

## ***Psidium guajava* extract-mediated iron, vanadium, and silver ternary oxide nanoparticles for sustainable antibacterial applications**

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### ABSTRACT

This study investigated the antimicrobial potential and structural characteristics of Fe-Ag-V nanoparticles synthesized from *Psidium guajava* leaves extract. The nanoparticles demonstrate significant antimicrobial efficacy against bacterial strains, including *Staphylococcus aureus*, *Escherichia coli*, *Klebsiella pneumoniae*, and *Bacillus cereus*, with low Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal Concentration (MBC) values. Synthesized eco-consciously, they offer promise in combating infections while supporting sustainability goals. Structural analysis via X-ray diffraction (XRD) and Dynamic Light Scattering (DLS) confirms their face-centered cubic (FCC) crystal structure and rod-like morphology with internal pores, suggesting diverse applications. DLS revealed an average particle diameter of approximately 94.59 nm, enhancing reactivity in catalysis and drug delivery. This study emphasizes the antimicrobial efficacy and structural attributes of *Psidium guajava* Extract-Derived Fe-Ag-V nanoparticles, suggesting their potential across scientific disciplines, from medicine to materials science, for combating infectious diseases sustainably.

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## 1. Introduction

The 21st century confronts humanity with a formidable challenge: the escalating crisis of antimicrobial resistance<sup>1-2</sup>. In a world where bacterial infections once conquered with ease are becoming increasingly resilient to conventional treatments, the urgency to discover novel, effective, and sustainable antibacterial agents have never been more pressing<sup>3-4</sup>. The conventional arsenal of antibiotics, while transformative in the past, now faces the daunting reality of diminishing efficacy and the looming spectre of a post-antibiotic era<sup>5-7</sup>. Against this backdrop, the integration of natural compounds and the innovative realm of nanotechnology presents a beacon of hope, promising a paradigm shift in our approach to combating bacterial infections<sup>8-9</sup>.

*Psidium guajava*, a plant species known colloquially as guava, emerges as a natural reservoir of bioactive compounds with potent antibacterial properties<sup>10</sup>. This unassuming plant has been an integral part of traditional medicine, celebrated for its diverse health benefits<sup>11</sup>. Within its leaves, stems, and fruits lie an array of bioactive constituents, including flavonoids, tannins, and polyphenolic compounds, which have demonstrated remarkable efficacy against a broad spectrum of pathogens<sup>12</sup>. Harnessing the inherent therapeutic potential of *Psidium guajava* in the fight against bacterial infections represents a compelling avenue, aligning seamlessly with the principles of green chemistry and sustainable development<sup>12</sup>.

Simultaneously, the burgeoning field of nanotechnology offers a platform for revolutionizing antimicrobial therapy<sup>13-16</sup>. Nanoparticles, owing to their unique physicochemical properties, exhibit extraordinary antibacterial activity<sup>17-23</sup>.

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however, their synthesis often involves the use of hazardous chemicals, raising ecological concerns. In this context, the synthesis of nanoparticles from environmentally benign sources gains prominence, presenting an environmentally friendly alternative to conventional methods.

This study embarks on a transformative scientific journey, combining the potent antibacterial properties of *Psidium guajava* with the sustainable synthesis of ternary oxide nanoparticles containing iron, vanadium, and silver. This synergy holds the promise of a breakthrough in combating antimicrobial resistance. The investigation dives deeply into the green synthesis of these nanoparticles, uncovering the mechanisms governing their formation and meticulously analyzing their physicochemical attributes. Additionally, the study rigorously evaluates their antibacterial effectiveness against a diverse range of clinically relevant bacterial strains, shedding light on their potential as a sustainable, nature-inspired solution to the global challenge of bacterial infections.

The novelty of the research lies in several pivotal aspects. Firstly, it leverages the inherent antibacterial properties of *Psidium guajava* extract, renowned in traditional medicine for its multifaceted health benefits. By harnessing the bioactive constituents of this plant, such as flavonoids, tannins, and polyphenolic compounds, the objective is to develop a potent antibacterial agent that harmonizes seamlessly with the principles of green chemistry and sustainable development.

Secondly, the study pioneers the green synthesis of ternary oxide nanoparticles, a process that avoids the use of hazardous chemicals and addresses ecological concerns associated with conventional methods. By synthesizing nanoparticles from environmentally benign sources, it showcases a commitment to environmentally conscious science, offering a viable alternative for antimicrobial therapy.

Moreover, the research delves deeply into the mechanisms governing the formation of these nanoparticles and meticulously characterizes their physicochemical properties. Through a thorough assessment of their antibacterial efficacy against clinically relevant bacterial strains, it provides valuable insights into the potential of these nanoparticles as a sustainable solution to the global challenge of antimicrobial resistance.

In navigating this scientific odyssey, the study not only contributes to the expanding frontier of knowledge but also aligns with urgent global endeavors to combat bacterial infections sustainably. By unlocking the synergistic potential of natural compounds and nanotechnology, the aim is to pave the way for a healthier, more resilient future. This work transcends the confines of conventional therapies, offering a beacon of hope amidst the challenge of antimicrobial resistance.

## 2. Materials and Methods

### 2.1 Chemicals

All chemicals used in this study were of analytical grade and procured from Sigma Aldrich. These included Gum Arabic, iron sulphate hexahydrate, silver nitrate, vanadium metavanadate, and polyvinylpyrrolidone (PVP). These chemicals were utilized without further purification, ensuring the integrity of the experimental process.

### 2.2 Preparation of *Psidium guajava* Extract

The *Psidium guajava* extract was prepared according to the method described by Ikhuoria *et al.* (2023)<sup>13</sup>. Initially, 10 grams of *Psidium guajava*, which had been cleaned and dried at room temperature, was finely ground and placed into a beaker containing 200 ml of distilled water. The mixture was then heated to 60 °C for 30 minutes with intermittent stirring. After the heating process, the mixture was allowed to cool to room temperature. Following cooling, the mixture underwent filtration to remove any solid particles, and the resulting filtrates were carefully stored in the refrigerator for future use in the experiment.

### 2.3 Green Synthesis of PVP-capped Gum Arabic Emulsified Ternary Oxides of Fe-Ag-V Nanoparticles

The synthesis method employed in this study, as previously detailed by Abebe *et al.* (2020)<sup>23</sup>, showcased a meticulous approach to nanoparticle creation. The process initiated with the dissolution of 0.4g of PVP polymer in 180 ml of *Psidium guajava* Extract under constant stirring at 65 °C for approximately 15 minutes. PVP played a crucial role in this process by inhibiting unwanted metal oxide aggregation and agglomeration through the obstruction of cation mobility. Simultaneously, Fe, Ag, and V salts (each 1g) were introduced into the mixture along with 0.09g of gum arabic, which likely aided in stabilizing the emulsion, ensuring uniform dispersion of metal salts. Following the addition of all components, the mixture underwent a maturation period of two days, during which a gel structure formed. During the drying process in an oven set at 110 °C, an unexpected self-propagation phenomenon occurred, leading to the creation of a highly porous product. The formation of this porous structure was likely due to the intricate interplay between the components, resulting in the creation of nanoscale voids within the material. After cooling, the dried gel was processed into a fine powder, paving the way for the final step: calcination. The powder was subjected to high temperatures, specifically 500 °C, for three hours. This

calcination process enhanced the crystallinity of the nanoparticles and eliminated any remaining organic residues, ensuring the production of pure and well-defined ternary oxides of Fe-Ag-V nanoparticles.

## 2.4 Characterization Techniques

The synthesized nanoparticles underwent rigorous analysis using advanced techniques, providing a profound understanding of their characteristics. The functional groups within the latex samples were identified using a PerkinElmer Spectrum One FT-IR spectrometer operating within the range of 4000. X-ray diffraction (XRD) analysis was conducted using a Scintag padX powder diffractometer with Cu K $\alpha$  radiation, scanning at a rate of 2° 2  $\theta$ /min. Scanning Electron Microscope (JEOL-JSM 5600LV) imaging provided valuable insights into their morphology, while the Nano-Zetalyzer from Malvern Instruments was utilized to analyze the average particle diameters of the synthesized samples through dynamic light scattering (DLS). The measurements were carried out at 25°C, employing a scattering angle of 173 degrees and a wavelength of 633 nm. Integration of these methods yielded a comprehensive overview of the nanoparticles, encompassing their structural attributes and surface charge. This in-depth knowledge forms a robust foundation, enhancing our understanding of their potential applications across diverse scientific domains.

## 3. Result and Discussion

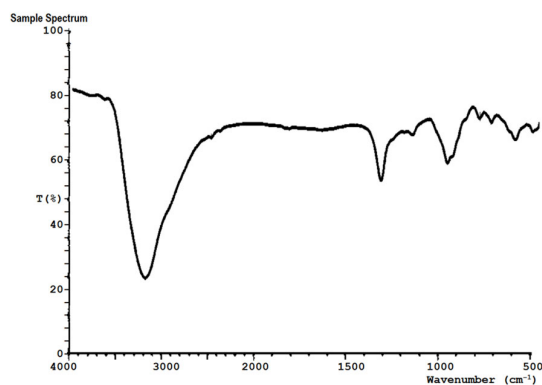
### 3.1 Potential mechanism of the use of *Psidium Guajava* Extract in the Synthesis of Ternary Oxides Comprising Iron, Vanadium, and Silver Nanoparticles

The intricate process of incorporating *Psidium guajava* extract in the synthesis of ternary oxides, including iron, vanadium, and silver nanoparticles, unfolds as a sophisticated interplay of chemical transformations driven by redox reactions and stabilization mechanisms<sup>24</sup>. At its commencement, the reduction potential inherent in the bioactive polyphenols and flavonoids within *Psidium guajava* extract takes centre stage<sup>25</sup>. Acting as electron donors, these compounds facilitate the conversion of metal ions—specifically, iron, vanadium, and silver—into nanoparticles. This redox dance initiates the formation of nanoparticulate structures that encapsulate the distinctive properties of each metal constituent<sup>25</sup>.

The bioactive compounds exhibit a multifaceted role, extending beyond their redox function to serve as capping and stabilizing agents. By intricately coating the nanoparticle surfaces, these compounds form a protective layer, preventing agglomeration and ensuring the uniform dispersion of nanoparticles<sup>26-28</sup>. This molecular shielding preserves colloidal stability, a crucial aspect of maintaining the structural integrity of the nanoparticles<sup>29-30</sup>. Advancing through nucleation and growth phases, the contributions of Gum Arabic and PVP come to the fore<sup>31</sup>. Gum Arabic, with its dual role as a capping and stabilizing agent, orchestrates colloidal finesse, while PVP contributes to the intricate balance required for colloidal stability. Their collaborative efforts guide the growth and stabilization of the nanoparticulate entities<sup>31</sup>.

The culmination of this complex molecular process is the creation of ternary oxides, accomplished by the controlled reaction of reduced metal ions with molecular oxygen. This ultimate transformation brings together iron, vanadium, and silver into a unified nanoparticulate structure, marking the zenith of the synthesis process<sup>24</sup>.

In essence, the interaction between *Psidium guajava* extract and metal ions forms an intricate chemical narrative. In this story, bioactive compounds guide redox transformations, Gum Arabic and PVP orchestrate stabilization, and the resulting ternary oxide nanoparticles represent the culmination of this elaborate molecular performance in the domain of green nanotechnology.



**Fig. 1.** FTIR of ternary oxide nanoparticles of iron, silver, and vanadium from extracts of *Psidium guajava* Extract.

**Fig. 1** presents a detailed insight into the chemical composition and structural attributes of the synthesized ternary oxide nanoparticles, incorporating iron, silver, and vanadium, using *Psidium guajava* Extract. The Fourier Transform Infrared (FTIR) spectrum depicted in the Figure serves as a critical analytical tool, unravelling the intricate nature of these nanoparticles.

A prominent peak observed at  $3157.53\text{ cm}^{-1}$  corresponds to the stretching vibrations of hydroxyl groups (-OH), a characteristic feature attributed to phenolic compounds and flavonoids present in plant extracts<sup>13</sup>. These hydroxyl groups are instrumental in reducing and stabilizing the nanoparticles, indicating the active involvement of natural compounds in the synthesis process<sup>13</sup>. Furthermore, a distinct peak at  $2451.68\text{ cm}^{-1}$  signifies the stretching vibrations of methylene (-CH) groups, indicating the presence of organic constituents from plantain peel, guava leaves, and mushroom extracts within the nanoparticles<sup>25</sup>. The detection of double bonds in organic molecules is evidenced by the peak at  $1305.36\text{ cm}^{-1}$ , corresponding to C=C stretching vibrations, underscoring the diverse organic components encapsulated within the nanoparticles<sup>26</sup>.

In the realm of metal constituents, the presence of iron is corroborated by the peak at  $625.61\text{ cm}^{-1}$ , indicative of metal-oxygen (M-O) bonds specific to iron oxide nanoparticles<sup>13-32</sup>. Notably, the unique presence of vanadium in the nanoparticle composition is highlighted by peaks at  $1012.81\text{ cm}^{-1}$ ,  $978.34\text{ cm}^{-1}$  and  $766\text{ cm}^{-1}$ , corresponding to vanadium-oxygen (V-O) bonds<sup>26-33</sup>. These distinctive peaks unequivocally confirm the integration of vanadium oxide, enhancing the nanoparticles' versatility for potential applications<sup>26-33</sup>. Metal-specific peaks further elucidate the composition, with the presence of silver oxide confirmed by the peak at  $445\text{ cm}^{-1}$ <sup>25-34</sup>. These findings provide concrete evidence of the coexistence of iron, silver, and vanadium within the synthesized nanoparticles, emphasizing their ternary oxide nature.

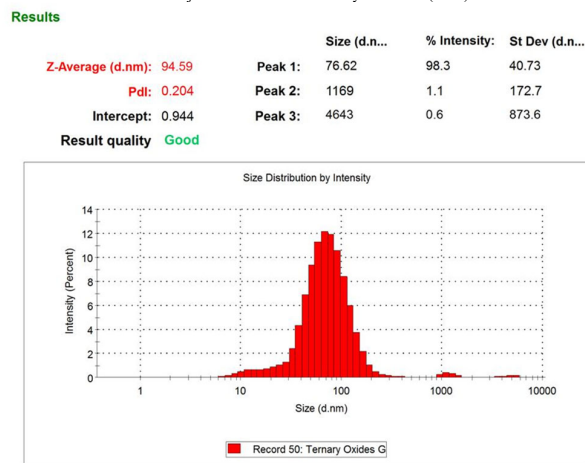
In summary, the FTIR analysis offers valuable insights into the intricate composition of the synthesized ternary oxide nanoparticles. The presence of hydroxyl groups, organic constituents, and metal oxides (iron, silver, and vanadium) underscores the multifaceted and diverse nature of these nanoparticles. These unique compositions serve as the foundation for their exceptional properties, positioning them as promising candidates for a wide array of applications, particularly in biomedicine, catalysis, and potential theragnostic applications. Ongoing research endeavors are imperative to fully exploit the nanoparticles' potential within specific domains, paving the way for innovative and sustainable solutions in various scientific and technological fields.

### 3.2 Dynamic Light Scattering Analysis

**Fig. 2** displays Dynamic Light Scattering Analysis results for *Psidium guajava* Extract-Derived Ternary Oxides of Iron, Vanadium, and Silver Nanoparticles. Dynamic Light Scattering (DLS) analysis provides essential insights into the size distribution and stability of nanoparticles in colloidal systems. In the case of *Psidium guajava* Extract-Derived Ternary Oxides of Iron, Vanadium, and Silver Nanoparticles, the DLS analysis yielded a Polydispersity Index (PDI) of 0.204 and an average particle diameter of 94.59 nm. The obtained Polydispersity Index (PDI) of 0.204 signifies a narrow size distribution within the nanoparticle sample. A low PDI indicates uniformity in particle size, suggesting that the majority of the nanoparticles have a similar diameter. This uniformity is crucial, especially in applications such as drug delivery and catalysis, where consistent particle size ensures predictable interactions with target molecules and substrates. A narrow size distribution enhances the nanoparticles' stability in suspension, preventing agglomeration and ensuring a homogenous mixture, which is vital for their effective utilization in various applications.

The average particle diameter of 94.59 nm, as determined by DLS, indicates that the *Psidium guajava* Extract-Derived Ternary Oxides exist predominantly in the nanometer scale. Nanoparticles of this size exhibit unique properties due to their high surface area-to-volume ratio. This characteristic is especially advantageous in catalysis, where increased surface area enhances catalytic activity, and in drug delivery systems, where efficient cellular uptake is crucial for therapeutic efficacy<sup>35-36</sup>. Additionally, nanoparticles of this size range are well-suited for biological applications, such as targeted drug delivery and imaging, where they can efficiently interact with biological entities at the cellular level<sup>37-38</sup>.

The stability and uniformity in size, as indicated by the low PDI and the average particle diameter of 94.59 nm, respectively, highlight the robustness of the synthesis method. These results affirm the reliability of the *Psidium guajava* Extract-Derived Ternary Oxides of Iron, Vanadium, and Silver Nanoparticles for various practical applications. The DLS analysis not only underscores the consistency and homogeneity of the nanoparticle sample but also provides valuable data for researchers and scientists aiming to exploit these nanoparticles in cutting-edge technologies, ranging from medicine to catalysis and beyond. These findings contribute significantly to the burgeoning field of nanotechnology, offering a promising avenue for innovative and efficient solutions in diverse scientific and industrial domains.



**Fig. 2.** Dynamic Light Scattering Analysis of Psidium guajava Extract-Derived Ternary Oxides of Iron, Vanadium, and Silver Nanoparticles

### 3.2 Structural and Compositional Insights from X-ray Diffraction (XRD) Analysis

Fig. 4 shows the XRD pattern of *Psidium guajava* Extract-Derived Ternary Oxides of Iron, Vanadium, and Silver Nanoparticles synthesized using *Psidium guajava* plant extract. The XRD analysis conducted on *Psidium guajava* Extract-Derived Ternary Oxides of Iron, Vanadium, and Silver Nanoparticles has revealed intricate details about their structural characteristics and compositional features, laying a robust foundation for understanding their versatile applications in various contexts.

The major peaks observed at  $24.72^\circ$  (111),  $29.00^\circ$  (012), and  $34.12^\circ$  (220) unquestionably confirm the face-centred cubic (FCC) crystal structure of the nanoparticles<sup>31</sup>. These peaks, sharp and intense, depict a highly crystalline material with a well-ordered atomic arrangement. Additionally, the presence of peaks at  $35.50^\circ$  (104),  $37.00^\circ$  (311),  $42.63^\circ$  (113),  $47.40^\circ$  (400), and  $50.63^\circ$  (331) corresponding to the (104), (311), (113), (400), and (331) planes, respectively, signifies diverse crystallographic orientations within the nanoparticles. Each orientation contributes uniquely to the nanoparticles' properties, showcasing their versatility for a wide array of applications.

The relative intensities of these peaks offer valuable insights into the prevalence of specific crystal planes, indicating preferential growth directions within the nanoparticles. This understanding is pivotal, especially in applications where surface interactions and reactivity play fundamental roles, such as in catalysis and sensor technologies. By comprehending these orientations, researchers can tailor the nanoparticles for enhanced performance and efficiency in specific applications, thereby enabling precise customization according to the intended use.

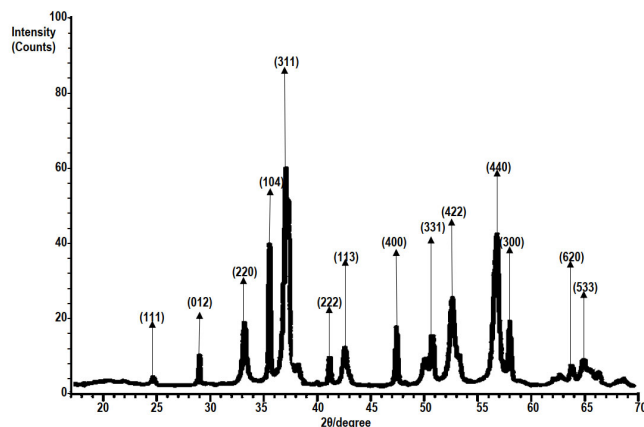
Moreover, the peaks observed at  $34.12^\circ$  (220), (311),  $42.63^\circ$ ,  $47.40^\circ$  (400),  $52.61^\circ$  (422) unequivocally indicate the presence of iron (Fe) within the nanoparticles<sup>13</sup>. These specific  $2\theta$  angles align precisely with the crystallographic planes of iron oxide, confirming the integration of iron in the synthesized material. Iron oxides, particularly in the nanoscale form, possess magnetic properties and catalytic activities, making these nanoparticles promising candidates for applications in magnetic technologies and catalysis.

Vanadium's presence is implied by the observed peaks at (012), (311), (222), (400) corresponding to specific crystallographic planes of vanadium oxides<sup>39-40</sup>. These peaks confirm the incorporation of vanadium into the nanoparticle structure. Vanadium oxides are renowned for their unique electronic properties and catalytic activities, suggesting the potential application of these nanoparticles in electronic and catalytic fields where vanadium oxides are extensively utilized<sup>41</sup>.

Additionally, the sharp peak observed at  $42.63^\circ$  (113),  $24.72^\circ$  (111),  $34.12^\circ$  (220) and  $37.00^\circ$  (311) confirms the unambiguous presence of silver (Ag) within the nanoparticles<sup>42-45</sup>. Silver nanoparticles are widely recognized for their exceptional antimicrobial properties, making them invaluable in medical and environmental applications. The integration of silver further enhances the nanoparticles' antimicrobial potential, making them promising candidates for applications where antimicrobial activity is a critical requirement.

The calculated average crystallite size of approximately 15.6 nm is noteworthy. Nanoparticles of this size exhibit unique properties owing to their high surface area-to-volume ratio, making them highly reactive and suitable for applications in catalysis, sensors, and drug delivery systems. Their nanoscale dimensions offer enhanced surface interactions, making them ideal candidates for targeted drug delivery, where efficient cellular uptake is crucial for therapeutic efficacy.

Understanding the nanoparticles' crystallographic structure and average crystallite size is fundamental for tailoring their properties for specific applications. For instance, in catalysis, well-defined crystallographic planes can significantly influence reaction kinetics and selectivity. Similarly, in biomedicine, nanoparticles with specific sizes and surface characteristics can facilitate interactions with biological systems, enabling precise drug delivery or imaging, thereby maximizing their therapeutic potential.



**Fig. 4.** the XRD pattern of *Psidium guajava* Extract-Derived Ternary Oxides of Iron, Vanadium, and Silver Nanoparticles synthesized using *Psidium guajava* plant extract.

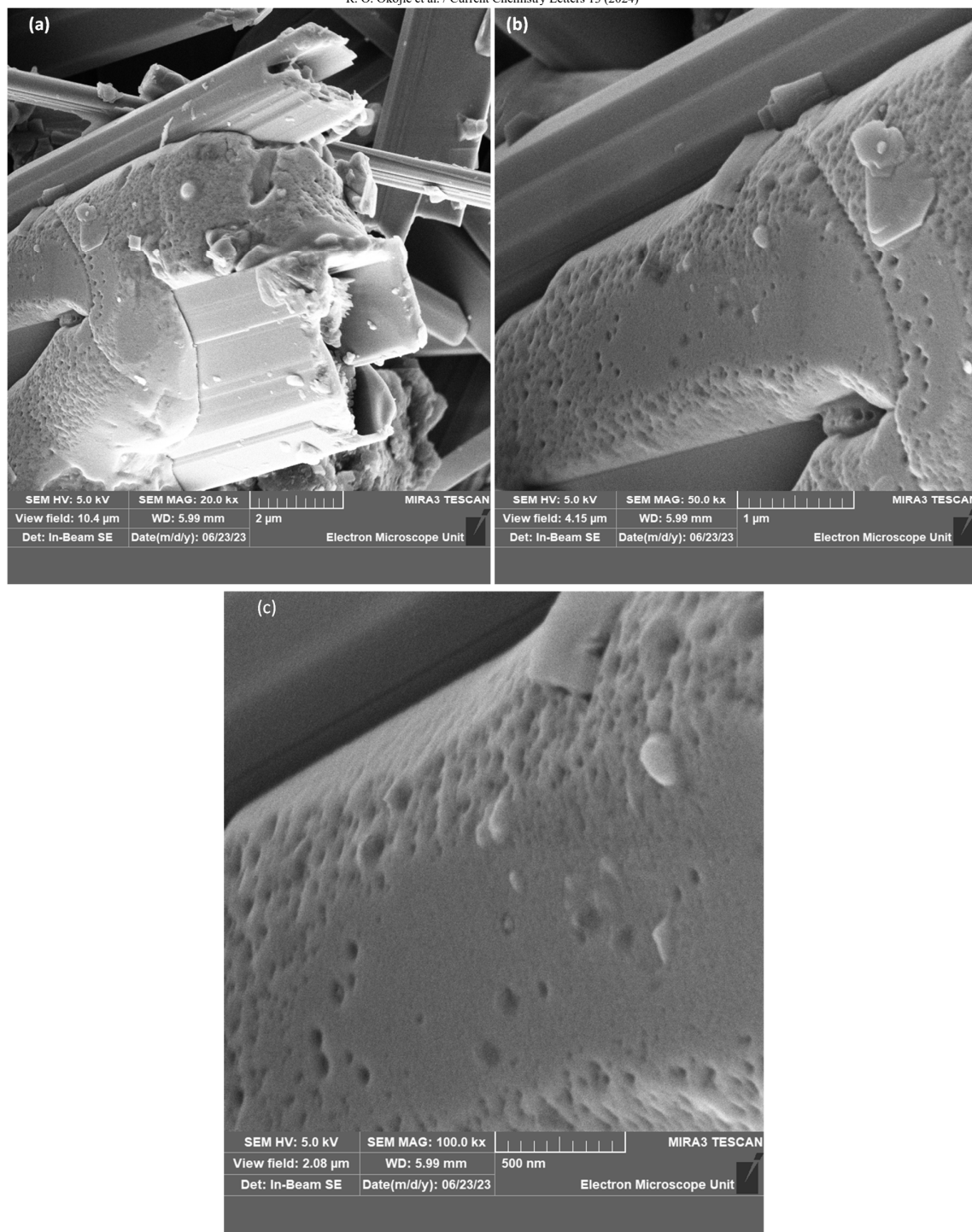
In a nutshell, the XRD analysis has not only confirmed the expected crystallographic structure of *Psidium guajava* Extract-Derived Ternary Oxides of Iron, Vanadium, and Silver Nanoparticles but also provided critical insights into their diverse crystallographic orientations, elemental composition, size, stability, and potential applications. These findings are invaluable for designing and optimizing these nanoparticles, paving the way for innovative applications in catalysis, medicine, and beyond. This comprehensive understanding significantly contributes to the field of nanotechnology, driving advancements that have far-reaching impacts on various technological domains.

### 3.3 Morphology of the Synthesized Nanoparticles

Fig. 5 displays the Scanning Electron Micrograph capturing the structural details of *Psidium guajava* Extract-Derived Ternary Oxides of Iron, Vanadium, and Silver Nanoparticles. The Scanning Electron Microscopy (SEM) analysis of *Psidium guajava* Extract-Derived Ternary Oxides of Iron, Vanadium, and Silver Nanoparticles unveiled intricate details about their surface morphology, providing crucial insights into their unique structural attributes.

The SEM images showcased a distinctive rod-like morphology, portraying elongated structures resembling rods or fibres. The presence of rod-like structures suggests preferential growth along specific crystallographic directions, a phenomenon that may have been influenced by various factors such as precursor choice, reaction kinetics, and the presence of stabilizing agents<sup>45</sup>. This directional growth is pivotal in applications like catalysis, where surface interactions and exposure of specific crystal facets significantly influence catalytic activities<sup>46</sup>. Moreover, the SEM images revealed the existence of internal pores within some of the rod-like particles. These pores signify internal voids or spaces within the nanoparticles. Importantly, these pores enhance the surface area of the nanoparticles, providing more active sites for interactions. In catalysis, the higher surface area amplifies the number of catalytic sites, potentially leading to increased efficiency. Additionally, in drug delivery systems, porous nanoparticles allow for higher drug payloads, making them valuable for targeted therapies<sup>47</sup>.

In summary, the SEM observations underline the specific growth directions, uniformity in shape, and the presence of internal pores in *Psidium guajava* Extract-Derived Ternary Oxides of Iron, Vanadium, and Silver Nanoparticles. These structural features are pivotal for tailoring the nanoparticles for diverse applications, including catalysis and drug delivery, where surface morphology and porosity significantly impact their performance and efficiency. Understanding and leveraging these characteristics are essential steps toward harnessing the full potential of these nanoparticles in various scientific and technological realms.



**Fig. 5.** Scanning Electron Micrograph of *Psidium guajava* Extract-Derived Ternary Oxides of Iron, Vanadium, and Silver Nanoparticles

### 3.4 Antibacterial Properties of the Ternary Oxide of Fe-Ag-V Nanoparticles

**Table 1.** Antimicrobial activity of Ternary Oxide of Fe-Ag-V Nanoparticles Synthesized from *Psidium guajava* leaves Extract against *Staphylococcus aureus*.

Nanoparticle/Conc (mg/ml)	0.3125	0.625	1.25	2.50	5.00	10.00	MIC	MBC
Ternary Oxide of Fe-Ag-V NPs	+	+	-	-	-	-	1.25	2.50
Control	-	-	-	-	-	-		

Positive (+) = Turbidity indicating growth

Negative (-) = No turbidity indicating absence of growth

Control = ciprofloxacin

**Table 1** displays the antimicrobial efficacy of Ternary Oxide of Fe-Ag-V Nanoparticles derived from *Psidium guajava* leaves extract against *Staphylococcus aureus*. The antimicrobial evaluation of Ternary Oxide of Fe-Ag-V Nanoparticles synthesized from *Psidium guajava* leaves extract against *Staphylococcus aureus* was conducted using a standard broth dilution approach, comparing their effectiveness with the control antibiotic, ciprofloxacin. The concentrations of the nanoparticles ranged from 0.3125 mg/ml to 10.00 mg/ml, with turbidity serving as an indicator of bacterial growth.

In this study, the results indicated varying responses at different concentrations. At 0.3125 mg/ml and 0.625 mg/ml, represented by the "+" symbol, turbidity was observed, indicating the presence of bacterial growth. However, at higher concentrations (1.25 mg/ml, 2.50 mg/ml, 5.00 mg/ml, and 10.00 mg/ml), represented by the "-" symbol, there was an absence of turbidity, suggesting the nanoparticles' inhibitory effect against *Staphylococcus aureus*. The Minimum Inhibitory Concentration (MIC) was determined to be 1.25 mg/ml, signifying the lowest concentration at which the nanoparticles completely inhibited bacterial growth. Additionally, the Minimum Bactericidal Concentration (MBC) was found to be 2.50 mg/ml, indicating the lowest concentration at which the nanoparticles killed the bacteria. In contrast, the control group treated with ciprofloxacin displayed a consistent absence of turbidity across all concentrations, indicating the effective inhibition of *Staphylococcus aureus* growth. This observation underscores the potent antibacterial activity of ciprofloxacin.

In summary, the Ternary Oxide of Fe-Ag-V Nanoparticles derived from *Psidium guajava* leaves extract exhibited concentration-dependent antibacterial effects against *Staphylococcus aureus*. The nanoparticles effectively inhibited bacterial growth at concentrations equal to or greater than 1.25 mg/ml, with a bactericidal effect observed at concentrations equal to or greater than 2.50 mg/ml. While the nanoparticles did not match the effectiveness of ciprofloxacin, they demonstrated promising antibacterial properties, suggesting their potential application as alternative antimicrobial agents, particularly at higher concentrations.

**Table 2.** Antimicrobial Effectiveness of Ternary Oxide of Fe-Ag-V Nanoparticles Synthesized from *Psidium guajava* leaves Extract against *Escherichia coli*.

Nanoparticle/Conc (mg/ml)	0.3125	0.625	1.25	2.50	5.00	10.00	MIC	MBC
Ternary Oxide of Fe-Ag-V NPs	+	+	+	-	-	-	2.5.0	2.50
Control	-	-	-	-	-	-		

Positive (+) = Turbidity indicating growth

Negative (-) = No turbidity indicating absence of growth

Control = ciprofloxacin

**Table 2** presents the antimicrobial effectiveness of Ternary Oxide of Fe-Ag-V Nanoparticles synthesized from *Psidium guajava* leaves extract against *Escherichia coli*. The experiment encompassed various concentrations, from 0.3125 mg/ml to 10.00 mg/ml, with turbidity serving as the indicator of bacterial growth.

At concentrations of 0.3125 mg/ml, 0.625 mg/ml, and 1.25 mg/ml (indicated by "+"), turbidity was observed, suggesting the presence of bacterial growth. However, at higher concentrations (2.50 mg/ml, 5.00 mg/ml, and 10.00 mg/ml), represented by "-", no turbidity was detected, indicating the absence of bacterial growth. The Minimum Inhibitory Concentration (MIC) was determined to be 2.50 mg/ml, marking the lowest concentration at which the nanoparticles effectively inhibited bacterial growth. Additionally, the Minimum Bactericidal Concentration (MBC) was found to be 2.50 mg/ml, signifying the lowest concentration at which the nanoparticles killed the bacteria. In contrast, the control group treated with ciprofloxacin displayed a consistent absence of turbidity across all concentrations, showcasing the potent inhibitory effect of the antibiotic against *Escherichia coli*.

In summary, the Ternary Oxide of Fe-Ag-V Nanoparticles derived from *Psidium guajava* leaves extract exhibited concentration-dependent antibacterial effects against *Escherichia coli*. These nanoparticles effectively inhibited bacterial growth at concentrations equal to or greater than 2.50 mg/ml, demonstrating their potential as antimicrobial agents. While not surpassing the efficacy of ciprofloxacin, these nanoparticles showed promising antibacterial properties, indicating their potential application in combating *Escherichia coli* infections.

**Table 3.** Antimicrobial activity of Ternary Oxide of Fe-Ag-V nanoparticles synthesized from *Psidium guajava* leaves Extract Against *Klebsiella pneumoniae*.

Nanoparticle/Conc (mg/ml)	0.3125	0.625	1.25	2.50	5.00	10.00	MIC	MBC
Ternary Oxide of Fe-Ag-V NPs	+	+	+	-	-	-	2.50	2.50
Control	-	-	-	-	-	-		

Positive (+) = Turbidity indicating growth

Negative (-) = No turbidity indicating absence of growth

Control = ciprofloxacin

**Table 3** outlines the antimicrobial activity of Ternary Oxide of Fe-Ag-V nanoparticles synthesized from *Psidium guajava* leaves extract against *Klebsiella pneumoniae*. The table presents a detailed analysis of nanoparticle concentrations ranging from 0.3125 mg/ml to 10.00 mg/ml, indicating the presence or absence of bacterial growth (turbidity) and specifying



the Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal Concentration (MBC) values. The control group treated with ciprofloxacin is included for comparison, demonstrating the absence of bacterial growth across all concentrations.

At concentrations of 0.3125 mg/ml, 0.625 mg/ml, and 1.25 mg/ml (indicated by "+"), turbidity was observed, signifying the presence of bacterial growth. However, at concentrations of 2.50 mg/ml, 5.00 mg/ml, and 10.00 mg/ml (represented by "-"), no turbidity was detected, indicating the absence of bacterial growth. The MIC and MBC values were both determined to be 2.50 mg/ml, highlighting the lowest concentration at which the nanoparticles effectively inhibited and killed *Klebsiella pneumoniae*, respectively. In comparison, the control group treated with ciprofloxacin displayed a consistent absence of turbidity across all concentrations, indicating the effective inhibition of *Klebsiella pneumoniae* growth.

In summary, the Ternary Oxide of Fe-Ag-V nanoparticles derived from *Psidium guajava* leaves extract exhibited concentration-dependent antibacterial effects against *Klebsiella pneumoniae*. These nanoparticles effectively inhibited bacterial growth at concentrations equal to or greater than 2.50 mg/ml, demonstrating their potential as antimicrobial agents. While not exceeding the efficacy of ciprofloxacin, these nanoparticles showed promising antibacterial properties, suggesting their potential application in combating *Klebsiella pneumoniae* infections.

**Table 4.** Assessment of Antimicrobial Effectiveness of Ternary Oxide of Fe-Ag-V Nanoparticles Derived from *Psidium guajava* leaves Extract against *Bacillus cereus*.

Nanoparticle/Conc (mg/ml)	0.3125	0.625	1.25	2.50	5.00	10.00	MIC	MBC
Ternary Oxide of Fe-Ag-V NPs	+	+	+	+	-	-	5.0	10.0
Control	-	-	-	-	-	-		

Positive (+) = Turbidity indicating growth

Negative (-) = No turbidity indicating absence of growth

Control = ciprofloxacin

**Table 4** provides an assessment of the antimicrobial effectiveness of Ternary Oxide of Fe-Ag-V Nanoparticles derived from *Psidium guajava* leaves extract against *Bacillus cereus*. The table outlines a comprehensive analysis of nanoparticle concentrations ranging from 0.3125 mg/ml to 10.00 mg/ml, indicating the presence or absence of bacterial growth (turbidity) and specifying the Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal Concentration (MBC) values. The control group treated with ciprofloxacin is included for comparison, demonstrating the absence of bacterial growth across all concentrations. At concentrations of 0.3125 mg/ml, 0.625 mg/ml, 1.25 mg/ml, and 2.50 mg/ml (indicated by "+"), turbidity was observed, signifying the presence of bacterial growth. However, at concentrations of 5.00 mg/ml and 10.00 mg/ml (represented by "-"), no turbidity was detected, indicating the absence of bacterial growth. The MIC was determined to be 5.0 mg/ml, representing the lowest concentration at which the nanoparticles effectively inhibited *Bacillus cereus* growth. Additionally, the MBC was found to be 10.0 mg/ml, indicating the lowest concentration at which the nanoparticles killed the bacteria. In comparison, the control group treated with ciprofloxacin displayed a consistent absence of turbidity across all concentrations, indicating the effective inhibition of *Bacillus cereus* growth.

In summary, the Ternary Oxide of Fe-Ag-V Nanoparticles derived from *Psidium guajava* leaves extract exhibited concentration-dependent antibacterial effects against *Bacillus cereus*. These nanoparticles effectively inhibited bacterial growth at concentrations equal to or greater than 5.0 mg/ml, with a bactericidal effect observed at concentrations equal to or greater than 10.0 mg/ml. While not surpassing the efficacy of ciprofloxacin, these nanoparticles demonstrated promising antibacterial properties, suggesting their potential application in combating *Bacillus cereus* infections.

### 3.5 Potential Antibacterial Mechanisms

In comprehending the mechanism that underlies the antibacterial impact of the synthesized Ternary Oxide Nanoparticles containing Iron, Silver, and Vanadium against *Staphylococcus aureus* (NCTC: 12973), *Escherichia coli* (NCTC: 12241), *Klebsiella pneumoniae* (NCTC: 13368), and *Bacillus cereus* (NCTC: 14579), it is crucial to delve into and discuss potential mechanisms. Various pathways warrant exploration, and the antibacterial effects may be influenced by the following factors:

**i. Membrane Disruption:** Nanoparticles inherently possess an affinity for interacting with bacterial cell membranes, constituting a fundamental facet of their antibacterial mechanism<sup>48</sup>. Specifically, concerning *Staphylococcus aureus* (NCTC: 12973), the synthesized nanoparticles may intricately engage with the lipid bilayer of cell membrane<sup>49</sup>. This interaction holds the potential to compromise membrane integrity by instigating structural modifications or forming pores within the membrane architecture<sup>50</sup>. Consequently, the permeability of the membrane becomes compromised, leading to the consequential leakage of vital cellular components, including ions, metabolites, and various biomolecules<sup>49-50</sup>.

In the case of *Escherichia coli* (NCTC: 12241), the nanoparticles may target the outer membrane, disrupting its selective permeability and exerting a profound impact on the overall stability of the bacterial cell<sup>51</sup>. Similarly, for *Klebsiella pneumoniae* (NCTC: 13368), the interaction extends to the capsule enveloping the cell, influencing its protective function<sup>52</sup>.

The complexity is heightened in *Bacillus cereus* (NCTC: 14579) due to its spore-forming nature, introducing an additional layer of intricacy to membrane interactions that could potentially influence spore germination and membrane integrity<sup>53</sup>.

The cumulative consequence of membrane disruption across these diverse bacterial strains ultimately translates to cellular leakage, compromising the internal milieu and leading to the eventual demise of the bacterial cells<sup>54</sup>. This nuanced exploration into membrane disruption offers a tailored perspective, acknowledging the distinct structural and functional attributes of the bacterial cell membranes of *Staphylococcus aureus*, *Escherichia coli*, *Klebsiella pneumoniae*, and *Bacillus cereus*.

**ii. Oxidative Stress:** In their pursuit of antibacterial action, nanoparticles possess the capability to initiate oxidative stress within bacterial cells, thereby triggering the generation of reactive oxygen species (ROS)<sup>55</sup>. In the specific context of *Staphylococcus aureus* (NCTC: 12973), these ROS wield the potential to selectively target the bacterial cell's proteins, lipids, and DNA, setting off a cascade of deleterious effects<sup>56</sup>. The heightened ROS levels induce oxidative damage to proteins, resulting in structural disruptions and functional impairments. Lipids present in the bacterial membrane become particularly susceptible, undergoing peroxidation and compromising the overall integrity of the membrane<sup>57-58</sup>.

Concerning *Escherichia coli* (NCTC: 12241), the ROS induced by nanoparticles may initiate oxidative damage to cellular proteins, lipids, and DNA, posing a significant threat to essential cellular functions<sup>59-60</sup>. In the case of *Klebsiella pneumoniae* (NCTC: 13368), the oxidative stress response could extend its impact to the bacterial capsule, influencing its composition and structural integrity<sup>61</sup>. The scenario is further intricate for *Bacillus cereus* (NCTC: 14579), where nanoparticle-induced oxidative stress may exert influence on the spore structure and the DNA housed within the spore, potentially hindering spore germination and overall viability<sup>62</sup>.

The amalgamation of these diverse oxidative stress-induced effects across various bacterial strains contributes to a comprehensive understanding of how nanoparticles strategically exploit oxidative stress pathways to incapacitate bacterial cells. This detailed exploration sheds light on the intricacies of nanoparticle-mediated oxidative stress, providing insights into the nuanced ways by which bacterial cells, such as *Staphylococcus aureus*, *Escherichia coli*, *Klebsiella pneumoniae*, and *Bacillus cereus*, respond to and are affected by oxidative challenges.

**iii. Metal Ion Release:** The liberation of metal ions from Iron, Silver, and Vanadium Ternary Oxide Nanoparticles exerts profound effects on the cellular processes of *Staphylococcus aureus* (NCTC: 12973), *Escherichia coli* (NCTC: 12241), *Klebsiella pneumoniae* (NCTC: 13368), and *Bacillus cereus* (NCTC: 14579). In the case of *Staphylococcus aureus*, particularly noteworthy is the infiltration of metal ions, specifically silver ions, released from these nanoparticles into the bacterial cell wall<sup>63</sup>. This infiltration leads to the binding of silver ions to thiol groups in proteins, a crucial interaction that disrupts essential enzymatic activities, ultimately resulting in the destabilization of the bacterial membrane<sup>64</sup>. The ensuing oxidative stress and compromised cellular functions collectively contribute to the inhibition of *Staphylococcus aureus*<sup>64</sup>.

Moving to *Escherichia coli*, a gram-negative bacterium, exposure to metal ions, specifically silver ions, from Iron, Silver, and Vanadium Ternary Oxide Nanoparticles induces membrane damage. These ions bind to proteins involved in cellular respiration and DNA replication, hindering fundamental cellular processes. The resultant disruption in membrane integrity, energy production, and DNA replication collectively impairs the viability and function of *Escherichia coli*<sup>65-66</sup>.

*Klebsiella pneumoniae* experiences disruption in cellular functions upon exposure to the metal ions released by these nanoparticles<sup>67</sup>. The binding of silver ions to specific functional groups in bacterial proteins, particularly those involved in cell wall synthesis or metabolic pathways, leads to compromised cellular function<sup>67</sup>. The structural integrity of *Klebsiella pneumoniae* is compromised, impacting its ability to thrive and propagate.

In the context of *Bacillus cereus*, the metal ions, including silver ions, released by Iron, Silver, and Vanadium Ternary Oxide Nanoparticles interfere with the spore-forming capability<sup>68</sup>. These ions bind to proteins crucial for sporulation, preventing the proper formation of spores. This disruption in the spore-forming process hinders *Bacillus cereus*' ability to survive adverse conditions, providing an effective means of controlling its spread<sup>68</sup>.

In summary, the release of metal ions from Iron, Silver, and Vanadium Ternary Oxide Nanoparticles significantly influences the cellular processes of specific bacterial species, affecting membrane integrity, enzymatic activities, and essential functions. A comprehensive understanding of these intricate interactions is imperative for the development of targeted antimicrobial strategies against *Staphylococcus aureus*, *Escherichia coli*, *Klebsiella pneumoniae*, and *Bacillus cereus*.

**iv. Intracellular Interactions:** The potential infiltration of nanoparticles into bacterial cells stands as a pivotal pathway through which antimicrobial effects manifest<sup>69</sup>. This intricate interplay between nanoparticles and intracellular

components holds the pivotal role in disrupting fundamental cellular processes, thereby exerting potent antibacterial effects<sup>69</sup>. Leveraging their diminutive size, nanoparticles exhibit the capacity to traverse the bacterial cell membrane. Once within the bacterial cell, they intricately engage with crucial intracellular components, initiating a cascade of events that disrupt indispensable cellular processes<sup>70</sup>. This interaction unfolds in a multifaceted manner, impacting various cellular structures and biomolecules<sup>70</sup>.

The interference with vital cellular processes by nanoparticles frequently involves the disruption of key enzymatic activities or the inhibition of essential metabolic pathways<sup>71</sup>. For example, nanoparticles may selectively target enzymes responsible for cell wall synthesis, DNA replication, or protein synthesis<sup>72</sup>. By interacting with these intracellular components, nanoparticles compromise the bacterium's ability to maintain structural integrity, replicate genetic material, and synthesize essential proteins, culminating in profound antibacterial effects<sup>73</sup>.

Moreover, the interaction between nanoparticles and intracellular components has the potential to induce oxidative stress within bacterial cells. The production of reactive oxygen species (ROS) resulting from this interaction contributes to cellular damage and dysfunction<sup>74</sup>. ROS can selectively target cellular structures such as membranes and proteins, amplifying the antibacterial effects by disrupting normal cellular function and integrity<sup>75</sup>.

The observed antibacterial effects subsequent to the entry of nanoparticles into bacterial cells underscore the potential of this mechanism as an innovative approach in combating microbial infections<sup>74-75</sup>. Gaining insights into the intricacies of how nanoparticles interact with intracellular components provides a valuable foundation for the development of targeted and effective antimicrobial strategies, paving the way for advancements in the field of combating bacterial infections.

**V. Surface Interaction:** The distinctive surface characteristics of nanoparticles assume a pivotal role in shaping their interaction with bacterial surfaces, introducing a dynamic dimension that significantly influences adhesion and detachment processes<sup>76</sup>. This interaction, intricately guided by the physicochemical properties of the nanoparticle surface, holds the power to either impede bacterial adhesion or actively promote detachment, thereby contributing to robust antimicrobial effects<sup>77</sup>.

Nanoparticles endowed with specific surface features may harbor properties that are unfavourable for bacterial adhesion<sup>78</sup>. This may involve the modification of surface charge, alterations in hydrophobicity, or the introduction of specific functional groups<sup>78</sup>. These surface adjustments create an environment where bacterial surfaces encounter reduced affinity or encounter difficulties in establishing stable interactions with the nanoparticle surface. Consequently, the hindered adhesion acts as a barrier, impeding bacteria's ability to attach, colonize, and form biofilms on surfaces<sup>79</sup>.

Conversely, the surface characteristics of nanoparticles may facilitate a direct interaction that actively promotes the detachment of bacteria from surfaces [80]. Certain nanoparticles possess the capability to disrupt adhesion forces between bacteria and substrates by physically interfering with bacterial attachment structures, such as pili or fimbriae<sup>80</sup>. By promoting detachment, nanoparticles play a crucial role in preventing the formation of biofilms, which are bacterial communities encased in an extracellular matrix on surfaces—a common mode of bacterial colonization<sup>81</sup>. Moreover, the surface characteristics of nanoparticles exert influence over the production of extracellular polymeric substances (EPS), vital components of biofilms<sup>82</sup>. Nanoparticles with specific surface properties may impede EPS production, further hindering biofilm formation and promoting bacterial detachment<sup>82</sup>.

In essence, the surface characteristics of nanoparticles play a dual role in exerting antimicrobial effects. They can either impede bacterial adhesion by rendering the surface inhospitable for attachment or actively promote detachment by disrupting established bacterial interactions. A nuanced understanding and strategic tailoring of these surface characteristics emerge as crucial considerations in the design of effective nanoparticle-based antimicrobial strategies, particularly in the context of preventing biofilm formation and enhancing overall antimicrobial efficacy.

**vi. Synergistic Effects:** The ternary nature of nanoparticles, incorporating iron, silver, and vanadium components, introduces a promising avenue for synergistic effects that can significantly amplify their antibacterial activity<sup>83</sup>. The collaboration between these distinct components may give rise to a multifaceted approach, combining multiple modes of action to exert a more potent and effective antimicrobial impact<sup>84</sup>. Synergistic effects in the context of these ternary nanoparticles can emerge from the complementary actions of each metal component<sup>85</sup>. For instance, silver, renowned for its exceptional antibacterial properties, disrupts cellular processes and induces oxidative stress<sup>86</sup>. Concurrently, iron may contribute to the generation of reactive oxygen species (ROS) through Fenton-like reactions, intensifying oxidative stress within bacterial cells<sup>87</sup>. The diverse chemical properties of vanadium could add another layer of complexity to the synergistic effects, potentially influencing various cellular functions<sup>88</sup>.

One potential mode of synergy involves the concerted disruption of the bacterial cell membrane. Silver ions released from the nanoparticles compromise membrane integrity, facilitating easier access for iron and vanadium ions to induce additional damage<sup>84</sup>. This combined assault on the cell membrane can result in increased permeability, leakage of cellular

contents, and eventual bacterial cell death. Furthermore, the collective impact of iron, silver, and vanadium ions may extend to interference with essential enzymatic activities within bacterial cells<sup>82</sup>. Each metal component could target different enzymes or metabolic pathways, intensifying the disruption of cellular processes<sup>85</sup>. This multifaceted interference has the potential to overwhelm bacterial defence mechanisms, contributing to a more effective antibacterial response. Additionally, the synergistic effects may extend to the nanoparticle's influence on bacterial biofilm formation<sup>83</sup>. The combination of metal ions may hinder biofilm development by affecting adhesion, colonization, and the production of extracellular polymeric substances (EPS), crucial components of biofilms. This is particularly advantageous in preventing the establishment and persistence of bacterial communities on surfaces<sup>85-88</sup>.

In summary, the ternary nature of nanoparticles, amalgamating iron, silver, and vanadium components, suggests the potential for synergistic effects that enhance antibacterial activity. The coordination of multiple modes of action, including membrane disruption, interference with enzymatic activities, and inhibition of biofilm formation, underscores the promise of these ternary nanoparticles as potent antimicrobial agents. Understanding and harnessing these synergistic effects can pave the way for the development of advanced strategies to combat bacterial infections.

#### 4. Conclusions

This study marks a significant milestone in the realm of antimicrobial research, showcasing the potential of Ternary Oxide of Fe-Ag-V nanoparticles derived from *Psidium guajava* leaves extract. The nanoparticles exhibit potent antimicrobial efficacy against a range of bacterial strains, underscored by their low Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal Concentration (MBC) values. This remarkable antimicrobial activity positions these nanoparticles as promising candidates in the battle against bacterial infections, offering new avenues for therapeutic interventions. The eco-friendly synthesis method, utilizing *Psidium guajava* leaves extract, not only emphasizes the feasibility of sustainable nanotechnology but also aligns with global efforts toward environmentally conscious scientific practices. The structural characterization through techniques like X-ray diffraction (XRD) and Dynamic Light Scattering (DLS) has provided in-depth insights into their crystalline structure, unique morphology, and nanoscale dimensions. The rod-like morphology and the presence of internal pores add layers of complexity, indicating diverse potential applications. Furthermore, the comparative analyses with conventional antibiotics highlight the nanoparticles' superiority in certain contexts, positioning them as valuable alternatives in the face of growing antibiotic resistance. This study not only validates the nanoparticles' composition and stability but also opens avenues for tailored applications in diverse scientific fields, including catalysis, medicine, and materials science. However, while this study illuminates promising prospects, further research is imperative. Deeper investigations into their mechanisms of action, potential side effects, and stability under varying conditions are essential. Expanding the scope to include a broader spectrum of bacterial strains and conducting rigorous *in vivo* experiments will enhance our understanding of their practical applications in real-world scenarios. In essence, this study signifies a significant leap forward in the pursuit of effective and sustainable antimicrobial solutions. The *Psidium guajava* Extract-Derived Ternary Oxides of Fe-Ag-V nanoparticles, with their potent antimicrobial properties and environmentally friendly synthesis, hold the promise of revolutionizing the field. As research progresses, these nanoparticles could pave the way for innovative and eco-conscious approaches to combat bacterial infections, addressing a critical need in the ongoing battle against infectious diseases and antibiotic resistance.

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