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Development and In Vitro Antibacterial Evaluation of a Nanotechnology-Enhanced Ginger (Zingiber officinale) Emulgel Incorporated with Silver Nanoparticles Against Staphylococcus Aureus

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Abstract

Antimicrobial resistance has become a major global public health concern because of the increasing prevalence of multidrug-resistant bacterial pathogens. The growing limitations of conventional antibiotics have intensified interest in plant-derived antibacterial agents and nanotechnology-based drug delivery systems. *Zingiber officinale* (ginger) possesses established antibacterial properties because of bioactive compounds such as gingerols and shogaols, while silver nanoparticles exhibit broad-spectrum antimicrobial activity through membrane disruption and oxidative stress induction. This study aimed to develop and evaluate a nanotechnology-enhanced ginger emulgel incorporated with silver nanoparticles against *Staphylococcus aureus* through in vitro antibacterial assessment.

Ethanolic ginger extract was formulated into emulgels containing ginger extract alone, silver nanoparticles alone, and a combined ginger–silver nanoparticle formulation. Physicochemical characteristics including pH, viscosity, spreadability, and centrifugation stability were evaluated to determine formulation suitability for topical administration. Antibacterial activity was assessed using the Kirby–Bauer disk diffusion method against *Staphylococcus aureus*, with vancomycin used as the positive control.

The ethanolic ginger extract yielded 89.84% and demonstrated the presence of 6-gingerol (0.023 mg/g) through ultraviolet–visible spectrophotometric analysis. All formulations exhibited acceptable physical stability without visible phase separation. The ginger extract formulation demonstrated greater antibacterial activity (10.17 mm) compared with the combined ginger–silver nanoparticle formulation (9.08 mm) and silver nanoparticles alone (8.55 mm). Statistical analysis revealed significant differences among treatment groups ($p < 0.001$). The findings indicate that ginger extract served as the principal contributor to antibacterial activity, whereas silver nanoparticle incorporation did not enhance antibacterial efficacy. Ginger-based emulgels may represent a promising alternative topical antibacterial formulation against *Staphylococcus aureus*.

Keywords: Antibacterial Activity, Emulgel, Ginger Extract, Nanotechnology, Silver Nanoparticles, *Staphylococcus aureus*, *Zingiber officinale*

Introduction

Antimicrobial resistance (AMR) has emerged as a major global public health concern because of the increasing prevalence of multidrug-resistant microorganisms that compromise the effectiveness of conventional antimicrobial therapies [1]. The continuous emergence of resistant bacterial strains has significantly increased morbidity, mortality, and healthcare expenditures worldwide. Among clinically important pathogens, *Staphylococcus aureus* remains one of the most frequently implicated organisms associated with skin infections, soft tissue infections, wound contamination, and hospital-acquired infections [2]. The increasing resistance of *Staphylococcus aureus* to commonly prescribed antibiotics has intensified the search for alternative therapeutic approaches capable of improving antibacterial efficacy while minimizing adverse effects and microbial resistance development [1], [2].

Medicinal plants have gained increasing scientific interest because of their therapeutic potential and relatively favorable safety profiles. Among medicinal plants, *Zingiber officinale*, commonly known as ginger, has been extensively utilized in traditional and modern medicine because of its antibacterial, antioxidant, anti-inflammatory, and pharmacological properties [4]. Ginger contains biologically active compounds such as gingerols, shogaols, paradols, and zingerone, which have been reported to inhibit bacterial growth through membrane disruption, inhibition of bacterial enzymes, and interference with bacterial metabolic pathways [5]. Previous investigations have demonstrated the antibacterial activity of ginger extracts against several pathogenic microorganisms, including *Staphylococcus aureus*, *Escherichia coli*, and *Bacillus subtilis*, suggesting its potential as a natural antibacterial agent [6].

Nanotechnology has likewise gained substantial attention in pharmaceutical and biomedical applications because of its ability to improve drug delivery, stability, and antimicrobial performance. Silver nanoparticles (AgNPs) possess broad-spectrum antibacterial properties and have demonstrated effectiveness against both Gram-positive and Gram-negative bacteria [3]. The antibacterial activity of silver nanoparticles is primarily attributed to their ability to penetrate bacterial cell membranes, induce oxidative stress through reactive oxygen species generation, disrupt intracellular metabolic processes, and interfere with bacterial DNA replication [7]. Previous studies have reported enhanced antibacterial effects when silver nanoparticles are combined with plant-derived phytochemicals, thereby supporting the hypothesis of synergistic antimicrobial interactions [8].

Topical drug delivery systems have increasingly incorporated plant-based compounds and nanoparticles to improve therapeutic outcomes. Among these systems, emulgels have emerged as an effective pharmaceutical dosage form because they combine the advantages of emulsions and gels, resulting in improved spreadability, stability, patient acceptability, and enhanced drug penetration [9]. Emulgels are particularly suitable for incorporating hydrophobic bioactive compounds such as ginger phytochemicals and nanoparticle-based agents because they facilitate sustained release and improve topical retention [10].

Although ginger extract and silver nanoparticles individually demonstrate established antibacterial activity, limited studies have investigated their combined incorporation into emulgel formulations against *Staphylococcus aureus*. Furthermore, while previous investigations generally report enhanced antibacterial performance following nanoparticle incorporation, contradictory findings may occur because of formulation incompatibility, nanoparticle aggregation, concentration-dependent interactions, and altered release behavior within pharmaceutical delivery systems [8]. Therefore, this study aimed to develop and evaluate a nanotechnology-enhanced ginger (*Zingiber officinale*) emulgel incorporated with silver nanoparticles and determine its in vitro antibacterial activity against *Staphylococcus aureus* using the Kirby–Bauer disk diffusion method.

Materials and Methods

Study Design

A laboratory-based experimental research design was employed to develop and evaluate a nanotechnology-enhanced ginger (*Zingiber officinale*) emulgel incorporated with silver nanoparticles (AgNPs) and determine its in vitro antibacterial activity against *Staphylococcus aureus*. The study involved ethanolic extraction of ginger rhizomes, emulgel formulation, physicochemical characterization, and antibacterial evaluation through the Kirby–Bauer disk diffusion assay.

Research Setting

Laboratory procedures involving extraction, emulgel preparation, and formulation development were conducted at the laboratory facilities of AMYA Polytechnic College, Inc., Davao City, Philippines. Antibacterial testing, ultraviolet–visible (UV–Vis) spectrophotometric analysis, and viscosity determination were performed in a testing and research laboratory in Davao City, Philippines, using standardized laboratory procedures to ensure reproducibility and reliability of experimental findings.

Collection and Preparation of Plant Material

Fresh rhizomes of *Zingiber officinale* were procured from local commercial sources in Davao City, Philippines. The collected ginger rhizomes were thoroughly washed under running water to remove adhering contaminants and impurities. The rhizomes were subsequently sliced into smaller portions and prepared for ethanolic extraction.

Preparation of Ethanolic Ginger Extract

The prepared ginger rhizomes were subjected to ethanolic extraction to isolate phytochemical constituents associated with antibacterial activity, particularly gingerols and shogaols. Ethanol was selected as the extraction solvent because of its ability to recover polar and moderately nonpolar bioactive compounds effectively.

Following extraction, filtration was conducted to separate the liquid extract from plant residues. The filtrate was concentrated to obtain crude ethanolic ginger extract suitable for formulation and antibacterial evaluation.

The percentage yield of the ginger extract was determined using the following equation:

$$\text{Percentage Yield (\%)} = \frac{Final\ Weight\ of\ Extract}{Initial\ Weight\ of\ Raw\ Material} \times 100$$

Ultraviolet–Visible Spectrophotometric Analysis of 6-Gingerol

Ultraviolet–visible (UV–Vis) spectrophotometric analysis was conducted to confirm the presence and concentration of 6-gingerol, a principal phytochemical constituent responsible for the antibacterial activity of ginger. Analytical testing was performed in a testing and research laboratory to quantify the concentration of 6-gingerol present in the ethanolic extract.

Preparation of Emulgel Formulation

The emulgel formulation was prepared through the incorporation of emulsion and gel phases in a 1:1 ratio following standard pharmaceutical formulation procedures. Ingredients utilized in the formulation included ginger extract, silver nanoparticles, Carbopol 934, Span 80, Tween 80, coconut oil, glycerin, propylene glycol, methylparaben, propylparaben, triethanolamine, and purified water.

Table 1: Composition of Ginger–Silver Nanoparticle Emulgel Formulation

Ingredients	Quantity
Ginger Extract	4 mL
Carbopol 934	12.5 g
Span 80	5.7 mL
Tween 80	4.65 mL
Methylparaben	0.30 g
Propylparaben	0.15 g
Propylene Glycol	24.15 mL
Glycerin	2.85 mL
Triethanolamine	q.s.
Coconut Oil	5.50 mL
Silver Nanoparticles	0.004 g
Purified Water	q.s.

Preparation of Emulsion Phase

The oil phase was prepared by mixing Span 80, glycerin, and coconut oil, whereas the aqueous phase consisted of Tween 80 dissolved in purified water. Both phases were heated separately at 40–50 °C and subsequently combined with continuous stirring until a stable emulsion was formed.

Preparation of Gel Phase

The gel phase was prepared by dispersing Carbopol 934 in purified water with continuous stirring for 90 minutes. Preservatives including methylparaben and propylparaben were dissolved separately and incorporated into the gel system, followed by the addition of propylene glycol and ginger extract. Triethanolamine was gradually added to adjust pH and achieve the desired gel consistency.

The final emulgel formulation was obtained through gradual incorporation of the prepared emulsion phase into the gel phase under continuous mixing until homogeneity was achieved.

Physicochemical Evaluation of Emulgel

The developed emulgel formulations were evaluated for physicochemical characteristics including pH, viscosity, spreadability, and centrifugation stability to determine formulation suitability for topical administration.

pH Determination

The pH of each formulation was measured using a calibrated digital pH meter to determine compatibility with skin application and formulation stability.

Viscosity Determination

Viscosity testing was performed using a Brookfield viscometer at WVN Testing and Research Laboratory. Viscosity assessment was conducted to evaluate formulation consistency and retention during topical application.

Spreadability Test

Spreadability testing was performed to determine the ease of application and uniform distribution of the emulgel formulation on topical surfaces.

Centrifugation Stability Test

A centrifugation test was performed to assess formulation stability and determine the presence or absence of phase separation under stress conditions.

Antibacterial Evaluation

Antibacterial activity was evaluated using the Kirby–Bauer disk diffusion method against *Staphylococcus aureus*. Mueller–Hinton agar plates inoculated with standardized bacterial cultures were prepared for susceptibility testing. Treatment disks impregnated with emulgel formulations were placed onto inoculated agar plates and incubated under controlled laboratory conditions.

The antibacterial activity of each formulation was determined by measuring the zone of inhibition in millimeters surrounding the treatment disks. Vancomycin served as the positive control, whereas the blank formulation served as the negative control.

Statistical Analysis

Experimental data obtained from antibacterial testing were analyzed using one-way analysis of variance (ANOVA) to determine significant differences in antibacterial activity among treatment groups. Tukey's Honestly Significant Difference (HSD) post hoc test was subsequently employed to identify pairwise differences among formulations when statistical significance was observed. Statistical significance was established at $p < 0.05$.

Results and Discussion

Percentage Yield of Ethanolic Ginger Extract

The percentage yield of ethanolic ginger extract was determined to evaluate extraction efficiency and recovery of phytochemical constituents associated with antibacterial activity.

Table 2: Percentage Yield of Ethanolic Ginger Extract

Parameter	Result
Percentage Yield	89.84%

The ethanolic extraction of *Zingiber officinale* yielded 89.84%, indicating substantial recovery of phytochemical constituents. The high extraction efficiency may be attributed to the polarity of ethanol, which effectively extracts biologically active compounds such as gingerols, shogaols, flavonoids, and phenolic compounds associated with antibacterial activity [4].

A high extraction yield is essential in pharmaceutical formulation because insufficient recovery of active phytochemicals may reduce therapeutic efficacy. The findings suggest that ethanol served as an effective extraction solvent for ginger and may have contributed to the antibacterial performance observed in subsequent analyses.

Physicochemical Evaluation of Emulgel Formulation

The developed emulgel formulations were evaluated to determine physical stability and suitability for topical administration.

Table 3: Physicochemical Properties of Emulgel Formulations

Parameter	Observation
Appearance	Homogeneous
pH	6.8–7.3
Spreadability	Acceptable
Viscosity	Stable
Centrifugation Test	No Phase Separation

The developed formulations demonstrated acceptable physicochemical characteristics suitable for topical application. The formulations remained homogeneous throughout the observation period and exhibited no visible evidence of instability or phase separation during centrifugation testing.

The observed pH range of 6.8–7.3 indicates compatibility with skin application and may minimize irritation risk associated with topical administration. Appropriate viscosity and spreadability are essential because these properties influence formulation retention, patient acceptability, and uniformity of application [9], [10]. The absence of phase separation further suggests favorable compatibility among formulation components, including ginger extract and silver nanoparticles.

Ultraviolet–Visible Spectrophotometric Analysis of 6-Gingerol

Ultraviolet–visible spectrophotometric analysis was performed to confirm the presence of 6-gingerol, a major phytochemical constituent responsible for the antibacterial activity of ginger.

Table 4: Quantification of 6-Gingerol in Ethanolic Ginger Extract

Parameter	Result
6-Gingerol Content	0.023 mg/g

The ethanolic ginger extract demonstrated a measurable 6-gingerol concentration of 0.023 mg/g, confirming the presence of bioactive constituents associated with antibacterial activity. Gingerol compounds have been reported to inhibit bacterial growth through membrane disruption, suppression of bacterial enzyme systems, and interference with microbial metabolic processes [5].

The successful detection of 6-gingerol supports the antibacterial activity observed among ginger-containing formulations. Previous investigations have likewise demonstrated that ginger phytochemicals contribute substantially to antibacterial activity against *Staphylococcus aureus* and other clinically important pathogens [6].

Antibacterial Activity of Emulgel Formulations Against *Staphylococcus aureus*

The antibacterial activity of the different formulations was evaluated using the Kirby–Bauer disk diffusion assay against *Staphylococcus aureus*.

Table 5: Mean Zone of Inhibition of Different Formulations Against *Staphylococcus aureus*

Formulation	Mean Zone of Inhibition (mm)
Ginger Extract	10.17
Silver Nanoparticles	8.55
Ginger + Silver Nanoparticles	9.08
Positive Control (Vancomycin)	23.21
Negative Control	0.00

The findings demonstrated that the ginger extract formulation exhibited the greatest antibacterial activity among the experimental treatments, producing a mean zone of inhibition of 10.17 mm. The combined ginger–silver nanoparticle formulation demonstrated a mean inhibition zone of 9.08 mm, whereas silver nanoparticles alone produced 8.55 mm. As expected, vancomycin demonstrated superior antibacterial activity, while the negative control showed no inhibitory effect.

Interestingly, incorporation of silver nanoparticles did not enhance the antibacterial activity of the ginger formulation. Contrary to expectations of synergistic enhancement, the ginger extract formulation demonstrated greater antibacterial activity than the nanoparticle-containing formulation. Previous investigations have reported synergistic interactions between phytochemicals and silver nanoparticles because of increased membrane penetration and microbial disruption [8]. However, inconsistent findings may occur depending on nanoparticle concentration, formulation compatibility, aggregation behavior, and drug release characteristics.

One possible explanation for the reduced performance of the combined formulation may involve physicochemical interactions between silver nanoparticles and ginger phytochemicals within the emulgel matrix, which may have interfered with bioavailability or antibacterial release. Nanoparticle aggregation may likewise have reduced effective antimicrobial contact with bacterial cells. These findings suggest that incorporation of nanoparticles does not necessarily guarantee enhanced antibacterial activity and highlight the importance of formulation optimization in nanotechnology-assisted pharmaceutical systems.

Statistical Analysis of Antibacterial Activity

One-way analysis of variance (ANOVA) was performed to determine significant differences in antibacterial activity among the different formulations.

Table 6: One-Way ANOVA of Antibacterial Activity

Statistical Test	p-value	Interpretation
One-Way ANOVA	< 0.001	Significant

Statistical analysis demonstrated significant differences in antibacterial activity among treatment groups ($p < 0.001$), indicating that at least one formulation significantly differed in antibacterial performance. Subsequent post hoc analysis using Tukey's Honestly Significant Difference test revealed that the ginger extract formulation demonstrated significantly greater antibacterial activity than both silver nanoparticles alone and the combined ginger–silver nanoparticle formulation.

These findings suggest that the antibacterial activity observed in the study was primarily attributable to ginger phytochemicals rather than silver nanoparticle enhancement. The results further emphasize the need for optimization of nanoparticle concentration and formulation compatibility to improve antibacterial performance in future pharmaceutical development.

Conclusion

The present study successfully developed and evaluated a nanotechnology-enhanced ginger (*Zingiber officinale*) emulgel incorporated with silver nanoparticles and assessed its in vitro antibacterial activity against *Staphylococcus aureus*. Ethanolic extraction of ginger resulted in a high percentage yield of 89.84% and confirmed the presence of 6-gingerol (0.023 mg/g), a phytochemical constituent associated with antibacterial activity.

The developed emulgel formulations demonstrated acceptable physicochemical characteristics, including homogeneity, favorable pH, suitable spreadability, stable viscosity, and absence of phase separation, indicating their suitability for topical pharmaceutical application. Among the tested formulations, the ginger extract formulation demonstrated greater antibacterial activity compared with the combined ginger–silver nanoparticle formulation and silver nanoparticles alone.

Statistical analysis revealed significant differences in antibacterial activity among treatment groups ($p < 0.001$). Contrary to the anticipated synergistic effect, incorporation of silver nanoparticles did not improve antibacterial performance. These findings suggest that ginger extract served as the principal contributor to antibacterial activity and may represent a promising plant-derived antibacterial agent for topical application against *Staphylococcus aureus*.

Future investigations may focus on optimization of nanoparticle concentration, enhancement of formulation compatibility, controlled-release systems, and evaluation against broader microbial strains to further improve antibacterial performance and pharmaceutical applicability.

Conflict of Interest

The author declares that no conflict of interest exists regarding the publication of this research. The study was conducted independently, and no external financial or organizational influence affected the findings or interpretation of the results.

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