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Synthesis of Polyvinylpyrrolidone Nanoparticle and its Antimicrobial Susceptibilities Against Gram-Negative and Gram-Positive Bacteria

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ABSTRACT

The proliferation of antibiotic-resistant microorganisms necessitates the development of new antimicrobials. Nanoparticles of strontium oxide (SrO) and polyvinylpyrrolidone (PVP) were synthesised, characterised, and shown to be effective against *Escherichia coli* and *Staphylococcus aureus* in this investigation. The research of strontium oxide nanoparticles made by co-precipitation was conducted using SEM, XRD, EDX, UV, and FTIR. There was an evaluation of antibacterial activity at 25% and 50% PVP strontium oxide concentrations using the Kirby-Bauer disc diffusion technique. With a 35-mm inhibition zone, a 50% concentration showed modest activity against *E. coli* and excellent antibacterial efficacy against *S. aureus*. These results raise the possibility that PVP nanocomposites containing SrO oxide have antibacterial properties, especially when applied to gram-positive bacteria.

INTRODUCTION

Staphylococcus aureus (*S. aureus*) and *Escherichia coli* (*E. coli*) are widely studied due to their importance to human health. Although they both live in different biological habitats and cause illnesses in different ways, the two types of bacteria are quite similar. Human and animal intestines are common habitats for the Gram-negative bacteria *Escherichia coli*. It aids digestion and nutrient absorption and is usually nontoxic (Kaper, Nataro, & Mobley, 2004). Contaminated meat, unpasteurised milk, and fresh fruit are frequent causes of *E. coli* outbreaks, but certain strains may cause serious disease. Pathogenic *Escherichia coli* is spread by direct touch or contaminated food or water, especially under poor hygiene and sanitation (Scallan et al., 2011). *E. coli* causes most UTIs and bloodstream infections in healthcare workers, especially in the elderly and immunocompromised (Russo & Johnson, 2003). *Staphylococcus aureus* is common in healthy people's nasal passages and skin. *S. aureus* is normally present in the body in modest levels (about 30%), but it may cause sickness if it enters via open wounds, punctures, or other penetrations (Lowy, 1998). *Staphylococcus aureus* scalp infections range from impetigo and boils to endocarditis, pneumonia, and sepsis (Tong et al., 2015).

S. aureus infections are harder to treat because the bacteria create biofilms and become resistant (Otto, 2008). Example: methicillin-resistant A notable *Staphylococcus aureus* strain has developed antibiotic resistance, causing clinical and community problems (DeLeo, Otto, Kreiswirth, & Chambers, 2010). *Staphylococcus aureus* may transmit hospital-acquired infections via personal contact or contaminated surfaces in patients with indwelling medical devices or surgical wounds (Klevens et al., 2007). Due to their infection-causing capacity and affiliation with healthcare institutions, *E. coli* and *S. aureus* pose serious health threats. Knowing how bacteria cause illness, spread, and may be avoided is essential for public health and reducing bacterial infections worldwide.

New antimicrobial medications and formulations are required to address the global health crisis of antibiotic resistance in harmful bacteria (Ventola, 2015).

Developing novel therapies is of utmost importance due to the global health danger posed by newly antibiotic-resistant bacteria. The potential for nanotechnology to create antimicrobial nanoparticles is promising (Li et al., 2019). Because they destroy many germs, silver nanomaterials are of interest to several sources (Rai et al., 2020). Polyvinylpyrrolidone (PVP) and strontium oxide (SrO) nanoparticles may disrupt bacterial membrane contacts and

biological processes, making them potentially effective in fighting microbial diseases (Gopinath et al., 2021).

We tested PVP-SrO nanoparticles for antibacterial activity against *Staphylococcus aureus* and *E. coli* using Kirby-Bauer disc diffusion. These bacterial strains were chosen for their resistance and medicinal potential. *Escherichia coli* and *Staphylococcus aureus* are bacteria. The former causes UTIs and food poisoning, whereas the latter causes skin infections and catastrophic systemic diseases (Ventola, 2015).

MATERIAL AND METHODS

CHEMICALS

Only analytical-grade substances were used in this experiment. The German pharmaceutical firm Merck supplied the polyvinylpyrrolidone (PVP), sodium hydroxide (NaOH), and strontium nitrate hexahydrate ($\text{Sr}(\text{NO}_3)_2 \cdot 6\text{H}_2\text{O}$). The German company Sigma-Aldrich provided us with the salt vinblastine sulphate, or VNB. Distilled water (DI) was used to create a stock solution of vinblastine sulphate with a concentration of 0.0002 M. The BRB buffer was made in DI water and has a concentration of 0.1 M. The Britton-Robinson buffer (BRB) electrolyte, which was composed of 0.1 M acetic acid, 0.1 M phosphoric acid, and 0.1 M boric acid, provided the necessary support. The buffer was adjusted to the required pH by adding 0.1 M NaOH and 0.1 M HCl. Reaching out to Sigma-Aldrich (based in the UK and Sweden) allowed us to get the 5% Nafion® solution. After that, it was diluted with DI water until it reached a concentration of 0.1%.

SYNTHESIS PROCEDURE FOR PVP-SRO NANOPARTICLES

Using a straightforward and easy co-precipitation approach, PVP-SrO nanostructures were effectively synthesised. Strontium nitrate hexahydrate ($\text{Sr}(\text{NO}_3)_2 \cdot 6\text{H}_2\text{O}$) was the salt precursor, sodium hydroxide (NaOH) was the reducing agent, and polyvinylpyrrolidone (PVP) was the stabilising agent. The following solutions were made in separate 50 ml volumetric flasks: a 0.1 M solution of ($\text{Sr}(\text{NO}_3)_2 \cdot 6\text{H}_2\text{O}$), a 0.5 M solution of NaOH, and a 5% solution of PVP. A 200 ml beaker was filled with a 0.1 M solution of ($\text{Sr}(\text{NO}_3)_2 \cdot 6\text{H}_2\text{O}$). After fifteen minutes of vigorous stirring, the liquid had homogenised. A 5% PVP solution and 0.5 M NaOH were added to an original salt solution in a step-by-step manner to improve it. With the foil still covering it, the solution was agitated for a further two hours. As a result of the process, $\text{Sr}(\text{OH})_2$ precipitates are formed. Filtering, washing with deionised water, and oven-baking at

120 °C for 2 hours dried the synthesised chemicals into precipitates. To complete the production of pure crystalline PVP-SrONPs, the $\text{Sr}(\text{OH})_2$ must be heated in a muffle furnace to 500°C for a duration of four hours.

RESULTS AND DISCUSSION

UV-VISIBLE SPECTROSCOPY

One of the best ways to make sure your nanoparticles are stable and uniformly distributed is to use ultraviolet-visible spectroscopy after production. By combining the SrNO_3 precursor salt with PVP for capping and NaOH for reducing, a colloidal dispersion of nanoparticles made of strontium oxide was produced. According to the previous research²⁹, which confirmed the effective synthesis of PVP functionalised SrO nanoparticles, the UV-Vis spectra of PVP-SrO exhibit an absorption peak at around 296.8 nm, as seen in Figure 1.

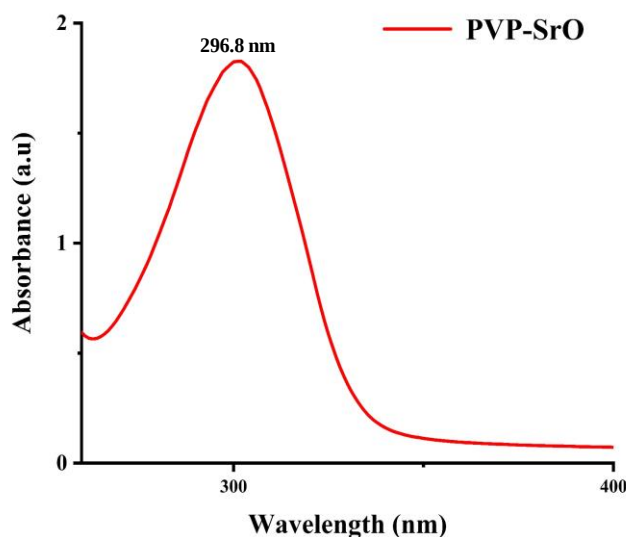


FIG 1. U.V VISIBLE SPECTRUM OF PVP-SRO NANOSTRUCTURES

FTIR ANALYSIS

The various functions included within the synthesised PVP-SrO nanostructures were identified using Fourier transform infrared spectroscopy. The FTIR spectra of PVP, SrO, and PVP-SrO were collected to confirm the procedures used for production and the modifications applied to the nanostructures. In Figure 2a, we can see the Fourier transform infrared spectra of pure PVP, a PVP-SrO hybrid, and SrO nanoparticles. Looking for the distinctive band at 1646 cm^{-1}

makes it easy to identify the pyrrolidone C=O group. In addition to the absorption peak at 1430 cm⁻¹ induced by the CH band of PVP and other pertinent bands, there are other notable bands at 1293 cm⁻¹ that come from the C-N stretching vibrations of PVP. O-H symmetric stretching yields vibrational peaks at 3416 cm⁻¹, CH₂ symmetric stretching at 2949 cm⁻¹, and CH₂ asymmetric stretching at 2894 cm⁻¹, all caused by a wide peak centred at around 3416 cm⁻¹. At 560 cm⁻¹ to 130, the PVP stretching of the C-N=O bands can be seen. The typical spectra of SrO NPs (Figure 2b) shows this. Sr-O may be stretched in symmetrical and asymmetrical ways, as shown by the bends at 848 cm⁻¹ and 701 cm⁻¹. The band at 1437 cm⁻¹ shows that as H₂O absorbs moisture from the air, it experiences an O-H bend. Figure 2c displays the PVP-SrO FTIR spectrum. According to Sr-O, symmetric stretching is indicated by the peak at 839 cm⁻¹, whereas asymmetric stretching is shown by the peak at 704 cm⁻¹. The PVP functional groups are shown by the stretching bands at 3612 cm⁻¹, 570 cm⁻¹, 1025 cm⁻¹, 1434 cm⁻¹, and 1680 cm⁻¹. These bands are caused by the following groups: O-H, C=O, CH₂, C=C, R=NH, and N=C=O. The little shift and weakening of peaks 32 demonstrate the effective synthesis of strontium oxide nanoparticles functionalised with PVP.

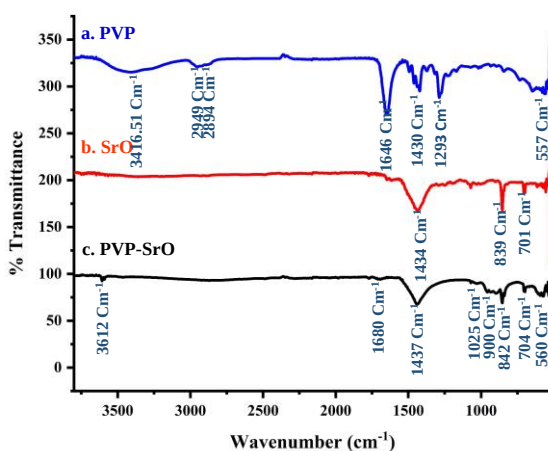


FIG. 2. THE PVP-SRO HYBRIDS' FTIR RESONANCE ENERGY DISTRIBUTION. SECTION A: PURE PVP, AND SECTION B: RU NANOSTRUCTURES. FUNCTIONALISED INORGANIC SRO NANOSTRUCTURES WITH PVP

XRD ANALYSIS

XRD is useful for determining crystallinity and phase transparency. XRD patterns in Bragg's

peaks may help explain the material's crystalline nature. Amorphous materials with short-range ordering have significantly humped diffractogram peaks. Strong peaks suggest crystallinity. Wide and narrow peaks imply crystalline and semi-crystalline characteristics. XRD may reveal the PVP-functionalized SrO's crystalline structure. Figure 3 shows the PVP-functionalized strontium oxide XRD pattern. Strontium oxide PVP333, 34's XRD pattern displays crystalline planes (110), (111), (101), (200), (102), (200), and (220) at 19.82° , 23.4° , 28.4° , 30.9° , 36.7° , 39.4° , 46.9° , and 48.7° . Strong diffractogram peaks indicate crystalline PVP-SrO NPs. The computational findings suggest that cubic PVP-SrO has a 20-nm crystalline dimension. The particle size was determined using the Debye-Scherrer formula: $D = K\lambda / (\gamma \cos \theta)$.

$\Delta 2\theta$, the adjusted full width at half maximum, and λ , the x-ray radiation wavelength, are defined here. Many mathematical calculations assume $K=1$. The X-ray diffraction pattern shows crystalline nanoparticles. The diffraction peaks are consistent with those in the database (JCPDS file No. 6-520)33, which proves that the PVP functionalised SrO NPs were successfully synthesised.

FIG. 3. XRD PATTERNS OF PVP-SRO NANOSTRUCTURES SEM AND MORPHOLOGICAL CHARACTERISTICS

Amazing structural details are shown in scanning electron micrographs (SEM) of PVP-SrO NPs, which are synthesised polyvinylpyrrolidone functionalised strontium oxide nanoparticles. Figure 4 shows that at low resolution, the SrO nanoparticles have a structure similar to tiny nano beads. Due to the high-temperature functionalisation of the PVP polymer, the SEM picture indicates that the produced particles aggregate. The unique shape is intriguing because it increases the sensing activity and improves electro-analytical applications by indicating a

larger surface area and more active spots. Additionally, SrO NPs have a surface that is somewhat rough, which is characteristic of nanoparticles and is very useful for electroanalysis. The nanosensors' enhanced surface properties allow them to enhance redox reactions by interacting with analytes. In general, scanning electron microscopy gives useful information on the morphology and structure of the PVP-SrO NPs that were made, which might be used to enhance redox processes.

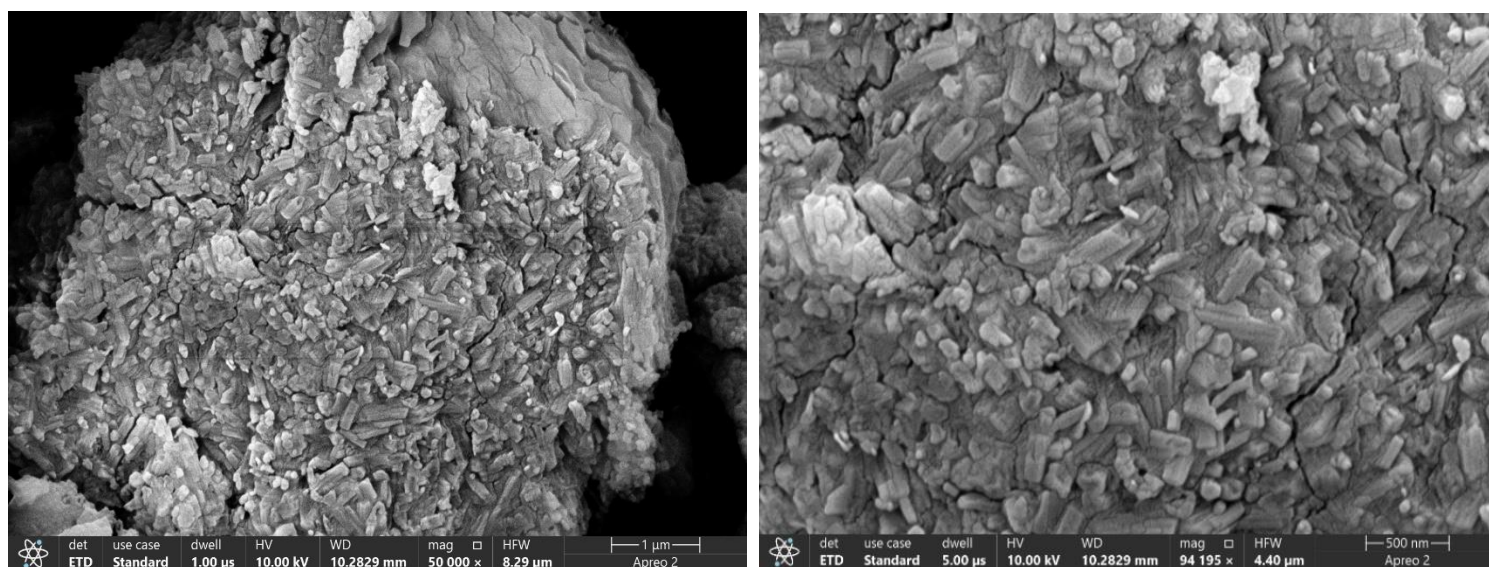


FIG. 4. FESEM PVP-SrO

NANOSTRUCTURES AT LOW AND HIGH RSOLUTION

THE ENERGY DISPERSIVE SPECTROSCOPY (EDS) ANALYSIS

Energy dispersive spectroscopy is crucial for sample component identification. EDS analysis found PVP-functionalized SrO NP components. Figure 5 depicts strontium (Sr), oxygen (O), and carbon (C) in PVP-SrO's EDS spectrum. Essential element percentages of 68.3%, 23%, and 8.1% support the projected atomic ratio, according to EDS data. Along with other example items, this technique tests for them. Polyvinylpyrrolidone, the functionalising agent used to make NPs, produces these components. PVP is made of carbon, nitrogen, and oxygen. PVP capping with strontium oxide worked because carbon is present. We synthesised SrO nanoparticles with the right composition, and this extensive EDS characterisation shows that the capping agent is present. These findings provide nanoparticle structure and chemistry, which are crucial for electroanalytical applications.

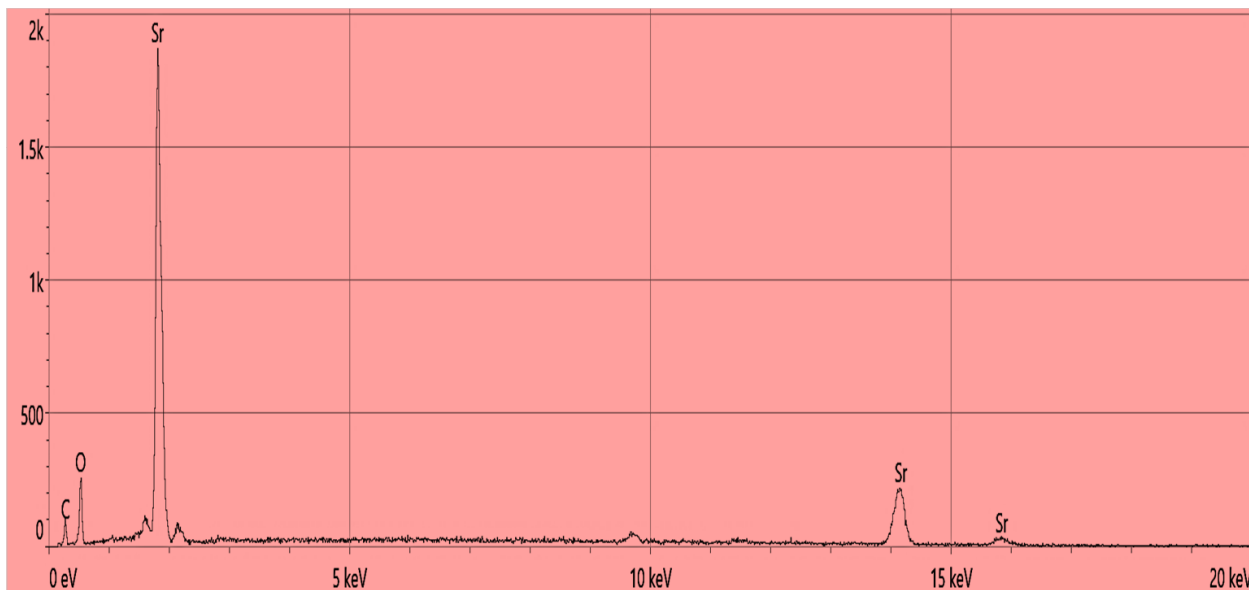


FIG. 5. EDX SPECTRUM OF PVP-SrO NANOSTRUCTURES

METHODOLOGY

SYNTHESIS OF PVP-SrO NANOSTRUCTURES

Every single experiment included some kind of chemical analysis. A simple co-precipitation technique was used to fabricate PVP and SrO nanostructures. In its original form, the sodium salt was referred to as $\text{Sr}(\text{NO}_3)_2 \cdot 6\text{H}_2\text{O}$. In order to stabilise and reduce the substance, we used NaOH and PVP. Solutions of 0.1 M $\text{Sr}(\text{NO}_3)_2 \cdot 6\text{H}_2\text{O}$, 0.5 M NaOH, and 5% PVP were produced in separate 50 mL flasks. Afterwards, transfer the 0.1 M $\text{Sr}(\text{NO}_3)_2 \cdot 6\text{H}_2\text{O}$ solution to a 200 mL beaker. The mixture was vigorously stirred for fifteen minutes to ensure uniformity. A single drop of both the 0.5 M NaOH and 5% PVP solutions was added to the precursor salt solution. After covering the mixture with aluminium foil, stir it for two hours. The formation of $\text{Sr}(\text{OH})_2$ precipitates was the end outcome of this procedure. Following filtering and washing with deionised water, the precipitates were dried in an oven at 120 °C for two hours. $\text{Sr}(\text{OH})_2$ was heated to 500°C for four hours in a muffle furnace as the penultimate stage. This process was used to make PVP-SrO NPs, which are nanoparticles of pure crystalline strontium oxide.

BACTERIAL CULTURE PREPARATION

We found two different bacterial strains in our microbiological susceptibility test. *Staphylococcus aureus* and *Escherichia coli*, two types of bacteria, were used. Procedure for the Kirby-Bauer Disc Diffusion Probability Test. All of the cultures did quite well on Mueller-

Hinton agar. Transferring the bacteria from the colony after they had grown overnight resulted in a bacterial solution with a concentration of 0.5 McFarland units (1.5×10^8 cfu/mL). The organisms were streaked around the plate using a sterile brush, with very narrow spacing in between each streak. Antibiotic particles started to fall as soon as they touched down on the plate. For every plate, four samples were added. The plates were maintained at a temperature of $35^\circ\text{C} \pm 2^\circ\text{C}$ for a period of 18 to 24 hours. After a day, we checked inhibitory zones.

RESULTS

The results indicate that at a lower concentration (25 mg), PVP-SrO nanoparticles exhibited antimicrobial activity against *E. coli* but were ineffective against *S. aureus*. At a higher concentration (50 mg), resistance was observed in both bacterial strains, suggesting a potential concentration threshold beyond which the nanoparticles lose efficacy.

TABLE 1: ANTIMICROBIAL SUSCEPTIBILITIES TEST

SNO.	Sample	Culture <i>E. coli</i>	<i>S.aureus</i>
1	PVP-SvO Nanoparticle (25 mg)	S	R
2	PVP-SvO Nanoparticle (50 mg)	R	R

Notes: The above-reported sensitivities are done by Kirby Bauer disk diffusion method.

Key: S =Sensitive, R =Resistant, I =Intermediate

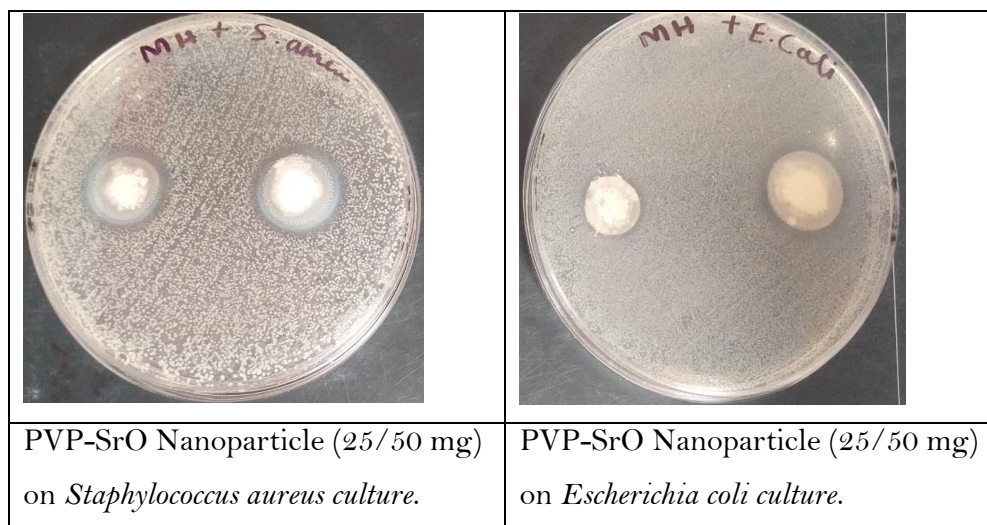
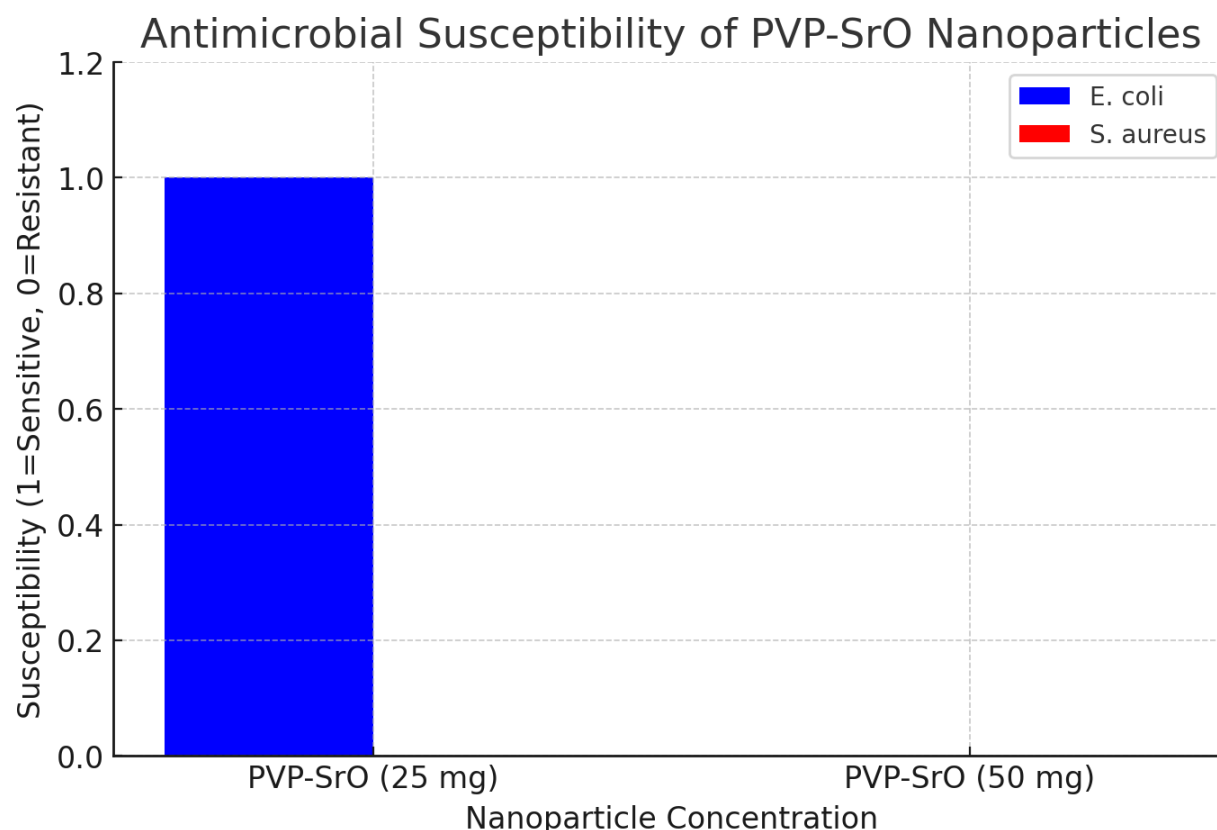


FIGURE:1 PVP-SRO NANOPARTICLE ON S.AUREUS AND E. COLI CULTURE.

The Kirby-Bauer disk diffusion method confirmed these findings, as the zone of inhibition was noticeable for *E. coli* at 25 mg but significantly reduced at 50 mg. The lack of effectiveness against *S. aureus* at both concentrations suggests species-specific differences in susceptibility,

possibly due to variations in bacterial cell wall composition and resistance mechanisms. These results show the importance of optimizing nanoparticle formulations to enhance antimicrobial efficacy while minimizing resistance development. Further studies, including mechanistic investigations and combination therapies with conventional antibiotics, may provide insights into improving the clinical applicability of PVP-SrO nanoparticles.



DISCUSSION

This study examines PVP-SrO nanoparticles' antibacterial effects against Gram-negative and Gram-positive microorganisms. *Escherichia coli* was sensitive to 25 mg of PVP-SrO, supporting earlier findings that metal-based nanoparticles may be more toxic to Gram-negative bacteria due to their thinner peptidoglycan layer and increased permeability to small molecules (Silhavy, Kahne, & Walker, 2010). Both PVP-SrO doses failed to kill Gram-positive *Staphylococcus aureus*. The thicker peptidoglycan layer may promote structural integrity and inhibit nanoparticle penetration, explaining this resistance (Otto, 2008).

Nanoparticle aggregation may have reduced absorption and antibiotic activity, causing

both bacterial strains to withstand 50 mg. Previous study has shown that nanoparticle clustering and surface charge interactions limit effectiveness at high nanoparticle concentrations (McDonnell & Russell, 1999).

Additionally, metal oxide nanoparticles may inhibit microbial development by creating reactive oxygen species (ROS), harming bacterial membranes, and inhibiting essential enzymes (Li, Plésiat, & Nikaido, 2015). Oxidative stress may be a crucial role in how PVP-SrO nanoparticles kill bacteria.

Staphylococcus aureus is resistant at both concentrations, therefore combination treatment may be needed. Traditional antibiotics mixed with metal-based nanoparticles may make bacteria more vulnerable to antibiotics and less prone to develop resistance (Yamaguchi, Nakamura, & Yamamoto, 2021). Future research should examine PVP-SrO nanoparticle synergy to boost antibiotic efficacy.

To ensure biomedical safety, PVP-SrO nanoparticles must pass cytotoxicity and biocompatibility tests. Metal-based nanoparticles show potential in antibacterial applications, but future study should carefully analyse their interactions with human cells and toxicity (Wang, Hu, & Yu, 2021).

CONCLUSIONS

This study examined PVP-SrO nanoparticles' antibacterial activity against *Staphylococcus aureus* and *E. coli* using Kirby-Bauer disc diffusion. PVP-SrO nanoparticles killed *E. coli* at 25 mg but not *S. aureus*. The resistance of both bacterial strains at 50 mg suggests that PVP-SrO nanoparticles' efficacy is species- and concentration-dependent. Since *Staphylococcus aureus* proved resistant to PVP-SrO at both doses, nanoparticle compositions must be optimised. Antibacterial efficiency decreases with nanoparticle concentration, as seen by resistance. The findings show that PVP-SrO nanoparticles may be beneficial as antimicrobial agents to improve bacterial susceptibility and limit resistance development, although they may work better with additional antibiotics. For safe biomedical usage, future research should examine PVP-SrO nanoparticle cytotoxicity, improve their formulations for bioavailability, and identify their antibacterial mechanisms. Nanoparticle-based antimicrobial medicines are vital to explore since antibiotic resistance is growing.

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