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ORIGINAL ARTICLE

Antibacterial Performance of Nanostructured Copper Nitride Thin Films

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Abstract

Objectives: To prepare nanostructured copper nitride (Cu_3N) films at different substrate temperatures, and to assess the antibacterial properties. **Methods:** Nanostructured copper nitride (Cu_3N) films are prepared at different substrate temperatures (303K to 573K) by the dc reactive magnetron sputtering technique on glass substrates and characterized with scanning electron microscopy (SEM), X-ray photoelectron spectroscopy (XPS), X-ray diffraction (XRD), UV-Vis-NIR double-beam spectrophotometer and Pseudomonas aeruginosa (*P. aeruginosa*). **Findings:** As the substrate temperature increased from 303 to 573K, the deposition rate diminished from 17.8 to 10.9 nm/min. The films deposited at room temperature (303K) exhibit a microstructure characterized by island-like features. As the substrate temperature reaches 473K, a significant transformation occurs, resulting in the films achieving a highly compact and uniform microstructure, characterised by well-defined nanorods. The optical transmittance of the films increased from 62% to 74% with the increase of substrate temperature from 303 to 473 K. The nanostructured Cu_3N films show notable antibacterial properties against *Pseudomonas aeruginosa*. **Novelty:** The present obtained nanostructured Cu_3N films are thermally stable even at 473K and exhibited significant antibacterial properties against *Pseudomonas aeruginosa*.

Keywords: Thin films; Nanostructured; Sputtering; Copper nitride; Antibacterial properties

1 Introduction

Microbial invaders can severely undermine health and pose serious health risks. Furthermore, microbial contamination can lead to the rapid degradation of various materials, including those used in industries, consumer products, pharmaceuticals, and food items, resulting in considerable economic losses. Therefore, there is growing emphasis on creating materials endowed with antibacterial properties to address these challenges effectively[(1)],[(2)]. Researchers developed and constructed a versatile antibacterial coating designed to combat bacteria effectively. This innovative coating exhibits properties that not only prevent bacterial growth but also facilitate the removal of both gram-negative and gram-positive bacteria from the surface, thereby enhancing its functionality[(3)]. *Pseudomonas aeruginosa* (*P. aeruginosa)*, a gram-negative bacterium, is an opportunistic pathogen and widely distributed in nature, which causes concern for humanity. Currently, *P. aeruginosa* is a significant focus of research both domestically and globally. It is primarily studied in the areas of clinical and basic medicine, as well as biology and pharmacy, due to its relevance and impact in these fields. Research shows that transition metals like gold, copper, and silver play crucial roles in enabling thin films to have antimicrobial properties. Among these metals, copper (Cu) stands out due to its wide abundance, low toxicity, good biological function and cost-effectiveness[4]. Advances in nanotechnology have shifted attention toward creating copper-based alloys and compound films, such as Cu-Ta[5], Cu_3N-CuO[6], CuTiZn[7], Ti-Cu-N[4], including nanoparticle and nanostructured coatings, to effectively utilize their antimicrobial benefits. Copper nitride (Cu_3N), a copper compound, has wide range of applications in different fields such as photocatalysts, photovoltaic cells, hydrogen gas sensors, high-speed integrated circuit materials, and optical storage devices, because of its unique properties, such as readily available elements, cubic anti-ReO_3 type structure, non-toxic nature, electro-catalytic activity, band gap energy can be easily adjusted, and hardness[8],[9]. However, research on the antibacterial properties of pure Cu_3N films is not extensive. Cu_3N thin films have been prepared using various thin film deposition techniques, with reactive DC magnetron sputtering emerging as a favoured choice. This technique offers distinct benefits such as precise control over the thickness, achieving high purity, ensuring uniformity of the film, and being well-suited for large-scale production[10]. The present study aimed to prepare high-quality nanostructured Cu_3N thin films on glass substrates by dc reactive magnetron sputtering at different substrate temperatures, and their antibacterial activities against *P. aeruginosa* were examined.

2 Methodology

The DC reactive magnetron sputtering technique is employed to prepare the nanostructured Cu_3N films using a high-purity (99.9%) Cu target. These films are deposited onto glass substrates, and specific deposition parameters are outlined in Table 1, detailing the precise conditions under which the nanostructured films are prepared.

Table 1. Deposition parameters were maintained during the preparation of nanostructured Cu_3N films

Base pressure	$5x10^{-6}$ mbar
Target to substrate distance	70mm
Nitrogen partial pressure (pN_2)	$3x10^{-3}$ mbar
Substrate temperature (Ts)	303K, 373K, 473K, 573K
Sputtering power (Pw)	80W
Working pressure (Sp)	$3x10^{-2}$ mbar

2.1 Characterization techniques

The films deposited in the study were characterised through various analytical methods. Scanning electron microscopy (SEM) was employed to investigate the films microstructure, providing detailed images of their surface features. To explore the compositional properties of these films, X-ray photoelectron spectroscopy (XPS) was used, enabling the identification and quantification of elemental components. The crystal structure of the films was analysed by using X-ray diffraction (XRD). Additionally, the optical properties of the films were measured using a UV-Vis-NIR double-beam spectrophotometer. The films thickness was measured by ellipsometry, and the thicknesses of the films were approximately 240 nm.

2.2 Bacterial culture and growth conditions

Pseudomonas aeruginosa (MTCC 424), a gram-negative bacterium, was sourced from the Microbial Type Culture Collection located in Chandigarh, India. For culture, the bacterium is grown under controlled laboratory conditions using LB media at a temperature of 30°C. Initially, *Pseudomonas aeruginosa* is cultured in LB broth overnight at 30°C, after which a loopful of the grown culture is transferred to Mueller Hinton broth (from Thermo Scientific) and placed on a rotary shaker at the same temperature until it reaches a turbidity matching the 0.5 McFarland standard. Subsequently, 0.25 mL of the microorganism suspension is inoculated into molten Mueller Hinton agar and distributed into petri dishes utilizing the pour plate technique.

3 Results and Discussion

3.1 Deposition rate

Figure 1 demonstrates how the deposition rate of nanostructured Cu_3N thin films varies with changes in substrate temperature. As the substrate temperature increased from 303 to 573K, the deposition rate diminished from 17.8 to 10.9 nm/min. This reduction occurs at higher substrate temperatures, which may be due to the adsorbed reactive species desorbing (reevaporating) from the substrate surface, resulting in a slower deposition rate. Similar observations were made by Majumder et al.[11], they noted a decrease in the deposition rate (from 4.8 to 4.3 nm/min) of rf magnetron sputtered Cu_3N films when the temperature increased from 100 to 300°C. This phenomenon is attributed mainly to the decrease in the sticking coefficient of copper at elevated temperatures. Moreover, the decrease in deposition rate becomes more pronounced when temperatures exceed 250°C as Cu_3N begins to decompose above approximately 300°C, potentially reducing the surface reaction rate of copper and nitrogen at such high temperatures. Magnetron sputtering delivers a superior deposition rate and enhances the quality of films produced, making it a popular choice across various applications such as solar energy, optics, electronics, and hard film production[12].

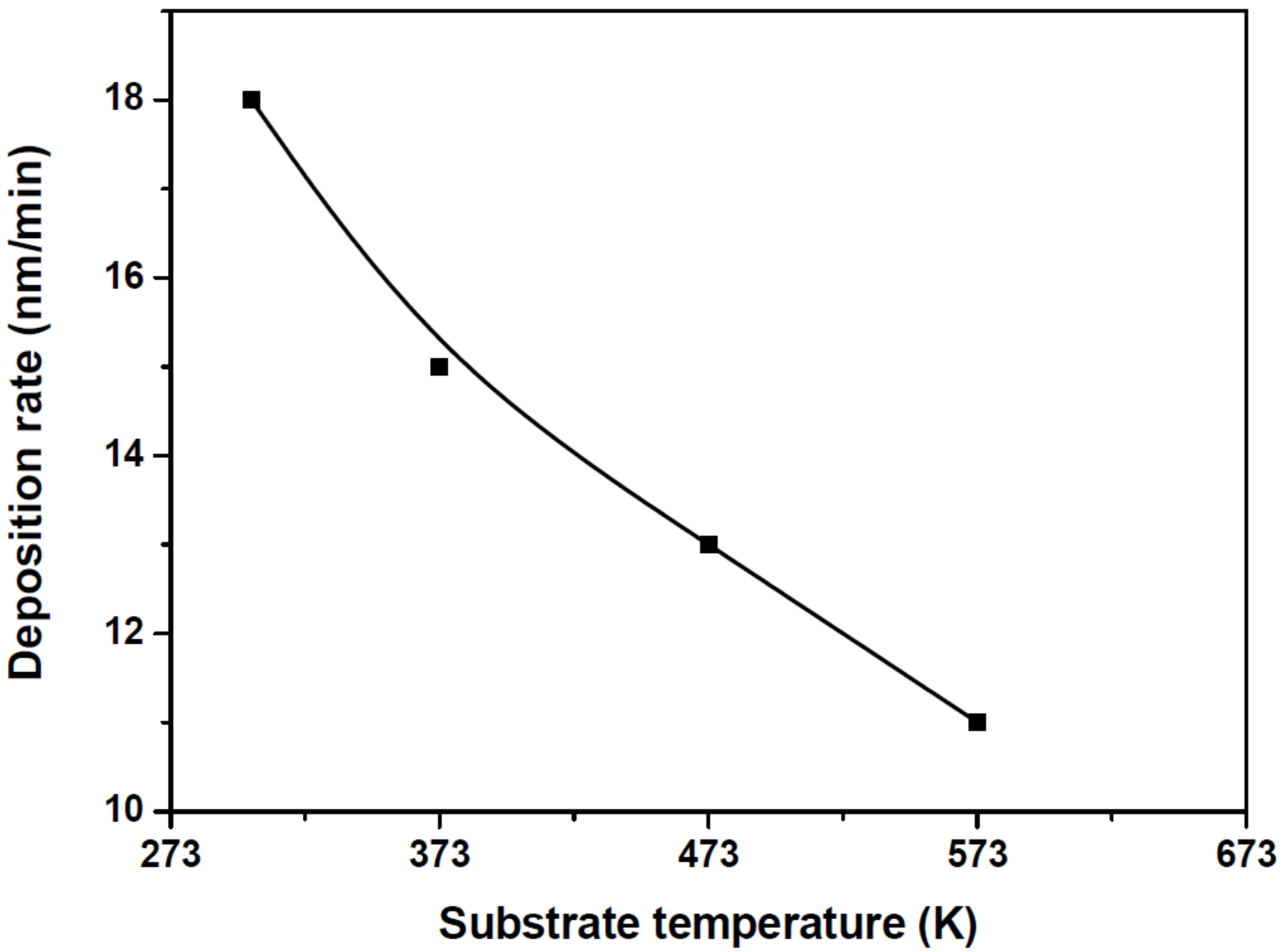


Fig 1. Deposition rate of nanostructured Cu_3N thin films as a function of substrate temperature

3.2 Microstructure

The microstructural characteristics of copper nitride films were found to be highly sensitive to the substrate temperature during deposition. The SEM images of copper nitride films at different substrate temperatures are shown in Figure 2(a-d). The films deposited at room temperature (303K) exhibit a microstructure characterized by island-like features composed of small, poorly defined, flake-shaped grains. This structure emerges due to the limited mobility of adatoms, which hinders the coalescence of nuclei. Increasing the substrate temperature to 373 K enhances surface diffusion, enabling adatoms to settle in energetically favourable positions, which leads to the merging of grains into a more compact and uniform structure. This process marks a transition from a loosely packed island configuration to the growth of rods and larger grains. As the substrate temperature reaches 473K, a significant transformation occurs, resulting in the films achieving a highly compact and uniform microstructure, characterised by well-defined nanorods and a reduction in surface defects. The smooth and dense appearance of the surface at 473 K indicates a favorable temperature range for optimal adatom mobility and grain boundary migration, contributing to improved densification. Nanorods are important in science and technology due to their high surface-to-volume ratio, which enhances their effectiveness in numerous applications, making them versatile and impactful in fields ranging from electronics to medicine. Further increasing the substrate temperature to 573 K results in the disappearance of the nanorod structures and development of small cracks and discontinuities on the films surface. This phenomenon is likely caused by the induced residual stress and the decomposition of Cu_3N. In the literature, similar results are observed by Monireh et al.[13] in RF magnetron sputtered WS_2 thin films deposited onto the glass substrates. When films were deposited at various substrate temperatures, such as 100°C, 200°C, and 300°C, a noticeable transformation in the films microstructure was observed. Beginning as nanoparticles, the microstructure transitions into nanosheets and eventually develops into distinctly separated grains on their surface. DC magnetron sputtering offers a higher deposition rate than RF magnetron sputtering, yet it still enables the precise formation of high-quality nanostructured Cu_3N films.

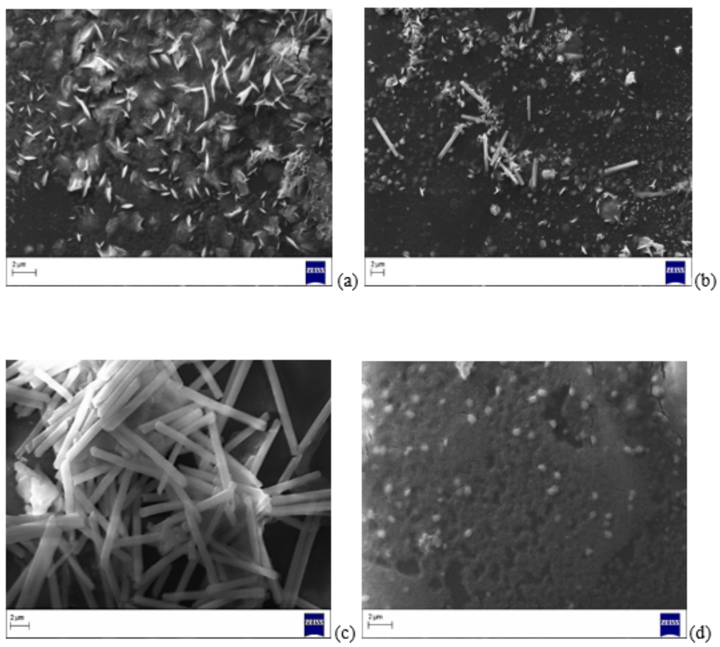


Fig 2. SEM images of Cu_3N films at different substrate temperatures: (a) 303K, (b) 373K, (c) 473K and (d) 573K

3.3 Compositional properties

To analyze the purity and composition of nanostructured Cu_3N films, the XPS measurements were performed. Figure 3(a) presents the XPS spectra of the nanostructured Cu_3N film deposited at a substrate temperature of 473K. The high-intensity peaks are located at 932.5 eV and 952.6, corresponding to Cu 2p3/2 and Cu 2p1/2, respectively. These peaks are indicative of copper-nitrogen (Cu-N) bonding[14]. The films deposited at a substrate temperature of 573K, the peaks position shifted towards the higher binding energy side. The similar effect was observed by Shikha et al.[15] in high-power impulse magnetron sputtered Cu_3N films. They found that the peak intensity related to copper bonding at 932 eV (perfect Cu-N binding) shifted significantly to 933.4 eV when the substrate temperature raised from 150 to 200°C. This substantial shift in peak position is largely due to the decreased nitrification rate of copper, as high substrate temperatures surpass the optimal range for effective nitrification. The current XPS results indicate that nanostructured Cu_3N films are thermally stable even at 473K, the peak position of Cu

2p3/2 is around 932.5 eV, which is 0.9eV lower than the reported[15] Cu_3N film deposited at 200°C. Figure 3(b) shows the photoemission peak of N1s. The N1s peak appears at 397.5 eV and is associated with the Cu-N bonding in Cu_3N[16].

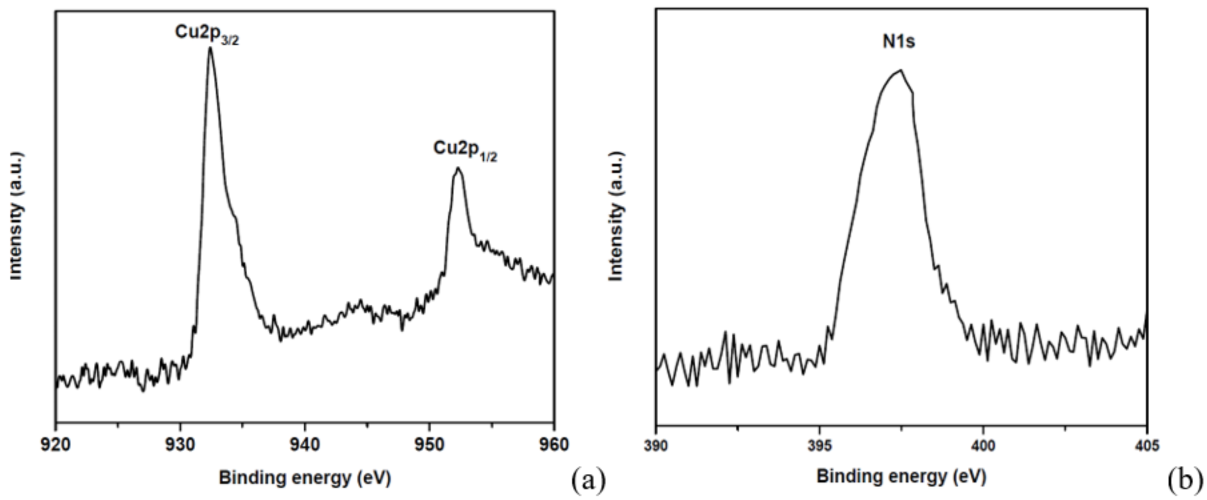


Fig 3. XPS spectra: (a) Cu 2p peaks and (b) N1s peak of the nanostructured Cu_3N film deposited at substrate temperature of 473K

3.4 Structural properties

Figure 4 shows the X-ray diffraction (XRD) patterns of nanocrystalline Cu_3N films at different substrate temperatures. These patterns reveal that the Cu_3N films deposited at substrate temperatures of 303 K and 473 K display broad humps, suggesting that the films possess a predominantly nanocrystalline or partially amorphous structure. Specifically, both patterns display a broad hump maximum centred around 2θ values of approximately 20–30°. Literature[17],[18] indicates that fully crystalline Cu_3N films typically exhibit distinct peaks associated with cubic Cu_3N structures, with peaks occurring near 2θ values of 23°, 33°, 41°, and 48°, corresponding to (100), (110), (111), and (200) orientations, respectively. Additionally, it is noted that increasing substrate temperatures to an optimal level generally sharpens these peaks and enhances the preferred crystalline orientation of the films. When films are deposited at a substrate temperature of 303 K, they exhibit a broad maximum around the expected area for the Cu_3N (100) orientation. However, the absence of well-defined peaks indicates that the grain growth is restricted, resulting in extremely small crystallites due to the limited mobility of adatoms during the growth process. Increasing the substrate temperature to 473 K results in a slight intensification and narrowing of the main hump, which implies an improvement in structural ordering and a modest increase in the average grain size. Higher substrate temperatures provide increased thermal energy that facilitates better migration of atoms, leading to improved structural properties. When the films are deposited at a substrate temperature of 573K, their intensity tends to slightly decrease. This reduction is possibly caused by the re-evaporation of the deposited material, which prevents the atoms from settling into their stable positions.

3.5 Optical properties

The optical transmittance spectra of nanostructured Cu_3N thin films prepared at different substrate temperatures are shown in Figure 5. The optical transmittance of the films increased with the increase of substrate temperature from 303 to 473 K. As the substrate temperature increases, the Cu-N bonding increases, which contributes to greater transmittance. However, when the films were prepared at a higher substate temperature of 523K exhibited low optical transmittance, attributed to a reduced nitrogen content and the development of a distinct microstructure. Haifa et al.[19] observed that the continuous dropping of transmittance with rising substrate temperatures in rf reactive sputtering Cu_3N films deposited on ITO substrate. As the substrate temperature increases, the absorption edge of the films moves towards a higher wavelength side.

The optical band gap (E_g) of the nanostructured Cu_3N films was determined by the extrapolating the linear portion of the plots of $(\alpha h\nu)^{1/2}$ versus $(h\nu)$ (α is the absorption coefficient, $h\nu$ is the photon energy). It was observed that the optical band gap decreased as the substrate temperature rose from 303K to 473K, but it increased again at a higher substrate temperature

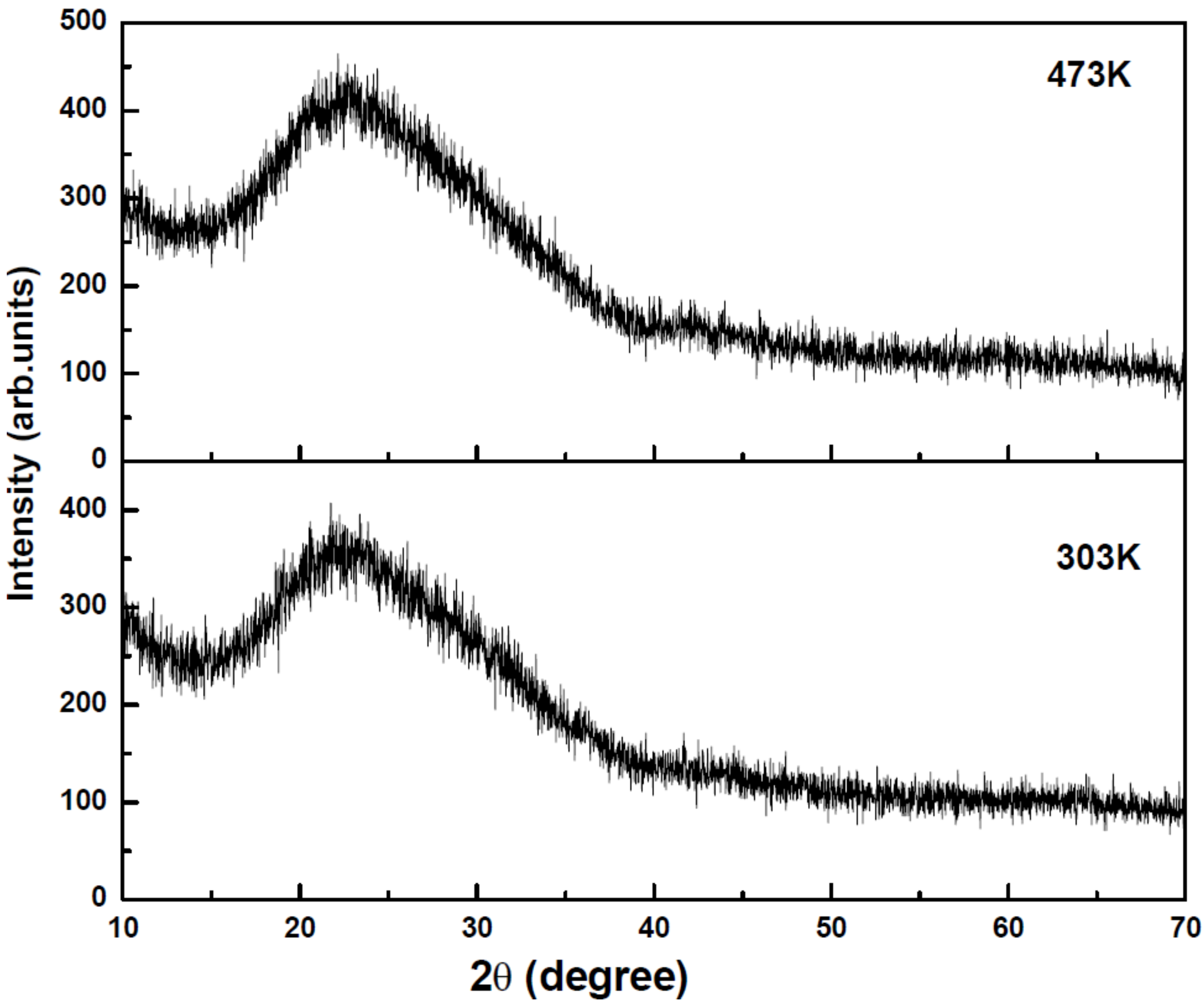


Fig 4. XRD patterns of nanostructured Cu_3N thin films at different substrate temperatures

of 573K. The present obtained band gap values are1.81eV, 1.73 eV, 1.67eV and 1.71 eV for the substrate temperature of 303K, 373K, 473K and 573K, respectively. These variations in the optical band gap with substrate temperature can be attributed to microstructural and compositional changes occurring in the films.

3.6 Antibacterial properties

This study examined the antimicrobial properties of nanostructured Cu_3N films against *Pseudomonas aeruginosa* (*P. aeruginosa*) by employing the pore plate technique. Pseudomonas aeruginosa was cultured in LB broth at 30°C overnight, and antibacterial results are shown in Figure 6(a&b). These findings illustrate that nanostructured Cu_3N films have notable antibacterial properties, which are influenced by their microstructure and composition. Specifically, films produced at substrate temperatures below 473K were ineffective in curbing the growth of *P. aeruginosa*, while those prepared at 473K created larger bacterial inhibition zones. This effectiveness is due to the improved nanostructure that offers a larger surface area for copper ion release and better stoichiometry, contributing to a sustained antibacterial effect. Bagdat et al. [5] observed that Ta-Cu coatings, especially after annealing at 600 °C, exhibited remarkable bactericidal activity, notably against Gram-negative bacteria, including *E. coli*, *S. enterica*, and *P. aeruginosa*. This study, nanostructured Cu_3N films, when prepared at a substrate temperature of

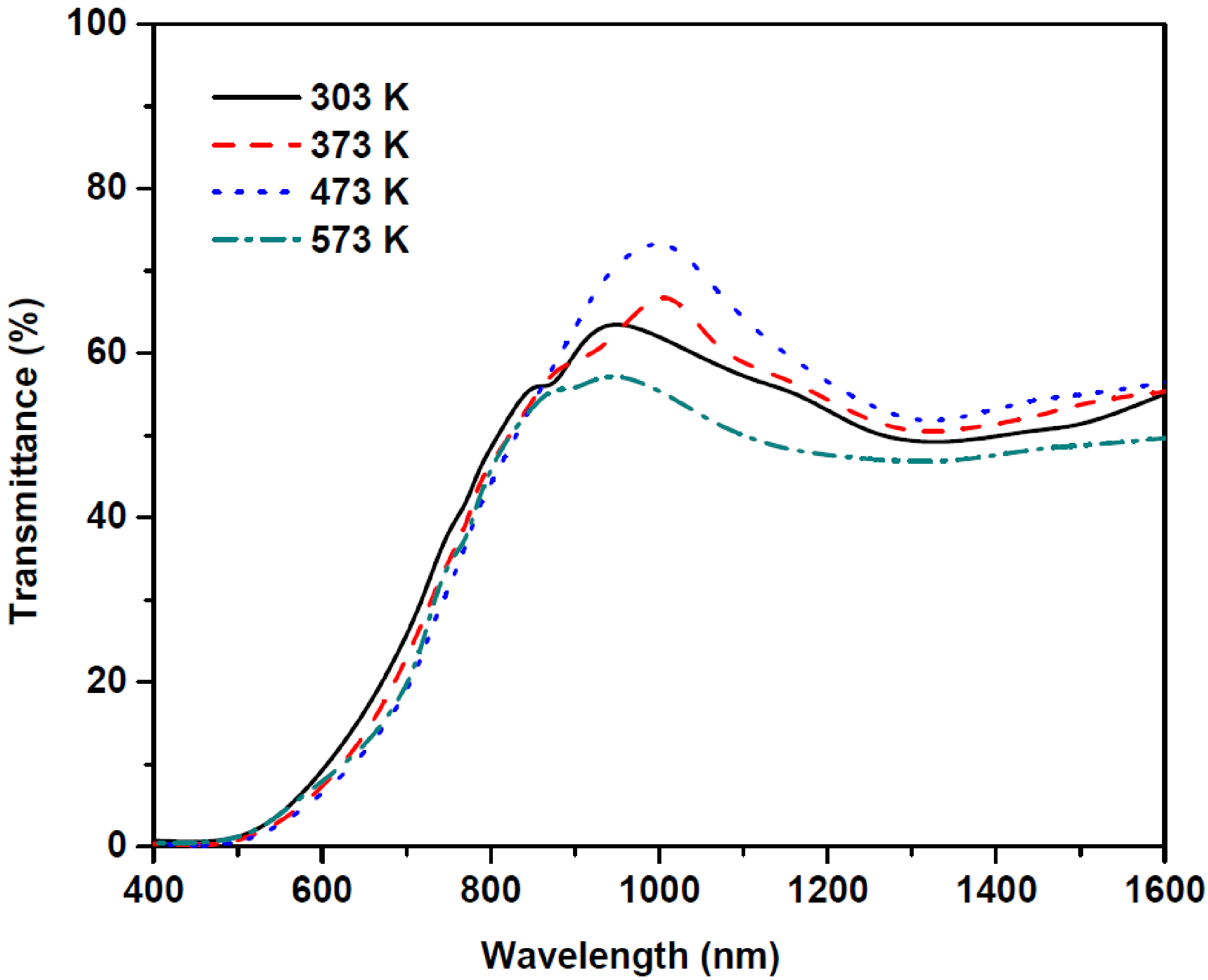
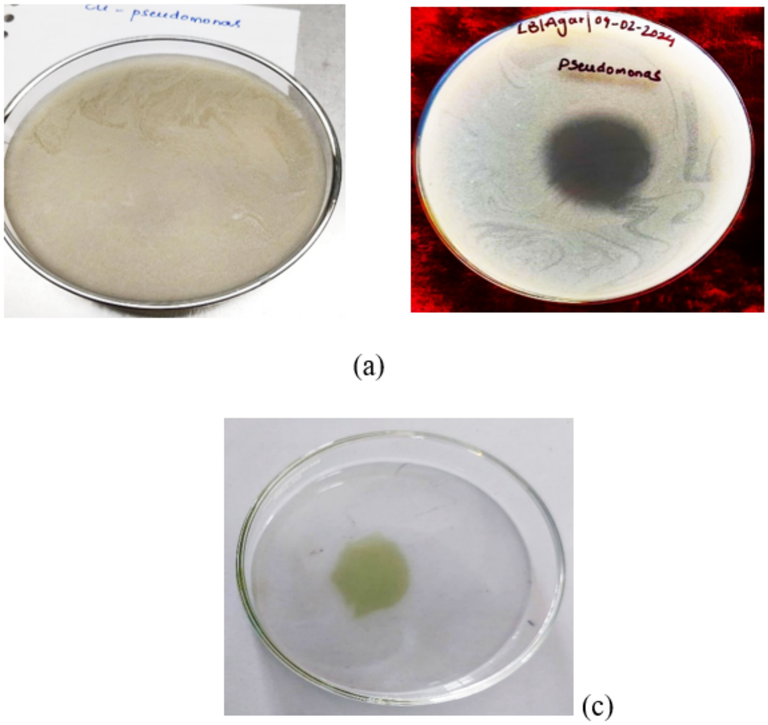


Fig 5. Optical transmittance spectra of nanostructured Cu_3N thin films prepared at different substrate temperatures

473K, demonstrates significant antibacterial effects against *P. aeruginosa*. Nanostructured Cu_3N films have shown exceptional effectiveness as antimicrobial materials, offering significant potential to prevent bacterial contamination.

4 Conclusion

This study investigated the influence of substrate temperature on the microstructural, compositional, optical and antibacterial properties of nanostructured Cu_3N films prepared by magnetron sputtering. Research into the antibacterial properties of pure copper nitride (Cu_3N) films is relatively very limited. The results showed that the Cu_3N films microstructure was significantly influenced by increasing the substrate temperature. This could be explained by the enhanced surface diffusion, enabling adatoms to settle in energetically favourable positions when the substrate temperature is high. The Cu-N bonding increased with increasing the substrate temperature from 373K to 473K. The XRD patterns show that the Cu_3N films exhibited broad peaks, indicating these films are largely nanocrystalline or partly amorphous in nature. The absorption edge shifted towards longer wavelength, and a reduction in band gap was observed by increasing the substrate temperature. Notably, nanostructured Cu_3N films prepared at a substrate temperature of 473K show excellent antibacterial properties against *P. aeruginosa*. The coating of nanostructured Cu_3N thin films within the petri dish substantially enhanced the anti-bacterial effectiveness, resulting in the formation of complete bacterial inhibition zones. These results suggest that magnetron-sputtered nanostructured Cu_3N



(a) (b) (c)

Fig 6. Antibacterial properties of nanostructured Cu_3N films at different substrate temperatures: (a) 303K, (b) 473 K and (c) uncultured film

thin films could serve as an effective approach for creating antimicrobial coatings. Nevertheless, further studies are required to accurately assess the antibacterial properties of nanostructured Cu_3N films and offer clear guidance for their development and practical use. It is crucial to establish controlled testing conditions that are specifically tailored to target objectives.

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