COLLAGEN-MEDIATED SILVER NANOPARTICLES FOR MULTIFUNCTIONAL FINISHING OF COTTON FABRIC WITH UV PROTECTION AND ANTIBACTERIAL ACTIVITY

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ABSTRACT

A novel approach for functional finishing of cotton fabric was developed using silver nanoparticles (AgNPs) synthesized with collagen extracted from Nile Tilapia scale. The green synthesis utilized fish derived collagen as both a reducing and stabilizing agent, ensuring eco-friendly fabrication and uniform nanoparticle distribution on the fabric surface. The synthesized AgNPs were characterized by UV–Vis's spectroscopy, X-ray diffraction, Fourier-transform infrared spectroscopy and scanning electron microscopy, revealing an average particle size of 2.58 nm. Optimization of process parameters for treating cotton fabric with collagen-mediated AgNPs was performed using Box bhenken design via a dip-coating method. The functionalized fabric was evaluated for antibacterial activity and ultraviolet protection. The optimized treatment exhibited a zone of inhibition (ZOI) of 25 mm against Escherichia coli and 24 mm against Staphylococcus aureus, while the ultraviolet protection reached 98.99%, demonstrating near complete UV-A and UV-B blocking. These findings indicate that silver nanoparticles synthesized with Nile Tilapia scale collagen provide an eco-friendly, efficient, and cost-effective method for producing multifunctional cotton textiles with excellent antibacterial and UV-protective properties.

Keywords: Antibacterial; collagen; green-synthesis; silver nanoparticles; UV protection

1. INTRODUCTION

In recent years the biodegradability ,softness and moisture absorbency of cotton make it the most widely used natural cellulosic fiber in the textile industries (Novaković, Popović et al. 2020). Cotton fabrics, however, are vulnerable to microbial contamination because of their hydrophilic and porous nature, which fosters the growth and proliferation of microorganisms like bacteria and fungi (Zambrano, Pawlak et al. 2020). This microbial activity lead to fabric deterioration, unpleasant odors, discoloration, and potential health risks, including skin irritation and infections (Bhat, Eraslan et al. 2021). In addition, cotton fabrics inherently exhibit poor resistance to ultraviolet (UV) radiation, particularly in lightweight and light-colored forms, thereby offering limited protection against harmful UV rays. Therefore this limitations significantly restrict the performance of cotton textiles in applications requiring hygiene, durability, and protective functionality(Bashari, Shakeri et al. 2018, Dai, Cao et al. 2025).To address these challenges, significant research efforts have focused on the development of multifunctional textile finishes that impart properties such as antibacterial activity and UV protection (Ghosh, Rupanty et al. 2025). Functionalization of cotton fabrics through surface modification has become a key strategy to enhance performance without compromising intrinsic textile properties (Nadi, Boukhriss et al. 2018). In this context, nanotechnology has emerged as a powerful tool due to its ability to introduce advanced functionalities at the nanoscale with minimal impact on fabric handle, weight, and breathability(Chy, Das et al. 2025).Among various nanomaterials, silver nanoparticles (AgNPs) have been widely studied and applied in textile finishing owing to their strong broad-spectrum antimicrobial activity and excellent UV-blocking properties(Verma and Maheshwari 2019, Ahmed, Mazumdar et al. 2024). AgNPs exert antibacterial effects through multiple mechanisms, including disruption of microbial cell membranes, generation of reactive oxygen species, and interaction with intracellular components, leading to cell death (Dakal, Kumar et al. 2016). Simultaneously, their unique optical characteristics enable efficient absorption and scattering of UV radiation, thereby enhancing the ultraviolet protection factor of treated fabrics. Despite these advancements, conventional chemical synthesis methods for AgNPs often involve toxic reducing agents and stabilizers, posing environmental and health concerns (Hosny, Gaber et al. 2025). Consequently, there has been a growing shift toward green synthesis approaches utilizing biological materials such as plant extracts, polysaccharides, and natural polymers as eco-friendly alternatives(Bhardwaj, Singh et al. 2020). Plant-mediated synthesis, particularly using phenolic compounds, has been widely reported in literature .However, these systems often suffer from limitations such as variability in composition, lower stability, and reduced durability of the functional finish on textile substrates (Islam and Sun 2022) These is due to the use of synthetic binders and crosslinking agents to improve durability, which compromise the eco-friendliness and sustainability of the finishing process (Rawat, Singh et al. , Rawat, Singh et al. 2026). To overcome these limitations, biopolymers as both reducing and stabilizing agents for nanoparticle synthesis were explored. For example, carboxymethyl cellulose (CMC) (Bozaci, Akar et al. 2015) and sodium alginate(Mahmud, Pervez et al. 2020) , chitosan (Gadkari, Ali et al. 2020) were employed to synthesize silver nanoparticles, and the antibacterial properties of cotton fabric have been investigated. Additionally collagen has been extensively studied in biomedical and drug delivery applications, its role in nanoparticle-mediated textile functionalization, particularly for AgNPs synthesis and finishing of cotton fabrics, remains limited in the existing literature(Karade, Patil et al. 2021, Zhang, Jiao et al. 2025). In the present study, a natural stabilizer was collagen from the scales of the fish was used as a stabilizer agent. This study aims to developing collagen-mediated silver nanoparticles for multifunctional finishing of cotton fabric with enhanced antibacterial and UV-protective properties. In this approach, collagen is employed as a green reducing and stabilizing agent for the synthesis of AgNPs, eliminating the need for hazardous chemicals. Simultaneously, its natural affinity toward cellulose is leveraged to improve nanoparticle binding, thereby enhancing the durability and performance of the finished fabric.

2. LITERATURE REVIEW

This section reviews previous studies on the development of functional materials, with a particular focus on the antibacterial and UV protection properties of cotton fabrics. The purpose of this review is to provide a clear background on the key challenges addressed in this research and to emphasize its importance. It also examines the various factors that influence the antibacterial performance and UV shielding ability of cotton, thereby establishing a foundation for further investigation into improving these properties through advanced treatments and innovative technologies.

2.1. MULTIFUNCTIONAL FINISHING OF TEXTILES

In recent years, the textile industry has shifted from conventional fabrics toward advanced materials with multiple performance characteristics (Thite, Krishnanand et al. 2018) . The use of multifunctional finishing methods, in which chemical agents are applied in a combined bath or a single step to impart specific functional properties to textiles, is becoming increasingly popular(Zhu, Ding et al. 2020). Improvement of existing properties and the creation of new material properties are the most important reasons for the functionalization of textiles(Gowri, Almeida et al. 2010). Environmental awareness has also increased with the globalization of the textile industry. As textile wet processing is considered as the most polluting one, multifunctional finishing in single bath has been extensively used in several industrial processes to reduce the negative impact on the environment(Singh, Chakraborty et al. 2023).Now a day, the development of functional and sustainable textiles has been the focus of researchers. The functional textiles are those that are developed specifically for an end purpose with added attributes such as self-cleaning, hydrophilicity, antibacterial activity, crease recovery, and super hydrophobicity. The demand of functionality in traditional clothing as well as in home textiles has significantly increased during recent years(Aslam, Hussain et al. 2019). To overcome the challenges of sustainability in textile wet processing, as well as to overcome the social and environmental challenges without the compromising economic growth, attempts have been made toward the water, energy and chemical conservations to reduce the pollution load and tend the textile industry toward sustainability(Eid and Ibrahim 2021)

2.2 ESSENCE OF NANO TECHNOLOGY

In the field of textiles, nanotechnology is a revolutionary breakthrough that has ushered in a new era of improved performance and functionality. Numerous enhancements are included with this innovation, such as resilience to water, odor, flame, stain, UV protection, and antibacterial qualities (Syduzzaman, Hassan et al. 2023). A number of metal oxides, including copper oxide, zinc oxide, and titanium dioxide, as well as metal nanoparticles, including gold, zinc, copper, and silver, have attracted a lot of interest because of their remarkable properties. These properties include a higher surface-area-to-volume ratio, which contributes to their antimicrobial action (Ribeiro, Shvalya et al. 2022). In recent years, silver nanoparticles have displayed the highest level of commercialization and account for 55.4% of the total nanomaterial-based consumer products available on the market. Silver nanoparticles have been utilized in diverse domains (Paladini and Pollini 2019).

SYNTHESIS OF AGNPS: An important area of research in nanotechnology is the synthesis of NPs with different chemical compositions, sizes, morphologies, and controlled dispersities (Iravani, Korbekandi et al. 2014). Today, a variety of preparation techniques have been reported for the synthesis of silver NPs, including physical, chemical, and biological synthesis (Almatroudi 2020). This chemical methods offer advantages such as convenience, simplicity, and cost effectiveness for large-scale production. However, their usage of toxic chemicals and the production of harmful by-products cause environmental damage, thereby limiting their clinical and biomedical applications (Almatroudi 2020). In recent years, the growing demand for reliable, nontoxic, and eco-sustainable methods in synthesis of silver metallic nanoparticles has prompted a shift away from conventional methods (Dikshit, Kumar et al. 2021)

MECHANISTIC INSIGHTS INTO AGNPS FORMATION:

Researchers explained the mechanism of how amino groups act as reducing agent in silver nanoparticle synthesis process. Mikhailova (2020) explain that while the amino groups of proteins take part in the silver reduction process (Mikhailova 2020) and a reducing agent first reduces the ions to atoms: $Ag^{+} (aq) + e^{-} \rightarrow Ag (s)$ then atoms are begin to nucleated (Nguyen, Dang et al. 2023).

3. MATERIALS AND METHODS

3.1. MATERIALS

The bleached plain woven 100% cotton fabric used in this study was obtained from the Textile and Garment Engineering Department laboratories at Hawassa University, Ethiopia. Fish scales from Nile Tilapia were gathered at a fish market in amorre gedel Hawassa, Ethiopia.
Silver nitrate ($AgNO_3$, ≥99% purity) was used as the precursor for silver nanoparticle synthesis. Additional chemicals, including sodium hydroxide (NaOH, ≥98% purity) and ethanol, were used for pH adjustment and cleaning purposes. All chemicals

used in this study were of analytical grade and were used without further purification. Distilled water was used throughout the experiments**.**

3.2. METHODS

3.2.1 EXTRACTION PROCESS OF COLLAGE:

Collagen was obtained from the scales of Nile Tilapia through a series of three processes: deproteinization, demineralization, and isolation. In the deproteinization phase, the scales underwent treatment with 0.1 M NaOH (1:10 w/v) at 80 °C for 8 hours to eliminate non-collagen proteins, and then they were rinsed to

reach a neutral pH. The demineralization process involved using 0.5 M HCl (1:12 w/v) at 90 °C for 12 hours to remove calcium minerals. Lastly, during the isolation phase, the demineralized powder was processed with 0.25 M acetic acid (pH 2.5) for 48 hours, followed by centrifugation, salting out with 2 M NaCl, dialysis, and lyophilization, ultimately resulting in dry acid-soluble collagen (ASC).

3.2.2 SILVERNANOPARTICLE SYNTHESIS PROCESSES

Silver nanoparticles (AgNPs) were synthesized using a green method with collagen acting as both a reducing and stabilizing agent. A 0.1 M silver nitrate was dissolved in 90 mL and heated to 60 °C under continuous stirring at 450 rpm for 1 hour. Collagen solution. at a 1:2 (v/v) ratio was gradually incorporated into the silver nitrate

Solution while maintaining continuous stirring with a magnetic stirrer, and the pH was adjusted to 8.The reaction was continued until the solution color changed from light yellow to brown, indicating the formation of AgNPs. The nanoparticles were then separated by centrifugation at 4000 rpm for 20 min, washed thoroughly with distilled water to remove impurities, and dried in an oven

Figure 1 Collagen–based silver nanoparticle synthesis process

3.2.3 CHARACTERIZATION AND EVALUATION

CHARACTERIZATION OF SILVER NANOPARTICLES: Silver nanoparticles were characterized using UV–Visible spectroscopy, FTIR, XRD, and SEM. UV–Vis spectroscopy (190–1100 nm, 20–25°C) was used to confirm nanoparticle formation. FTIR analysis (4000–500 cm^{-1}, 0.5 cm^{-1} resolutions) identified functional groups involved in reduction and stabilization. XRD was employed to determine crystal structure, phase composition, and crystallite size. Surface morphology and particle distribution were examined using scanning electron microscopy (SEM).

ANTIBACTERIAL-ACTIVITY:

Antibacterial activity of treated fabrics was evaluated using the agar diffusion (zone of inhibition) method according to SNV 195920-1992(Ristić, Zemljič et al. 2011) against Staphylococcus aureus and Escherichia coli. Sterile agar plates were prepared, inoculated with 24 h bacterial cultures, and fabric samples (10 × 10 mm) were placed on the surface. After incubation at 37°C for 18–24 h, antibacterial activity was assessed by measuring the inhibition zones. The percent inhibition antifungal and antibacterial will be calculated using the following formula

$$\% \text{ Inhibition } = \frac{\text{Total area of disc} - \text{Area of ZOI}}{\text{Total area of disc}} X100 \qquad (1)$$

UV PROTECTION PERFORMANCE: UV protection was evaluated according to AATCC 183-2000 by measuring UV transmittance and calculating the Ultraviolet Protection Factor (UPF). UPF values were classified as good (15–24), very good (25–39), and excellent (40–50+) categories (Bashari, Shakeri et al. 2019)The calculation was based on erythemal spectral effectiveness (Eλ), solar spectral irradiance (Sλ), transmittance (Tλ), and wavelength interval (Δλ). Each sample was calculated using the following formula as per the AATCC 183-2000 method.

$$\text{UPF} = \frac{\sum_{280\ nm}^{400\ nm} E\lambda x S\lambda x \Delta\lambda}{\sum_{280\ nm}^{400\ nm} E\lambda x S\lambda x \Delta T \Delta\lambda} \qquad (2)$$

Where, Eλ = relative erythemal spectral effectiveness, Sλ =solar spectral irradiance, Tλ = average spectral transmittance of the sample, and Δλ = measured wavelength interval (nm)

PHYSICAL AND MECHANICAL PROPERTIES:

Tensile strength was determined according to ISO 13934-2, Fabric stiffness was evaluated in both warp and weft directions using ISO bending length method on 25 × 25 cm samples, and wash durability results were reported as the average of three measurements.

4. RESULT AND DISCUSSION

4.1 CHARACTERIZATION OF AgNP

FTIR ANALYSIS: As shown in Graph A in Figure **2** the FTIR spectrum of pure silver nitrate exhibited characteristic nitrate-associated vibrational bands in the 450–1750 cm^{-1} region. According to Augustine, Kalarikkal et al. (2014), Following biosynthesis, the FTIR spectrum of AgNP–collagen revealed pronounced compositional and structural modifications, indicating successful biomolecule-mediated reduction and stabilization of silver nanoparticles. A broad absorption band at 3320 cm^{-1} is assigned to N–H stretching vibrations, while the band at 2973 cm^{-1} corresponds to aliphatic C–H stretching, both confirming the presence of proteinaceous collagen on the nanoparticle surface. The emergence of strong amide I (C=O stretching) at 1613 cm^{-1} and amide II (N–H bending) at 1566 cm^{-1} further verifies the retention of collagen backbone structures after nanoparticle formation (Mudhafar, Zainol et al. 2021). Importantly, a comparative decrease in the intensity of amide I and amide II bands in the AgNP–collagen spectrum, relative to native collagen, indicates a significant perturbation of the peptide bond environment. This spectral attenuation suggests that carbonyl (C=O) and amine (N–H) groups are actively involved in coordination with Ag^{+} ions during nanoparticle formation. Such interaction likely proceeds through lone pair donation from nitrogen and oxygen atoms to silver ions, facilitating complexation and subsequent electron transfer required for reduction to metallic Ag^{0}.

The observed spectral shifts and intensity variations therefore support a mechanism in which collagen acts not merely as a passive stabilizer but as a bifunctional agent, simultaneously serving as a mild reducing and effective capping matrix. The binding of Ag^{+} to amine-rich regions of collagen promotes nucleation sites for nanoparticle growth, while steric hindrance from the protein backbone prevents agglomeration, thereby enhancing colloidalstability.

UV-VISIBLE ANALYSIS: As show in Graph B in figure 2 Silver nanoparticles were characterized using a UV–Vis spectrophotometer, typically operating within a wavelength range of 190–1100 nm, with a spectral resolution of 1–5 nm and an operating temperature of 20–25 °C. The UV–Vis absorption spectrum of the synthesized silver nanoparticles in the presence of collagen extracted absorption peak at 418 nm. The observed color change and the appearance of this characteristic absorption band are attributed to the surface plasmon resonance (SPR) phenomenon, which is typically observed in the UV–Vis region of 400–460 nm for silver nanoparticles. These results confirm the successful synthesis of silver nanoparticles (Anith Jose, Devina Merin et al. 2022). Similar Uv-Vis spectrum results were reported by other researchers, with (Murugan, Subramaniam et al. 2021) noting a peak at 460 nm and (Sharma, Vishwakarma et al. 2022) observing a peak at 418 nm. These consistent findings validate the synthesis and characterization of silver nanoparticles using collagen extracted from fish scales

TGA ANALYSIS: Graph C in figure 2 shows three distinct weight loss stages corresponding to water loss, degradation of small organics, and decomposition of polymer residues. The curve displays three prominent areas indicating weight reductions. The first one is primarily attributed to the protein's loss of water, which caused a weight reduction of 10.96% at 116. The initial reduction in weight is due to the evaporation of adsorbed water and other volatile substances from the sample. The second weight loss (3.52%) took place in the range of 163°C – 254°C as a result of the degradation of small organic molecules and loosely bound organic residues. The third notable weight reduction (49.27%) occurred within the temperature span of 258.64 °C - 520 °C, as a result of the breakdown of more stable organic compounds or polymers linked to the silver nanoparticles (Cardoso, Quelemes et al. 2014) and also indicated the breakdown of organic capping agents that were used to stabilize the nanoparticles. After reaching the decomposition temperature, the graph displayed a stabilized region from fish scales, showing a distinct that signifies the removal of all volatile and degradable components.

SEM ANALYSIS:

Scanning Electron Microscopy (SEM) images in **Graph D** revealed clustered AgNPs with rough and dense surface morphology. At 3000× magnification, particles formed larger aggregates, indicating agglomeration. At 5000×, the nanoparticles appeared predominantly spherical and within the nanometer range, consistent with previous reports (Khodashenas and Ghorbani 2019). The observed agglomeration is attributed to the tendency of nanoparticles to minimize surface free energy (Altammar 2023). The SEM examination verifies the successful synthesis of silver nanoparticles with moderate aggregation and varied morphologies, indicating effective bio-reduction and stabilization made possible by the components of fish scale collagen.

XRD Analysis: Graph E in Figure 2 shows the X-ray diffraction (XRD) analysis of the synthesized silver nanoparticles (AgNPs) validated their crystalline structure. The diffractogram displayed several distinct and intense peaks at roughly 2θ values of 38°, 44°, 64°, and 77°, which align with the (111), (200), (220), (311) and (222) planes of face-centered cubic (fcc) silver, respectively, as per the standard JCPDS card no. 04-0783. The prominence of the (111) peak suggests that the silver nanoparticles exhibit a preferential orientation along this plane, a characteristic often seen in biologically synthesized AgNPs. The presence of sharp and narrow peaks further establishes the high crystallinity of the produced particles (Mudhafar, Zainol et al. 2024). The mean crystal size was calculated using Scherrer’s formula. The value corresponding to the highest intensity peaks is 2.58 nm.

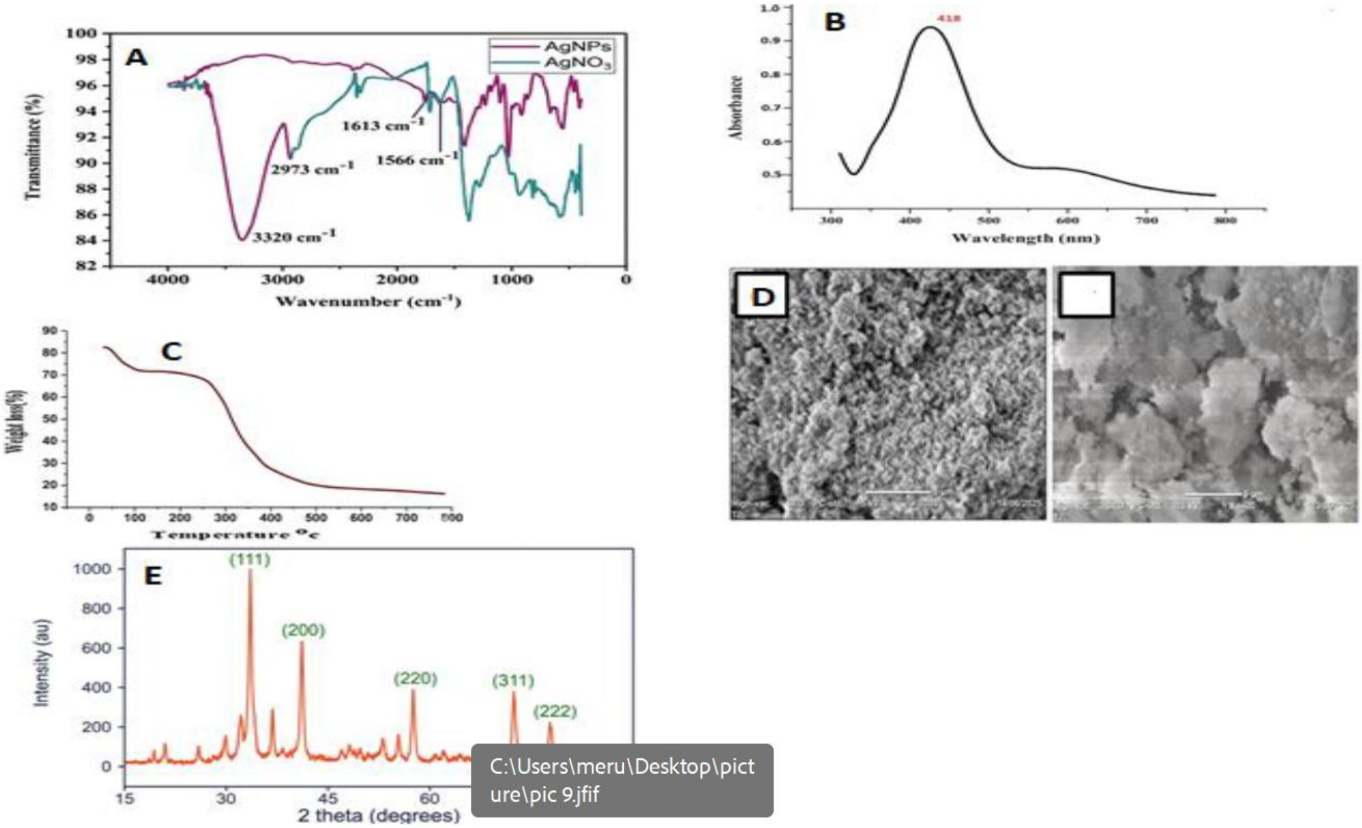

Figure 2 FTIR spectra of AgNPS (A) , UV-visible analyses of AgNPS (B), TGA image of AgNPS (C) , SEM images of AgNPS (D) and XRD patterns of AgNPS (E)

4.2 DATA PRESENTATION AND ANALYSIS

DATA PRESENTATION: The antibacterial test was done by putting a 10x10mm fabric sample on a nutrient agar plate, inoculating it with a gram negative and gram positive bacteria culture, and then incubating it. Following incubation, the circumference of the inhibition zone surrounding the fabric sample was measured carefully. The measured test results were expressed in millimetres of diameter, as shown in **Figure 3**. Each sample was levelled with a number.

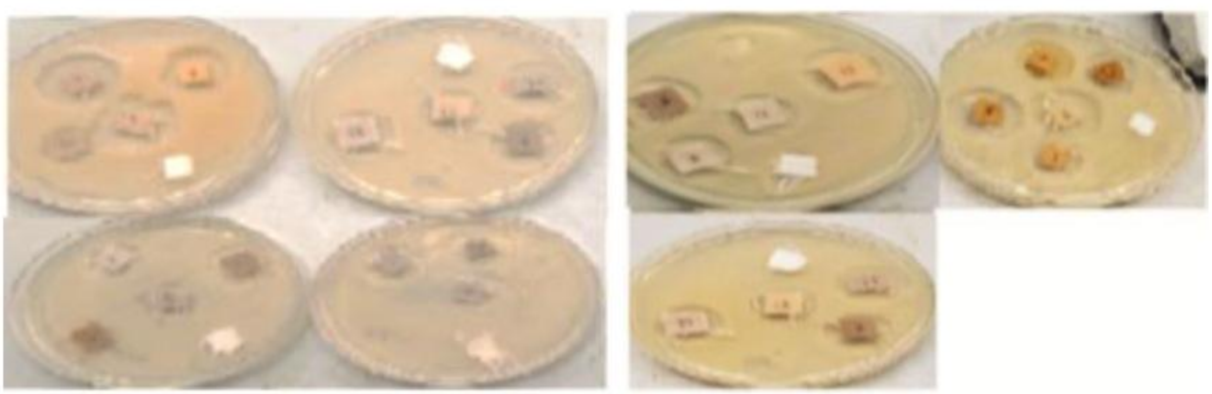

Figure 3 Antibacterial tests for total samples

Table 1 Experimental run for the optimization process

		Factor 1	Factor 2	Factor 3	Response 1	Response 2	Response 4
Std	Run	A:concentration of AgNPS	B:Temperature	C:Time	ZOI gram negative bacteria	ZOI gram positive bacteria	%Blocking UVR
		Wt%	^{o}C	minute	Mm	Mm	%
13	1	0.2	60	30	19	21	97.8
2	2	0.1	60	30	22	23	96.73
1	3	0.1	40	30	16	14	95.81
11	4	0.1	40	40	21	18	96.9
12	5	0.5	30	40	22	22	94.99
4	6	0.5	40	30	25	24	98.99
9	7	0.1	60	20	18	16	95.81
8	8	0.2	30	40	24	23	95.8
5	9	0.2	60	20	16	16	90
3	10	0.5	40	30	20	18	89
14	11	0.2	60	30	22	22	97.8
6	12	0.2	40	20	23	21	92.8
10	13	0.5	30	20	18	21	95.07
7	14	0.2	30	40	17	17	87.22

DATA ANALYSIS: The experimental data were analyzed to evaluate the responses, including the zone of inhibition against Gram-negative bacteria, zone of inhibition against Gram-positive bacteria, and ultraviolet protection efficiency. Design-Expert software was used for statistical analysis and optimization based on the Box–Behnken design . The analysis involved model development, analysis of variance (ANOVA), and assessment of the significance of individual factors and their interactions.

Table 2 ANOVA for the responses

	Gram -ve bacteria					gram +ve bacteria					%Blocking UVR				
Source	Some of squares	DF	Mean Square	F-value	P-value	Some of Squares	DF	Mean Square	F-value	P-value	Sum of Squares	DF	Mean Square	F-value	p-value
Model	161.46	9	17.94	19.14	0.0040	103.61	9	11.51	36.84	0.0017	151.96	9	16.88	38.59	0.0016
A-AgNPs	112.50	1	112.50	120.00	0.0007	53.78	1	54.79	184.78	0.0005	28.12	1	28.12	64.29	0.0083
B – Temperature	28.12	1	28.12	30.00	0.0054	34.12	1	34.12	97.60	0.0008	120.12	1	120.12	274.57	< 0.0001
C- Time	6.13	1	6.13	6.53	0.0629	3.13	1	3.13	10.00	0.0341	2.00	1	2.00	4.57	0.0993
AB	119.8	1	119.95	125.78	0.0004	60.50	1	60.50	193.60	0.0002	36.32	1	36.32	76.76	0.0013
AC	46.9	1	47.57	50.4	0.0049	6.90	1	6.90	7.12	0.0458	3.78	1	3.78	5.87	0.0789
BC	27.49	1	27.87	29.91	0.0068	2.50	1	2.65	8.00	0.0421	3287	1	32.87	58.71	0.0091
A^2	4.05	1	4.05	4.32	0.1062	2.45	1	2.45	7.84	0.0488	0.0500	1	0.0500	0.1143	0.7523
B^2	0.0500	1	0.0500	0.0533	0.8287	1.25	1	1.25	4.00	0.1161	1.25	1	1.25	2.86	0.1662
C^2	11.25	1	11.25	31.00	0.0257	1.25	1	1.25	4.00	0.1161	0.4500	1	0.4500	1.03	0.3679
Residual	3.75	4	0.9375			1.25	4	0.3125			1.75	4	0.4375		
Lack of Fit	1.75	3	0.5833	0.897	0.6588	0.7500	3	0.2500	0.5000	0.7478	1.25	3	0.4167	0.8333	0.6466
Pure Error	0.0012	1	0.0012			0.0002	1	0.0002			2.00	1	2.00		

As shown in **Table 2** the ANOVA results show that the developed regression models are statistically significant for all responses, indicating good model fit and reliability in explaining the variability in Gram-negative ($p = 0.0040$), Gram-positive ($p = 0.0017$) antibacterial activity, and UVR blocking efficiency ($p = 0.0016$).Among the main factors, AgNPs (A) is the most dominant variable, showing a highly significant effect on Gram-negative ($F = 120.00$, $p = 0.0007$) and Gram-positive bacteria ($F = 184.78$, $p = 0.0005$), indicating its strong antibacterial contribution. It also significantly influences UVR blocking ($F = 38.59$, $p = 0.0083$), confirming its multifunctional role in both antimicrobial and UV-protective performance. Temperature (B)
also exhibits a strong influence, particularly on UVR blocking, where it shows an exceptionally high F-value ($F = 274.57$, $p < 0.0001$). Its effect on antibacterial activity is also significant for both Gram-negative ($p = 0.0054$) and Gram-positive bacteria ($p = 0.0008$), though comparatively lower than AgNPs in antibacterial responses. Time (C) demonstrates a weaker and more selective influence, being significant only for Gram-positive bacteria ($p = 0.0341$). Its effect on Gram-negative bacteria ($p = 0.0629$) and UVR blocking ($p = 0.0993$) is statistically insignificant, indicating limited contribution to overall performance compared to A and B factor.

4.2.1 EFFECT OF THE MAIN FACTORS ON THE RESPONSES

INDIVIDUAL FACTORS ON ZOI GRAM NEGATIVE BACTERIA:

When the concentration of silver nanoparticle increase the zone of inhibition of gram negative bacteria increased, The previous research's also show the more nanoparticles interact with bacterial cell membranes, causing structural damage and increased permeability (Vasiliev, Kubo et al. 2023). Ahmed, B., et al. (2018), have been reported where higher AgNP concentrations enhanced antibacterial activity due to stronger interaction with bacterial membranes and increased Ag^+ ion release, leading to disruption of cellular functions(Ahmed, Hashmi et al. 2018).

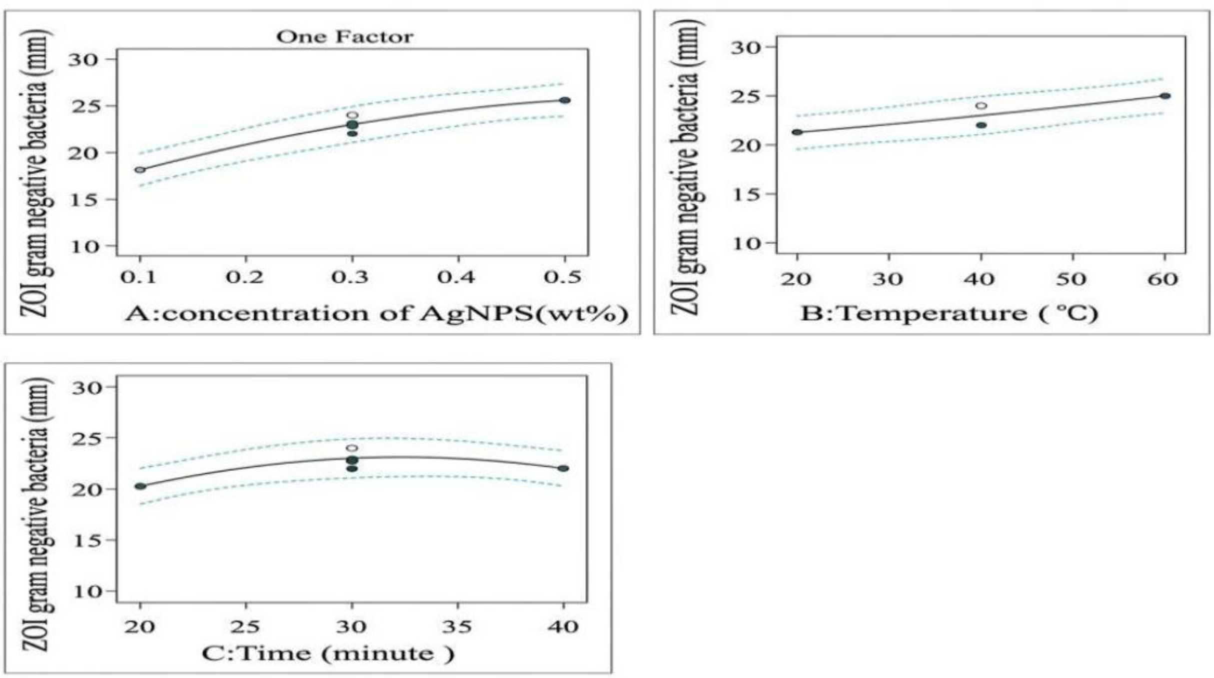

Figure 4 Effect of main factors on the zone of inhibition of negative bacteria

The graph (Figure 4) shows after maximum points the zone of inhibition start decrease because silver nanoparticles (AgNPs) tend to aggregate, which reduces their effective surface area and limits interaction with bacterial cells(Tang and Zheng 2018)When the temperature increase the zone of gram negative bacteria increase. Based on previous literature the increase in the zone of inhibition against Gram-negative bacteria with rising temperature is attributed to enhanced nanoparticle cell interactions, increased ion release, and higher reactive oxygen species (ROS) generation, which collectively damage the bacterial membrane and cellular components(Qi, Chen et al. 2018, Yang, Wang et al. 2020)Furthermore, increasing of time have an increasing effect on the zone of inhibition with prolonged exposure due to greater accumulation and sustained antibacterial action. However, after the optimum time, a decreasing effect is observed, likely due to nanoparticle aggregation and reduced active surface area, which limits further antibacterial efficiency (Arsene, Viktorovna et al. 2021).

INDIVIDUAL FACTORS ON ZOI GRAM POSITIVE BACTERIA: The effect of silver nanoparticles (AgNPs) on Gram-positive bacteria shows in **Figure** 5 an increasing zone of inhibition with increasing concentration, as observed in the one-factor interaction graph. This is consistent with previous studies reporting that AgNPs damage the thick peptidoglycan layer of Gram-positive bacteria through direct cell wall interaction and Ag^{+} ion release, which inhibits essential enzymatic functions and leads to cell death (Dakal, Kumar et al. 2016)The effect of temperature shows The zone of inhibition against Gram-positive bacteria with rising temperature. Based on previous studies this is due to enhanced nanoparticle activity, increased Ag^{+} ion release, and higher ROS production(Song, Wu et al. 2019) Similarly, as shown in **Figure 5** time shows an increasing effect due to prolonged nanoparticle bacteria interaction and sustained antibacterial action, while the slight decrease at higher time is attributed to nanoparticle aggregation and reduced active surface area (Tang and Zheng 2018)

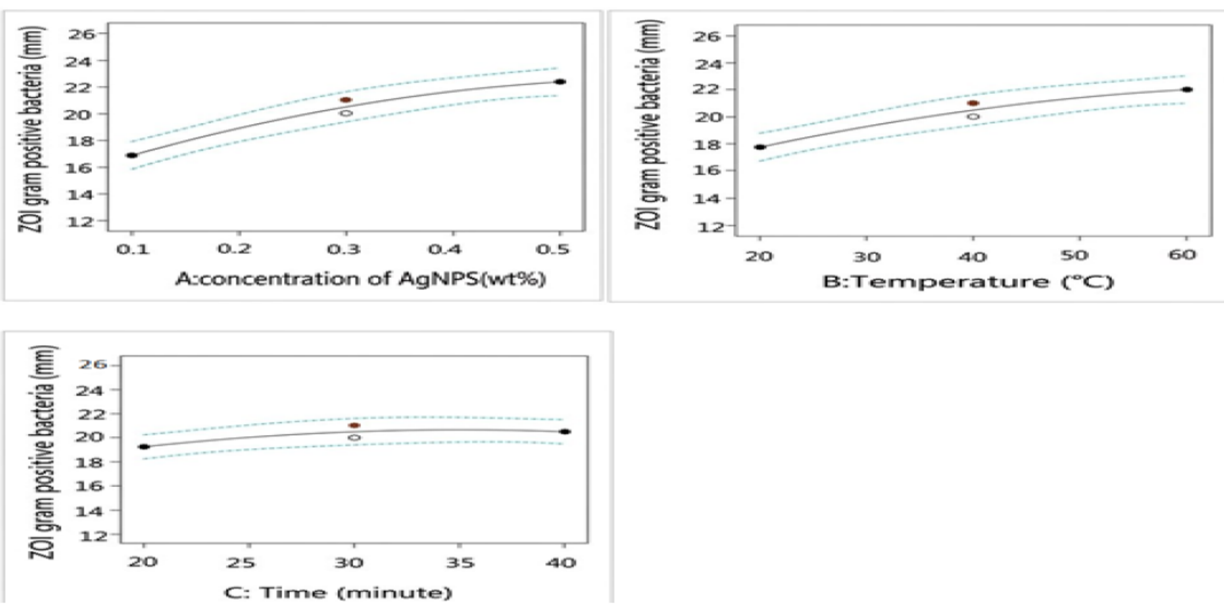

Figure 5 Effect of main factors on the zone of inhibition of positive bacteria

INDIVIDUAL FACTORS ON UVR: As shown in Figure 6 concentration increased from 0.1 to 0.5M, there is an increase in UPF for is observed. The broad-spectrum UV-blocking property of silver nanoparticles, which can block both UV-A and UV-B rays, makes them a popular and efficient UV-protective agent. They provide long-lasting protection because of their great photostability, which means they do not break down readily in the presence of sunshine. Better skin adhesion and more surface area are made possible by their nanoscale size, which also improves UV absorption and scattering without producing an obvious white residue(Noor, Mutalik et al. 2023).

In the case of temperature ,the UPF initially increases due to improved dispersion and interaction of silver nanoparticles within the matrix at moderate thermal conditions, enhancing UV absorption and scattering; however, at higher temperatures, a decrease in UPF is observed due to nanoparticle aggregation and structural instability, leading to reduced UV-blocking efficiency, as reported in previous studies (Rajaboopathi and Thambidurai 2018, Rehan, Mashaly et al. 2024).Similarly, the effect of time increase in UPF due to better nanoparticle distribution and stabilization within the matrix, while a gradual decrease at longer exposure times is attributed to particle agglomeration and structural rearrangement, which reduces effective surface coverage and UV protection efficiency, as observed in previous studies (Rajaboopathi and Thambidurai 2018)

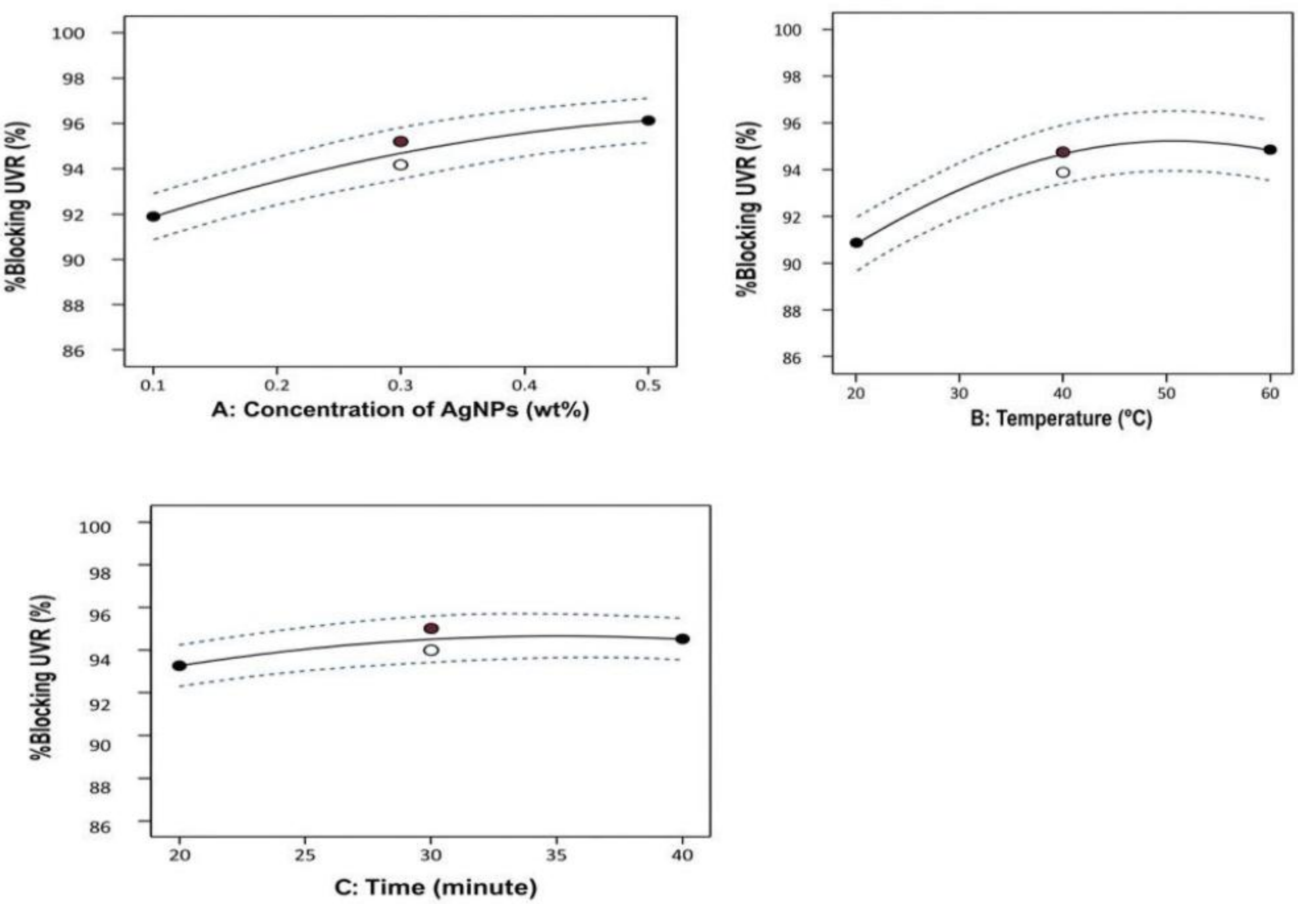

Figure 6 Effect of main factors on the UVR

INTERACTION EFFECT: The 3D surface shows the effect of significant independent variables, which are concentration of silver nanoparticle, temperature, and time on the response, per cent reduction of gram-negative and gram-positive bacteria, as well as UPF activity, with the three-dimensional graph. The interaction effects of factors on the three-dimensional graph were shown for each response as follows.

The 3D response surface plots show the three interaction effects of AgNPs concentration, temperature, and time on the antibacterial activity. The results indicate that the zone of inhibition for Gram-negative bacteria is strongly influenced by these combined factors. Overall, the highest inhibition zone of 25 mm was observed at 0.5 M AgNPs concentration and 40°C temperature. In contrast, lower concentration (0.1 M) and lower temperature conditions produced reduced inhibition zones indicating weaker antibacterial performance.

INTERACTION EFFECT ON ZOI OF GRAM NEGATIVE BACTERIA:

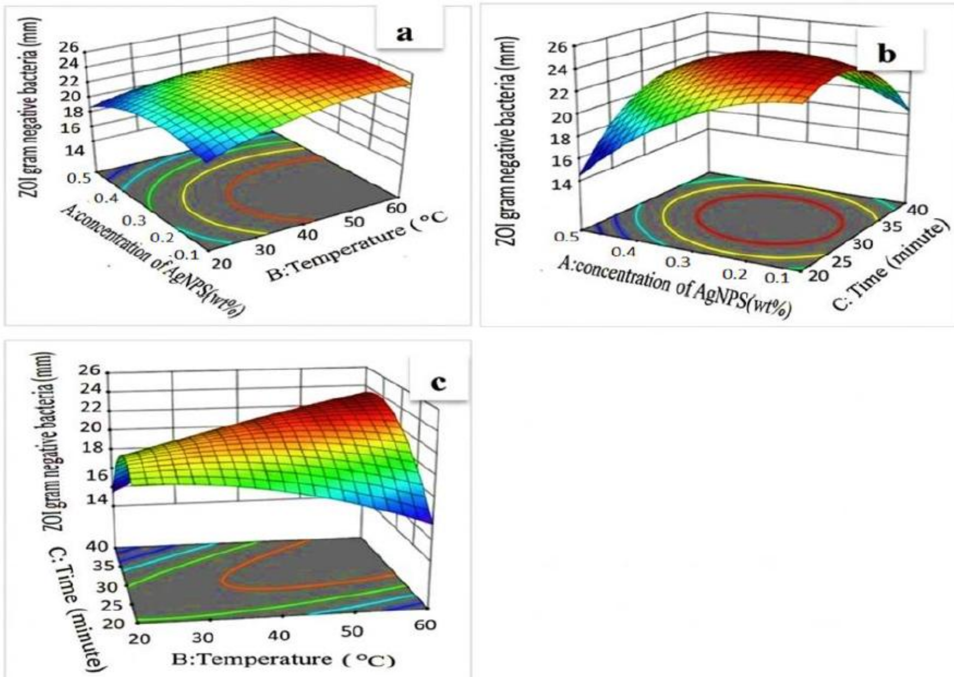

Figure 7 Interaction effects of independent factors on the percent reduction of gram-negative bacteria

INTERACTION EFFECT ON ZOI OF GRAM POSITIVE BACTERIA

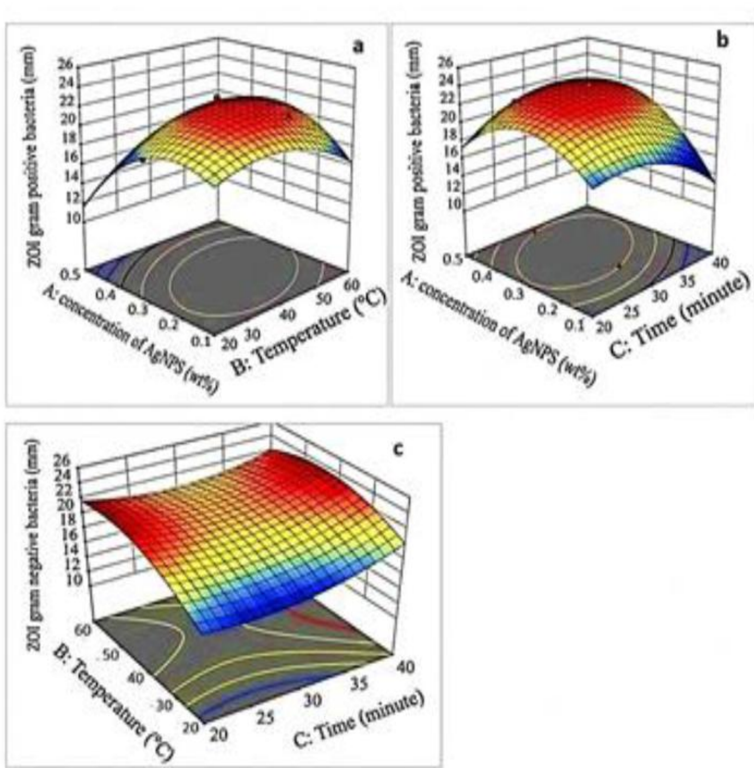

Figure 8 Interaction effects of independent factors on the percent reduction of gram-positive bacteria

As shown in Figure 8 the 3D response surface for Gram-negative bacteria shows that the zone of inhibition is strongly affected by the interaction of AgNPs concentration, temperature, and time. The AB interaction (AgNPs–temperature) gives higher antibacterial activity at increased levels due to improved nanoparticle formation. The AC interaction (AgNPs–time) shows that longer treatment enhances nanoparticle deposition and bacterial inhibition, while the BC interaction (temperature–time) improves diffusion and stabilization of AgNPs on the fabric.. The maximum inhibition zone of 24 mm was recorded at 0.5 M AgNPs and 40°C, confirming that optimized conditions of all three factors enhance antibacterial performance.

INTERACTION EFFECT ON UVR:

AS shown in Figure 9 the interaction between AgNPs concentration and temperature (AB) indicates that increasing both parameters enhances UV blocking efficiency. The maximum UVR blocking efficiency of 98.99% was obtained at 0.5 M AgNPs concentration and 40°C temperature. This improvement is attributed to better dispersion and stronger deposition of silver

nanoparticles on the fabric surface at moderate thermal conditions, which increases UV absorption and scattering. The interaction between AgNPs concentration and time (AC) also shows a positive effect on UV protection. Longer treatment time allows sufficient interaction between nanoparticles and the fiber surface, improving uniform coating and stability(Lodeiro, Achterberg et al. 2016).

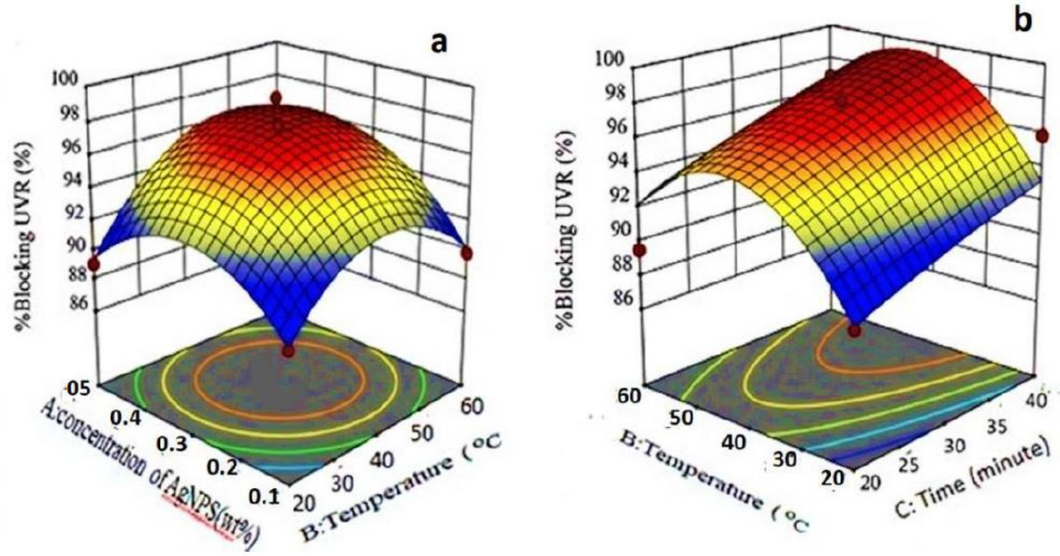

Figure 9 Interaction effects of independent factors on the percent reduction of blocking UVR

4.3 TESTING AND EVALUATION

AIR PERMEABILITY: Fabrics with high air permeability feel lighter and drier, enhancing wearability in apparel(Rajabov 2024). The control fabric exhibited an air

permeability of approximately 17.8 cm³/cm²/s, whereas the optimized treated fabric showed a reduced value of 15.5 cm³/cm²/s. This decline in air permeability can be attributed to modifications in the fabric's surface structure after treatment. The deposition of active substances on the fiber surface leads to partial pore blockage, increased surface roughness, and relaxation shrinkage, all of which contribute to higher resistance to airflow (Mazzon 2021).

WASH DURABILITY: Wash fastness test for assessing the antimicrobial and percent blocking of ultraviolet protection was done. Their durability up to 5 and 10 washing cycles

are shown by in table 19. The decrease in antibacterial activity and UV protection of cotton fabric treated with silver nanoparticles after washing is mainly due to the gradual removal of nanoparticles from the fabric surface(Wu, Yang et al. 2019). During washing cycles, mechanical agitation and detergent action cause loosely bound AgNPs to detach from the cotton fibers, leading to a reduction in active antimicrobial sites. In addition, the continuous leaching of silver ions (Ag^+), which are responsible for most of the antibacterial effect, further reduces the fabric's effectiveness.

Table 3 Wash fastness for antimicrobials and %blocking of UV test

	Gram negative bacteria	Gram positive bacteria	%block UVR	%block UVA	%block UVB	UVR
	ZOI (mm)	ZOI (mm)	%	%	%	
5- Washing Cycle	22	20	95.65	95.52	96.8	Very good
10- Washing Cycle	18	16	87.7	87.63	91.09	Medium
Untreated Sample	_	_	86.36	86.3	86.57	Poor

TENSILE STRENGTH TEST: The increase in tensile strength of cotton fabric after treatment with silver nanoparticles can be explained in a simple way by how the nanoparticles interact with the fibers. According to Grandolfo, A. (2025) and another recent research shows when AgNPs are deposited onto the cotton, they attach to the surface and also penetrate into small spaces between the fibers. This helps the fibers stick together more firmly and reduces the movement or slipping of fibers when force is applied(TANIA 2021, Grandolfo 2025)In addition, the nanoparticles act like tiny reinforcing fillers inside the fabric structure. They fill small gaps and make the fabric structure more compact and stable. Because of this improved bonding and tighter structure, the fabric becomes stronger and can withstand more force before breaking (Anik, Tushar et al. 2025).

Table 4 Tensile strength result of optimized sample

Sample	Force (N/mm^2)		Elongation at break (%)	
	Warp	Weft	Warp	Weft
Control sample	286	276	18.1	12.4
Optimized sample				
S1	289	274	17.6	12.4
S2	291	278	17.7	11,5
S3	289	276	16.9	12.2
S4	293	275	17.4	11.1
S5	292	278	16.1	12.1

4. CONCLUSION

An eco-friendly finishing strategy was successfully developed to improve the antibacterial and ultraviolet protective performance of cotton fabric through the application of collagen-mediated silver nanoparticles (AgNPs). Collagen extracted from fish scales was effectively utilized as both a reducing and stabilizing agent, enabling a green synthesis route that avoids the use of hazardous chemicals. This approach not only ensured the formation of stable nanoparticles but also promoted better interaction and adhesion between the AgNPs and the cotton fibers. Comprehensive characterization using X-ray diffraction (XRD), scanning electron microscopy (SEM), UV–Vis spectroscopy, and Fourier-transform infrared spectroscopy (FTIR) confirmed the successful synthesis and deposition of well-dispersed nanoparticles,

with an average particle size of 2.58 nm.The functionalized cotton fabric demonstrated remarkable antibacterial performance against both Gram-negative and Gram-positive bacteria, exhibiting zones of inhibition of 25 mm and 24 mm, respectively. In addition to its antimicrobial efficacy, the treated fabric showed excellent ultraviolet protection, achieving a UPF value of 98.99, which indicates near-complete blocking of harmful UV radiation. The enhanced performance can be attributed to the synergistic effect between collagen and silver nanoparticles, where collagen improves nanoparticle stability and binding affinity to the cellulose structure, while AgNPs impart strong antibacterial and UV-blocking properties.This study highlights the effectiveness of integrating bio-derived materials with nanotechnology to produce high-performance textiles. The developed method is not only environmentally friendly and cost-effective but also scalable for industrial applications. It presents a viable alternative to conventional chemical finishing processes, with significant potential for use in medical textiles, protective garments, outdoor fabrics, and other applications where hygiene and UV protection are critical.

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