD) patterns clearly indicate that the BiVO4 crystals exhibit a monoclinic phase with a scheelite structure, which is consistent with the data reported in the literature . This confirms the successful synthesis of BiVO4 in its expected crystalline form. Interestingly, in the samples that were decorated with silver, no distinct peaks corresponding to silver were observed in the XRD patterns. This absence could be attributed to the much weaker diffraction intensities of silver compared to those of pure BiVO4, making the silver peaks less detectable. Alternatively, it might be due to the very low content of silver in the samples, which further contributes to the lack of noticeable silver peaks in the diffraction data. [(Nga et al., 2022)](https://paperpile.com/c/DpxpCn/G3PVG)The key concern in photocatalyst development is improving performance while reducing production costs. [(Tiwari & Jain, 2023)](https://paperpile.com/c/DpxpCn/GoyFQ)[(Graf et al., 2023)](https://paperpile.com/c/DpxpCn/d01eX) This study presents a novel synthesis of BiVO4 photocatalyst via homogeneous precipitation, using surfactants like PVP, EG, CTAB, and SDBS to assist crystal orientation. The samples were evaluated with XRD, SEM, Raman, and DRS. XRD confirmed the purity of all samples, except for the SDBS sample, which showed a mixture of tetragonal and monoclinic phases. [(Helal et al., 2020; Nga et al., 2022)](https://paperpile.com/c/DpxpCn/G3PVG+kC1rm)FTIR spectroscopy is significant for analyzing water content in samples, as water molecules strongly absorb in IR spectral regions. This creates a challenge in isolating water's contribution, particularly in the bending vibrations at about 1640 cm–1, overlapping with peptide bond vibrations (1620–1690 cm–1). Special approaches are required to exclude water's interference, including drying microbial biomass to maintain biomacromolecular integrity. [(Sabarathinam & Madhulaxmi, 2021)](https://paperpile.com/c/DpxpCn/in8VQ)[(Sushanthi et al., 2021)](https://paperpile.com/c/DpxpCn/fnZrA)[(Harsha et al., 2022)](https://paperpile.com/c/DpxpCn/RMrMd) Dried samples can be studied using ATR or DRIFT modes, providing detailed structural and quantitative information without extensive sample preparation. [(Kamnev & Tugarova, 2023)](https://paperpile.com/c/DpxpCn/0NM8Z)The Fourier-transformed amplitude of V K-edge EXAFS k3χ data shows that the peak intensity increases with the concentration of decorative Ag nanoparticles (NPs). This intensity change typically indicates a variation in the coordination number of the metal center. During the formation of oxygen vacancy defects, lattice oxygen atoms escape, reducing the coordination number of the V atoms. [(Neha et al., 2021)](https://paperpile.com/c/DpxpCn/hn2C2)[(Maliael et al., 2021)](https://paperpile.com/c/DpxpCn/iPEd0)[(Lakshmi, 2021)](https://paperpile.com/c/DpxpCn/ItU8g)The increase in peak intensity of the V–O bond in R-space due to Ag NP decoration on BiVO4 suggests an increase in the oxygen coordination number around the V atom centers, reflecting a reduction in oxygen vacancy defects, which can act as recombination centers in BiVO4. [(Nga et al., 2022)](https://paperpile.com/c/DpxpCn/G3PVG)The UV–visible absorption spectra show that BiVO4 absorbs in the UV–visible region with a band centered around 500 nm. The absorption edges of Ag–BiVO4 photoanodes are redshifted with different concentrations of Ag NPs, indicating that Ag NPs enhance light absorption through the localized surface plasmon resonance (LSPR) effect. [(Nga et al., 2022)](https://paperpile.com/c/DpxpCn/G3PVG)Transmission Electron Microscopy (TEM) with immunogold labeling enables high-resolution visualization and localization of nanoparticles (NPs) by using gold-conjugated antibodies to detect antigens. This method provides detailed insights into NP distribution within cells and their interaction with subcellular structures. Although effective, immunogold labeling’s use in toxicology may be limited due to the potential influence of NP surface coatings on biological effects. For targeted NPs, this technique is useful for understanding their cellular localization and distribution, with double-labeling and EFTEM enhancing the analysis of NP association with cellular components. [(Mühlfeld et al., 2007)](https://paperpile.com/c/DpxpCn/O0aTh) TEM images were obtained using a Tecnai T20 G2 (Thermo Fisher Scientific) with a LaB6 electron source and a CETA 16 M 4k × 4k CMOS camera, operating at 200 kV. Imaging was done at various magnifications: 25,000× with 8.53 Å pixels, 50,000× with 4.24 Å pixels, 80,000× with 2.67 Å pixels, and 100,000× with 2.13 Å pixels, all with a 1 s acquisition time. For specimen preparation, a 1 wt % sample suspension in absolute ethanol was ultrasonicated and drop-cast onto a CF200-Cu copper grid with a carbon film. The grid was then dried, inserted into the TEM holder, and pumped to high vacuum. [(Shkatulov et al., 2020)](https://paperpile.com/c/DpxpCn/sCPS4)This study introduces a carbon quantum dots (CQDs)/BiVO4 composite with enhanced photocatalytic activity, effectively degrading rhodamine B (RhB) under simulated solar light. The CQDs' unique properties improve the composite's performance, suggesting a strategy for high-performance CQD-based catalysts. [(Z. Zhang et al., 2019)](https://paperpile.com/c/DpxpCn/ZRAgs)XRD patterns identify BiVO4, Ag2S, and their composites, but distinct peaks for Ag or Ag2S are absent, likely due to their low concentrations and the high crystallinity of BiVO4. SEM and HRTEM images show smooth, plate-like BiVO4 particles. Ag nanoparticles (50-100 nm) are seen on BiVO4, and fine Ag2S nanoparticles (~5 nm) are observed on BiVO4/Ag. TEM and HRTEM confirm lattice spacings of 0.24 nm and 0.34 nm for Ag (111) and Ag2S (110), respectively, confirming Ag2S formation. [(Y. Liu et al., 2020; Z. Zhang et al., 2019)](https://paperpile.com/c/DpxpCn/ZRAgs+6zQmi) BVO@C/Ag MTs were created by coating BiVO4 microtubes with carbon and embedding Ag nanoparticles. This material showed enhanced photocatalytic activity for rhodamine B degradation under visible light, thanks to improved charge separation and light absorption. It maintained high activity after three cycles and could be easily recovered due to its one-dimensional structure. [(M. Zhang et al., 2012)](https://paperpile.com/c/DpxpCn/v6Pwy)

# CONCLUSION

The study demonstrates that Ag-BiVO4 composites, enhanced with seaweed-derived carbon, effectively scavenge reactive oxygen species (ROS) and exhibit strong antifungal activity. The inclusion of seaweed-derived carbon enhances the photocatalytic performance of Ag-BiVO4, leading to increased ROS generation under visible light. This boost in ROS production results in significant inhibition of Candida species, including their biofilm formation and growth, indicating the composites' effectiveness in reducing fungal cell viability and preventing growth.These findings emphasize the biocompatibility and potential of Ag-BiVO4 composites with seaweed-derived carbon for advanced antifungal treatments. Their improved ROS scavenging capability suggests they could be valuable in biomedical applications. Future research should investigate their effectiveness against a wider range of fungal pathogens and explore their clinical potential, offering a novel approach to address the issue of antifungal resistance.

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