

Short Communication

Antioxidant, anti-collagenase, and antibacterial activities of *Fucus vesiculos*us silver nanoparticles

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Abstract

Fucus vesiculosus is an alga with high fucoxanthin, phlorotannin, fucoidan, sterol, and astaxanthin. The silver nanoparticles of F. vesiculosus (AgNPs-Fv) are expected to have high antioxidant, anti-collagenase, and antibacterial activities. The aim of this study was to characterize the distribution and size of AgNPs-Fv and determine their antioxidant, anti-collagenase, and antibacterial activities. The distribution and size of AgNPs-Fv were measured using particle size analyzer (PSA) analysis. The nanoparticle compound and their functional groups were characterized using a scanning electron microscope (SEM) and Fourier-transform infrared spectroscopy (FTIR), respectively. The antioxidant activity of AgNPs-Fv was determined using a 1.1-diphenyl-2-picrylhydrazyl (DPPH) assay, while the anti-collagenase activity was examined using the spectrophotometric method. The antibacterial activity was assessed using an inhibition zone test, minimum inhibitory concentration (MIC), and minimum bactericidal concentration (MBC). Results indicated that the AgNPs-Fv had a dominant volume of about 86.3% with a diameter of 113.3 nm. SEM analysis revealed the spherical AgNPs with sizes between 27 and 54 nm. The FTIR analysis of the AgNPs-Fv absorption band at 1,046 cm⁻¹ demonstrated the bond between the Ag metal and the O-H hydroxyl group. The antioxidant activity of AgNPs-Fv was higher than F. vesiculosus extract (24.23±3.55 mg/L vs 47.45±3.16 mg/L). AgNPs-Fv also had a higher anti-collagenase activity compared to F. vesiculosus extract (66.74±6.352 mg/L vs 145.1±6.326 mg/L). The inhibition zone diameter of AgNPs-Fv was greater than F. vesiculosus extract. The MIC and MBC of AgNPs-Fv were 18.75 and 18.75 ppm, while F. vesiculosus extract was 37.5 and 75 ppm, respectively. These results suggested that AgNPs of F. vesiculosus had higher antioxidant, anti-collagenase, and antibacterial activities than F. vesiculosus extract alone.

Keywords: Fucus vesiculosus, AgNPs, antibacterial, anti-collagenase, antioxidant

Introduction

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N atural sources from the sea have been shown to have high biological activity, such as the *Fucus* vesiculosus alga, which has many health benefits [1]. *F. vesiculosus* is mainly found in subtropical areas such as the north of the Baltic Sea, the Swedish Bothanian coast, and northeast of the Bothanian coast [1,2]. It has many active compounds such as fucoxanthin, chlorophyll a and c pigments, xanthophylls, and β -carotene [2]. In addition, it has a very abundant fucoidan compound that acts as an antioxidant to counteract reactive oxygen species (ROS) and induces endogenous antioxidant defense activities, including glutathione transferase, superoxide dismutase (SOD), catalase, and glucose-6-phosphate dehydrogenase [3]. Other active compounds

in *F. vesiculosus* extracts, such as fucoxanthin, astaxanthin, and phlorotannin (polyphenolic), also have many biological activities. Fucoxanthin has antiaging and antioxidant activities, while astaxanthin protects cells from photooxidation and is considered a strong antioxidant [4]. Phlorotannin has several activities to prevent skin aging, such as anti-melanogenic and anti-collagenase properties. The phlorotannin in *F. vesiculosus* has very high antioxidant activity and potentially prevents several diseases, such as viral infections, bacterial infections, aging-related conditions, and cardiovascular disease [5]. Some bioactive compounds in the *F. vesiculosus* exhibit great potential in European cosmetic industries [4]. Several cosmetics made from algae have been marketed in many forms, such as moisturizers, sunscreens, and anti-aging serums [6].

Most of the high-value biochemicals in several natural plants have been well-studied, including polyphenolic or phlorotannin. These compounds are the main contributor to *F*. *vesiculosis*'s antioxidant capacity [7]. Polyphenols have a strong antioxidant activity similar to ascorbic acid, vitamin E, β -carotene, and selenium [6]. Polyphenols have various biological effects such as antibacterial, antiviral, antiradical, antiallergic, antiaging, protection against ROS, and reduction of α -tocopheryl radicals [6].

Some of the active compounds in *F. vesiculosus* can be developed with a nanoparticle system to increase the effectiveness of their usage. Nanoparticles have an effective delivery system for active compounds from plants [8]. One of the methods to synthesize nanoparticles is green synthesis, which is environmentally friendly, non-toxic, and safe for human health. The green synthesis of silver nanoparticles refers to the synthesis of silver or argentum nanoparticles (AgNPs) using plants as bio-reductors [9,10]. In addition, silver is an inorganic antibacterial agent that is non-toxic and has been safely used for centuries and it can kill various disease-causing microorganisms [11]. A previous study has characterized AgNPs from several algae, such as *Palmaria palmata* and *Gelidium spinosum*, resulting in particle sizes of 185.5 and 107.1 nm, respectively [12,13]. Although the main compound of *F. vesiculosus* as an antioxidant has been characterized [7], there is no research on the antioxidant activity and characterization of AgNP from *F. vesiculosus* (AgNPs-Fv). The aim of this study was to determine the antioxidant, anticollagenase, and antibacterial activities of AgNPs-Fv. It is expected that AgNPs-Fv have higher antioxidant, anti-collagenase, and antibacterial biological activities than non-nanoparticle *F. vesiculosus* extract.

Methods

Preparation of Fucus vesiculosus extract

The extraction method of *F. vesiculosus* was adapted from Rajivgandhi *et al.* [14], with several modifications. Briefly, the powdered samples were dissolved in distilled water at a ratio of 1:20. Then, samples were homogenized by stirring and heated at 35°C. The samples were centrifugated at 4,000 relative centrifugal force (RCF) for 15 minutes at 4°C to collect the supernatant.

Preparation of silver nanoparticles from F. vesiculosus (AgNPs-Fv)

A total of 40 ml *F. vesiculosus* filtrate was mixed with 40 ml of 1 mM $AgNO_3$ solution (1:1) and left in a dark room at 60°C for two hours until the color of the solution changed from white to pale yellow. Then, the solution was centrifuged at 4,000 RCF for 30 minutes at 20°C. The pellets were dried at 45°C for 24 hours in the oven [14].

Particle size analyzer (PSA) analysis

PSA analysis was used to determine the distribution and size of AgNPs-Fv. The samples were vortexed and dissolved in aquadest (1 mg in 10 mL). Next, the solution was placed into a tube that could hold a maximum of 15 mm of solution height. After that, the Malvern Zetasizer Nano Particle Analyzer (Malvern Panalytical Ltd, Malvern, UK) was used to measure the sample's diameter distribution [15].

Characterization of nanoparticle compounds

The AgNPs-Fv was characterized using a scanning electron microscope (SEM) (TM 3000 Hitachi with SwiftED 3000 X-ray Microanalysis, Japan). The stub was covered with nanoparticles using double-sided tape. The powder was conditioned to be electrically conductive with a tuft of a thin

layer of platinum and a current strength of 30 mA. The image was captured with the appropriate magnification at 10 kV electron voltage [16].

Characterization of functional groups

The functional groups of AgNPs-Fv were characterized using Fourier-transform infrared spectroscopy (FTIR) (Shimadzu, Shimadzu Corp, Kyoto, Japan). A total of 100 mg of potassium bromide (KBr) and 2 mg of powdered AgNPs-Fv were combined. The powder mixture was dried in a vacuum freeze dryer for one day. Subsequently, the powder mixture was exposed to 100 scans using a Spectrum One Spectrometer (Perkin Elmer, Norwalk, CT, USA) at a wavelength range of 400–4,000 cm⁻¹ [17].

Antioxidant activity assay

To determine the antioxidant activity, a 1.1-diphenyl-2-picrylhydrazyl (DPPH) assay was used, as previously described by Qarani *et al.* [18], with some modifications. A series concentration of *F. vesiculosus* extract and AgNPs-Fv was prepared at 50, 100, 150, and 200 ppm concentrations. Ascorbic acid was used as a standard solution by preparing it at 2, 4, 6, 8, and 10 ppm. Then, 500 μ L of each sample was mixed with 500 μ L of DPPH solution. Each sample was replicated three times and incubated in a dark place at room temperature for 30 minutes. The absorbance of each sample was measured using a UV-Vis spectrophotometer (Bio-Rad Laboratories, Inc., California, US) at 517 nm. The DPPH inhibition was then calculated using the formula provided elsewhere [18].

Collagenase inhibitory assay

The collagenase inhibitory test was conducted using a method modified by Geeta *et al.* [19] and Riani *et al.* [20]. A total of 20 μ L AgNPs-Fv at 50, 100, 150, 200, and 250 ppm were dissolved in 20 μ L of collagenase enzyme from *Clostridium histolyticum*, then mixed with 20 μ L of tricine buffer. The solution was incubated for 30 minutes at 37°C. Furthermore, 100 μ L of bovine collagen substrate was added to the incubated solution and incubated at 37°C for 10 minutes. Then, 400 μ L of 5% trichloroacetic acid (TCA) and 200 μ L of Folin Ciocalteu reagent were added. Each sample was replicated three times.

The blank solution consisted of $40 \ \mu\text{L}$ buffer as a substitute for the sample, and it was treated the same as the sample, then the enzyme solution was replaced with $20 \ \mu\text{L}$ distilled water. A standard tyrosine solution was prepared by reacting $40 \ \mu\text{L}$ of tyrosine as a substitute for the samples and enzymes, then treated the same as the samples. Furthermore, the collagenase enzyme was prepared but reacted without an inhibitor. All samples were determined for absorbance using a UV-Vis spectrophotometer (Bio-Rad Laboratories, Inc., California, USA) at 578 nm three times. The percentage of enzyme inhibition was calculated using the following formula: collagenase inhibition percentage = 1 - (the activity of collagenase with inhibitor / theactivity of collagenase without inhibitor).

Antibacterial activity assay

The disc diffusion method, adapted from Bhuyar *et al.* [21], was used against *Staphylococcus aureus* and *Escherichia coli*. The suspension of *S. aureus* and *E. coli* (10⁷–10⁸ CFU/mL) was inoculated on 0.1 mL of nutrient agar (NA) medium and leveled. The paper discs were immersed in AgNPs-Fv, *F. vesiculosus* extract, chloramphenicol (positive control) and distilled water (negative control). Six different concentrations of AgNPs-Fv, *F. vesiculosus* extract, and chloramphenicol were used (2.34, 4.68, 9.37, 18.75, 37.5, and 75.0 ppm). The clear zone surrounding the paper disc served as an indicator of the level of bacterial inhibition, and this zone was quantified using a digital caliper. This assay was repeated three times.

The minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) were also determined [22]. The MIC assay was conducted using the microdilution method in a sterile 96-well plate containing nutrient broth (NB) media, based on the previous method [23], with several modifications. A total of 200 μ L of the AgNPs-Fv was added to the first well. Then, a multiple dilution was carried out by consecutively transferring 100 μ L of the test solution from the first well. The microbial suspension was adjusted with 100 μ L of 0.5 Mc Farland solution to all wells and incubated at 37°C for 24 hours.

Statistical analysis

Antioxidant, anti-collagenase, and antibacterial data were expressed as mean \pm standard deviation (SD).

Results

Characterization of silver nanoparticles of F. vesiculosus (AgNPs-Fv)

The results of the characterization of the AgNPs-Fv showed that the particle size was >100 nm (**Figure 1**). AgNPs-Fv had a dominant volume percentage of about 86.3% with a diameter of 113.3 nm and the smallest volume percentage of about 13.7% with a size of 400 nm.



Figure 1. Characterization of AgNPs size distribution of silver nanoparticle of *Fucus vesiculosus* (AgNPs-Fv) using particle size analyzer (PSA) analysis.

The observation of SEM revealed that the constituents of AgNPs-Fv were C, O, Na, Mg, Cl, and K. Generally, a lighter white color in SEM image was produced by constituent metal elements with a higher atomic number than by those with a lower atomic number. The results of the SEM characterization demonstrated the formation of spherical particle surfaces. The uniformly distributed spherical particles were observed (**Figure 2A**). The particle shape had sharp corners, as presented in **Figure 2B**. When the particles formed angular plates with different sizes ranging from 2.2 to 3.2 µm, their shape became more apparent (**Figure 2B**). The scanning electron microscopy with energy-dispersive X-ray spectroscopy (SEM-EDX) spectrum obtained from AgNPs-Fv indicated that the peaks correspond to several elements (**Figure 2C**). The elemental composition of AgNPs-Fv was comprised of carbon (C), oxygen (O), sodium (Na), magnesium (Mg), chlorine (Cl), and potassium (K) (**Figure 2D**). AgNPs showed strong absorption spectra at 2.1 keV, characteristic of elemental gold.

AgNPs-Fv at 1,046 cm⁻¹ demonstrate the bond between the Ag metal and the O-H hydroxyl group (**Figure 3**). It demonstrates that the absorption band signals the formation of silver nanoparticles. The flavonoid compounds in the *F. vesiculosus* extract that contain functional groups such as O-H, N-H, C=C, and C-H, serve as reductants of silver ions during the nanoparticle synthesis process. Furthermore, a C-N amine functional group stabilizing silver nanoparticles is present in the AgNPs-Fv.



Figure 2. The characterization of particle size and shape from AgNPs of *F. vesiculosus* using a scanning electron microscope (SEM) at (A) 5 μ m and (B) 40 μ m. (C) SEM with energy-dispersive X-ray (EDX) spectrum analysis. (D) The elemental composition of AgNPs of *F. vesiculosus* from the results of EDX analysis. CK: carbon K electron shell; CIK: chlorine K electron shell; KK: potassium K electron shell; MgK: magnesium K electron shell; NaK: sodium K electron shell; OK: oxygen K electron shell; ZAF: ZAF correction (atomic number effect/Z, self-absorption effect/A, fluorescent effect/F); Wt(%): weight %; At%: atomic %.



Figure 3. Spectrum and peak value of Fourier-transform infrared spectroscopy (FTIR) from $AgNO_3$ (blue line), AgNPs from *Fucus vesiculosus* (AgNPs-Fv) (red line), and *Fucus vesiculosus* extract alone (black line).

Antioxidant activity of silver nanoparticles of *F. vesiculosus* (AgNPs-Fv)

Antioxidant activity test suggested that the AgNPs-Fv had a powerful antioxidant activity compared to the *F. vesiculosus* extract (**Table 1**). The IC_{50} value of the AgNPs-Fv (24.23±3.55 ppm) was lower than the *F. vesiculosus* extract (47.45±3.16 ppm). However, antioxidant activity in ascorbic acid (2.604±0.08 ppm) was higher compared to the AgNPs-Fv (**Table 1**).

Sample Concentration (ppm) IC₅₀, mean±SD (ppm)* Category [22] AgNPs-Fv Very strong 24.23±3.55 50 100 150 200 250 F. vesiculosus extract Very strong 47.45±3.16 50 100 150 200 250 Ascorbic acid 2 2.604 ± 0.08 Very strong 4 6 8 10

Table 1. Antioxidant activity of *Fucus vesiculosus* silver nanoparticles (AgNPs-Fv), *F. vesiculosus* extract, and ascorbic acid

* The antioxidant activity was measured in triplicate

Inhibition of collagenase of silver nanoparticles of *F. vesiculosus* (AgNPs-Fv) The results of collagenase inhibition revealed that AgNPs-Fv (IC₅₀: 66.74±6.35 ppm) had a stronger anti-collagenase activity than *F. vesiculosus* extract (IC₅₀: 145.1±6.33 ppm) (Table 2). This suggested that AgNPs-Fv increased the inhibitory activity of the collagenase enzyme, possibly due to its strong antioxidant contents. AgNPs-Fv is expected to be a good source for antiaging treatments because the natural antioxidant content in *F. vesiculosus* acts as an inhibitor of the collagenase enzyme. However, the ascorbic acid still had the strongest anti-collagenase activity (1.69±0.002 ppm) than others (Table 2).

| Sample | Concentration (ppm) | IC ₅₀ , mean±SD (ppm)* | Category [23] |
|------------------------|---------------------|-----------------------------------|---------------|
| AgNPs-Fv | 50 | 66.47±6.35 | Strong |
| | 100 | | |
| | 150 | | |
| | 200 | | |
| | 250 | | |
| F. vesiculosus extract | 50 | 145.10±6.33 | Medium |
| | 100 | | |
| | 150 | | |
| | 200 | | |
| | 250 | | |
| Ascorbic acid | 2 | 1.69±0.002 | Very strong |
| | 4 | | |
| | 6 | | |
| | 8 | | |
| | 10 | | |

Table 2. Collagenase inhibitory activity of *Fucus vesiculosus* silver nanoparticles (AgNPs-Fv), *F. vesiculosus* extract, and ascorbic acid

*The anti-collagenase activity was measured in triplicate.

Antibacterial activity of silver nanoparticles of *F. vesiculosus* (AgNPs-Fv)

The AgNPs-Fv against *E. coli* had the largest inhibition zone diameter $(9.19\pm4.35 \text{ mm})$ compared to AgNPs-Fv against *S. aureus* $(8.33\pm3.45 \text{ mm})$, *F. vesiculosus* extract against *E. coli* $(7.26\pm4.68 \text{ mm})$ and *F. vesiculosus* against *S. aureus* $(7.47\pm1.61 \text{ mm})$ (**Table 3**). These four samples were categorized as having moderate antibacterial activity [24]. In contrast, chloramphenicol exhibited strong antibacterial activity, with average inhibition zone diameters of $13.52\pm5.06 \text{ mm}$ against *E. coli* and $13.23\pm4.24 \text{ mm}$ against *S. aureus* (**Table 3**).

| Sample | Inhibition zone, mean±SD (mm) | Category [26] |
|--------------------------------------|-------------------------------|---------------|
| F. vesiculosus extract + E. coli | 7.26±4.68 | Moderate |
| F. vesiculosus extract + $S. aureus$ | 7.47±1.61 | Moderate |
| AgNPs-Fv + $E. coli$ | 9.19±4.35 | Moderate |
| AgNPs-Fv + S. aureus | 8.33±3.45 | Moderate |
| Chloramphenicol + E. coli | 13.52±5.06 | Strong |
| Chloramphenicol + S. aureus | 13.23±4.24 | Strong |
| Distilled water + <i>E. coli</i> | 0.00 ± 0.00 | - |
| Distilled water + <i>S. aureus</i> | 0.00±0.00 | - |

Table 3. Antibacterial activity of *Fucus vesiculosus* silver nanoparticles (AgNPs-Fv), *F. vesiculosus* extract, and chloramphenicol against *Escherichia coli* and *Staphylococcus aureus*

The MIC of AgNPs-Fv against *E. coli* and *S. aureus* was the same, 18.75 ppm. The MIC of *F. vesiculosus* extract against *E. coli*, and *S. aureus* was 18.75, and 37.5 ppm, respectively. The MBC of AgNPs-Fv against *E. coli* and *S. aureus* was also the same at 18.75 ppm. The MBC of *F. vesiculosus* extract against *E. coli* and *S. aureus* was 75 ppm and 37.5 ppm, respectively. This indicated that the MBC of AgNPs-Fv was lower than that of the other groups.

Discussion

The present study found that *F. vesiculosus* extract has successfully synthesized silver nanoparticles. The size of nanoparticles with an effective transmission system is 10-1,000 nm. The nanoparticle diameter should be 200-400 nm [13,25]. Another study revealed that the distribution of five AgNPs on the surface of *Gelidium amansii* was morphologically uniform, with spherical AgNPs with sizes between 27-54 nm. AgNPs' biosynthesis with hydrodynamic diameters ranging from 27-54 nm. Biosynthesis of AgNPs using seaweed extracts has also been reported to produce hydrodynamic diameters between 20-95 nm [26, 27].

A previous study showed that FTIR spectra of the *C. roxburghii* extract revealed the presence of N-H, O-H, C=C, and C-H groups, proving that hydroxyl and amine groups were present in the plant extracts. Flavonoid compounds are present in both groups [28]. The AgNPs of *F. vesiculosus* have a powerful antioxidant activity compared to the *F. vesiculosus* extract, as indicated by a lower IC₅₀ value. The smaller the IC₅₀ value indicated a stronger antioxidant activity. Molyneux [22] reported that IC50 values <50 indicate very strong antioxidant activity, IC₅₀ values of 50–100 indicate strong antioxidant activity, IC₅₀ values of 100–150 indicate moderate antioxidant activity, and IC₅₀ values of 150–200 indicate weak antioxidant activity.

The IC₅₀ value of AgNPs-Fv was greater than *F. vesiculosus* extract, which means that the synthesis of AgNPs-Fv was successful in increasing antioxidant activity. The polyphenol compounds can coat AgNPs to produce stable nanoparticles and increase antioxidant activity [29]. The flavonoid compounds can inhibit free radicals through the hydrogen-atom transfer (HAT) mechanism. The O-H bonds of the flavonoid hydroxyl groups will separate, and the H atoms will be transferred to free radicals [30]. In addition, nanoparticles have a larger surface area when compared to the extract. Thus, the potential for nanoparticles to bind to the enzyme's active site is bigger, and the large surface area of nanoparticles has a great ability in its activity [31].

AgNPs-Fv have a stronger anti-collagenase activity compared to the *F. vesiculosus* extract. Thus, the smaller the IC₅₀ value, the better the anti-collagenase activity. Findrianny *et al.* [32] showed that IC₅₀ values (<50) indicate very strong, (50–100) indicate strong, (101–150) indicate moderate, and (>150) indicate weak activity. Natural antioxidant compounds such as terpenoids and flavonoids have a synergistic effect in inhibiting the collagenase enzyme, which can cause wrinkles on the skin [33]. MMP enzyme is responsible for degrading and decreasing collagen production in the skin. The lack of collagen fibers in skin tissue stimulates the formation of wrinkles. However, the hydroxyl group (OH) in the polyphenolic content of *F. vesiculosus*, along with the active side of the Zn ion contained in collagenase, can bind to polyphenolic compounds, thus preventing the substrate from digesting collagenase [34].

AgNPs-Fv and *E. coli* showed the highest inhibition zone diameter $(9.19\pm4.35 \text{ mm})$ compared to the combined AgNPs of *F. vesiculosus* and *S. aureus* $(8.33\pm3.45 \text{ mm})$, *F. vesiculosus* extracts and *E. coli* $(7.26\pm4.68 \text{ mm})$, and *F. vesiculosus* extracts and *S. aureus* $(7.47\pm1.61 \text{ mm})$.

These four samples were in the moderate category [35]. This study found that the diameter of the inhibition zone in AgNPs-Fv was greater than *F. vesiculosus* extract (**Table 3**). These results indicated that the nanoparticle successfully increased antibacterial activity. Silver nanoparticles can attach to the surface of cell membranes, penetrate the cell, and interact with sulfur compounds on the membrane surface and inside bacterial cells. Thus, it can interfere with cell viability, cell membrane permeability, cellular respiration, and degradation of bacterial DNA, resulting in bacterial lysis [36].

The fucoidan compound might be responsible for the inhibitory and antibacterial effects of *F. vesiculosus* extract. This compound consists of three main components, including sugar, sulfate, and uronic acid, which can act as an antibacterial agent. Fucoidan protects brown algae from microorganisms [24]. The present study also revealed that chloramphenicol's highest inhibition zone diameter was a positive control (**Table 3**). Chloramphenicol is a broad-spectrum antibiotic against Gram-positive, Gram-negative, and anaerobic bacteria. Thus, it can inhibit protein synthesis by binding to the 50S ribosomal subunit [37]. Meanwhile, the distilled water was not detected to have an antibacterial activity. Distilled water cannot inhibit the growth of Gram-positive and Gram-negative bacteria [38].

Gram-positive and negative bacteria have different cell wall compositions, which cause different potential antibacterial resistance. Gram-negative bacteria have a more complex cell wall structure and consist of lipopolysaccharide, periplasm, and peptidoglycan membrane. So, it has a strong defense to block the antibacterial compounds. Meanwhile, Gram-positive bacterial cells comprise a simpler peptidoglycan structure than Gram-negative bacteria, so antibacterial compounds can easily enter the cell and damage the bacterial cell wall, causing the bacteria to lyse and die [39]. These differences can affect the inhibition zone value and diffusion speed of antibacterial compounds in the paper disc test.

F. vesiculosus contains several active compounds, such as fucoidans, flavonoids, and tannins, which also have the potential as an antibacterial agent. The flavonoids are hydroxylated phenolic compounds with C6-C3 units bound to aromatic rings, which can inhibit nucleic acid synthesis, form bonds with proteins and extracellular cell walls, and degrade cell membranes. Meanwhile, tannins are a group of polymeric phenolic substances that can inhibit extracellular enzymes, precipitate proteins, and degrade polypeptides, which can interfere with bacterial cell wall synthesis [40].

AgNPs-Fv was a potent antibacterial agent; even the smallest concentration was able to inhibit bacterial growth and kill the bacteria (**Table 3**). A study suggested that silver nanoparticles continuously release ions in the microbial water environment [41]. Due to their small size and large surface area, silver nanoparticles exhibit a strong antibacterial effect even in small concentrations. In addition, silver nanoparticles can produce reactive oxygen species (ROS) that can destroy bacterial cell walls, bind to respiratory enzymes, and inhibit DNA replication, ultimately killing bacteria.

AgNPs exhibited antibacterial properties through various mechanisms, such as producing reactive oxygen species (ROS), unwanted interactions between AgNPs and bacterial cell wall DNA, and the AgNPs' release of silver ions. For example, antimicrobial properties of variously shaped nanoparticles led by silver ions. In this instance, the bacterial cell wall and silver nanoparticle ions interacted to inhibit one or more respiratory enzymes, which aided in producing reactive oxygen species and ultimately caused cell damage [11,42,43]. This study offers valuable insight into the discovery of novel antioxidant, anti-collagenase, and antibacterial agents. However, this study only tested the antibacterial effect of AgNPs-Fv on two species of bacteria. Future studies should test the antibacterial effect of AgNPs-Fv on other bacterial species and strains to validate their antibacterial effect. Further characterization of AgNPs-Fv is also recommended.

Conclusion

Particle size analyzer (PSA) analysis showed that AgNPs-Fv has a dominant volume of about 86.3% with a diameter of 113.3 nm. SEM analysis found the spherical AgNPs with sizes between 27 and 54 nm. The FTIR analysis of AgNPs-Fv at 1,046 cm⁻¹ demonstrates the bond between the Ag metal and the O-H hydroxyl group. AgNPs-Fv had more powerful antioxidant, anti-

collagenase, and antibacterial activities than *F. vesiculosus* extract. Further research is needed, including the in vivo toxicity and bioactivity properties of AgNPs from *F. vesiculosus*.

Ethics approval

Not required.

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Competing interests

All the authors declare that there are no conflicts of interest.

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Underlying data

Derived data supporting the findings of this study are available from the corresponding author on request.

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