

Environmentally Friendly Production of Silver Nanoparticles Using an Ethanolic Extract of Saffron Petals

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ABSTRACT

In this study, silver nanoparticles were synthesized via a green method using an ethanolic extract of saffron (*Crocus sativus* L.), which is rich in bioactive compounds. The use of ethanol as a solvent enabled the extraction of a wider range of phenolic and flavonoid compounds, which acted as both reducing and stabilizing agents during the synthesis process. The formation of nanoparticles was also visually confirmed by a color change of the solution from colorless to yellowish-brown. UV-Vis spectroscopy results showed a characteristic peak around 425 nm, confirming the formation of silver nanoparticles. FT-IR analysis further indicated that functional groups such as -OH, C=O, and C-O played a key role in the reduction of silver ions and stabilization of the nanoparticles. XRD analysis confirmed the crystalline nature of silver with a face-centered cubic (FCC) structure, and the particle size was estimated to be approximately 20 nm using the Scherrer equation. SEM images revealed that the nanoparticles were mostly irregular in shape, showed a tendency to agglomerate, and had a relatively broad size distribution. The synthesized nanoparticles show potential for applications in antimicrobial, pharmaceutical, and antioxidant fields. However, further studies, including biological activity assessment and cytotoxicity evaluation, are necessary for practical applications.

INTRODUCTION

Silver nanoparticles are among the most well-known and widely used metallic nanomaterials, attracting significant attention in recent years due to their unique properties [1]. Typically ranging in size from 1 to 100 nanometers, these particles exhibit behaviors that differ markedly from bulk silver, which has led to their broad range of applications [2]. One of the most important characteristics of silver nanoparticles is their strong antimicrobial activity. They are capable of eliminating bacteria, fungi, and even some viruses [3]. Their mechanism of action involves disrupting cell membranes, generating reactive oxygen species (ROS), and interfering with essential enzymatic functions in microorganisms [4]. As a result, they are widely used in wound dressings, disinfectant coatings, medical textiles, and water purification systems. In the medical and pharmaceutical fields, silver nanoparticles have numerous applications [5]. They can be used as carriers in targeted drug delivery systems, enhancing the effectiveness of therapeutic agents. Additionally, they have been

extensively studied in areas such as tissue engineering, bioimaging, and the treatment of diseases like cancer. Their anti-inflammatory properties and ability to accelerate wound healing further increase their importance in medical applications. In industry, silver nanoparticles are utilized in the production of paints, antibacterial coatings, food packaging, electronics, and catalysts [6]. For instance, in food packaging, they help extend the shelf life of products. Moreover, due to their unique optical properties, they are also employed in sensors and optoelectronic devices. Overall, silver nanoparticles play a crucial role in advancing modern technologies and remain a highly active area of research.

In recent years, there has been growing interest in environmentally friendly approaches, leading to increased attention toward the green synthesis of silver nanoparticles [7]. In this method, plant extracts are used instead of harmful chemicals, serving both as reducing and stabilizing agents. Bioactive compounds such as flavonoids, polyphenols, alkaloids, and proteins present in

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these extracts facilitate the conversion of silver ions into nanoparticles while preventing their aggregation [8].

The synthesis process is generally straightforward: a plant extract (prepared using water or ethanol) is mixed with a silver nitrate solution, and parameters such as temperature, pH, and reaction time are controlled [8]. A color change of the solution to yellowish-brown indicates nanoparticle formation, which is associated with surface plasmon resonance (SPR) and can be confirmed by UV-Vis spectroscopy.

Various plants have been used for this purpose, including tea leaves, mint, aloe vera, pomegranate peel, and turmeric [9-11]. Each plant, due to its unique chemical composition, influences the size, shape, and stability of the nanoparticles. Therefore, selecting an appropriate plant source is crucial for controlling the final properties of the nanoparticles. The advantages of this method include simplicity, low cost, minimal need for complex laboratory conditions, and reduced environmental impact. However, challenges such as reproducibility, precise size control, and a complete understanding of the reaction mechanisms still require further investigation. Nevertheless, green synthesis using plant extracts is considered a promising approach for the sustainable production of silver nanoparticles. Saffron (*Crocus sativus* L.), is a valuable medicinal and spice plant known for its beneficial compounds [12]. It contains phenolic and flavonoid compounds with antioxidant, anti-inflammatory, and anticancer effects.

Key chemical constituents of saffron include crocin, picrocrocin, and safranal, all of which possess structures rich in active functional groups [13]. Additionally, the presence of polyphenols enhances its ability to neutralize free radicals and reduce oxidative stress. These properties make saffron a suitable natural source for biological applications. In green synthesis, these compounds can act as reducing agents to convert metal ions into nanoparticles and as stabilizing agents to prevent their aggregation [14]. Functional groups such as hydroxyl (-OH) and carbonyl (C=O) play a crucial role in this process. Therefore, beyond its medicinal value, saffron can also serve as an efficient biological resource in modern processes such as the green synthesis of nanoparticles. This dual functionality strengthens its position in interdisciplinary research and makes it a valuable candidate for developing sustainable technologies. Many studies use aqueous extracts because they are simple, safe, and eco-friendly [15]. However, this approach has limitations, as not all bioactive compounds especially those with lower polarity are effectively extracted in water. For this reason, the use of ethanolic saffron extract can be considered a more effective and innovative approach. Ethanol, as a semi-polar solvent, is capable of extracting a broader range of bioactive compounds, including those that are not readily soluble in water but play a significant role in the reduction and stabilization processes. This can improve the synthesis efficiency and lead to nanoparticles with smaller size, better uniformity, and enhanced stability.

MATERIALS AND METHODS

In this study, saffron (*Crocus sativus* L.) was first obtained from the Torbat Heydariyeh Faculty, and its petals were collected and dried. To prepare the ethanolic extract, 5 g of dried powder was mixed with 200 mL of ethanol and kept at room temperature for 24 hours. The mixture was then filtered using filter paper and subsequently centrifuged to obtain a clear solution. The supernatant was used for the synthesis process.

In the next step, for the synthesis of silver nanoparticles, 35 mL of silver nitrate (AgNO_3) solution with a concentration of 25 mM was mixed with 5 mL of the saffron ethanolic extract. After about 30 minutes, a color change from light yellow to yellowish-brown was observed, indicating the initial formation of silver nanoparticles and the occurrence of surface plasmon resonance. Following nanoparticle formation, the particles were separated by centrifugation and washed several times with distilled water or ethanol to remove residual plant compounds and free silver ions. The obtained precipitate was then dried and stored for further analysis. To confirm nanoparticle formation, UV-Vis spectroscopy was performed in the range of 200–700 nm. FT-IR analysis was also conducted to identify functional groups such as phenols, flavonoids, hydroxyl, and carbonyl groups. The crystalline structure of the nanoparticles was examined using XRD, and the approximate particle size was calculated using the Debye Scherrer equation. Additionally, SEM images were used to evaluate the morphology, shape, and distribution of the nanoparticles.

RESULTS AND DISCUSSION

During the synthesis of silver nanoparticles using the ethanolic extract of saffron, the first noticeable sign is the change in the color of the solution, which is clearly visible in the image. Initially, the silver nitrate solution is completely clear and colorless (left vial). After adding the saffron extract (middle vial), the solution turns light yellow, indicating the presence of bioactive compounds in the extract. As the two are mixed, the color gradually changes to yellowish-brown (right vial). This color change is an important indication of the formation of silver nanoparticles and is related to a phenomenon known as SPR [16]. In this process, the free electrons on the surface of the nanoparticles respond to light and oscillate collectively, resulting in a characteristic optical absorption in the visible region [17]. The intensity and position of this color can even provide information about the size and shape of the nanoparticles. In fact, this simple transition from a colorless solution to a brownish one indicates that silver ions (Ag^+) have been reduced to metallic silver (Ag^0). For this reason, the color change is considered one of the first signs of successful synthesis and is usually confirmed by UV-Vis analysis. In the UV-Vis spectrum, a distinct peak appears around 425 nm, which corresponds to the surface plasmon resonance of silver nanoparticles. This peak is one of the most important indicators for confirming nanoparticle formation in a colloidal system. In the spectrum, it appears clearly and indicates the collective oscillation of surface electrons in response to

light, something that occurs only when nanoparticles are successfully formed. Additionally, the absorption observed below 300 nm is likely related to the organic compounds present in the saffron extract, such as phenols and flavonoids, which play a role in the reduction and stabilization processes.

Similarly, in the case of biosynthesis using *Alpinia officinarum* extract, the successful formation of silver nanoparticles was evidenced by the appearance of a distinct SPR absorption peak around 468 nm in the UV–Vis spectrum [18].



Fig. 1. Visual observation of color change during the green synthesis of silver nanoparticles using ethanolic saffron extract: (left) colorless AgNO_3 solution, (middle) light yellow saffron extract, and (right) yellowish-brown solution indicating the formation of silver nanoparticles due to SPR.

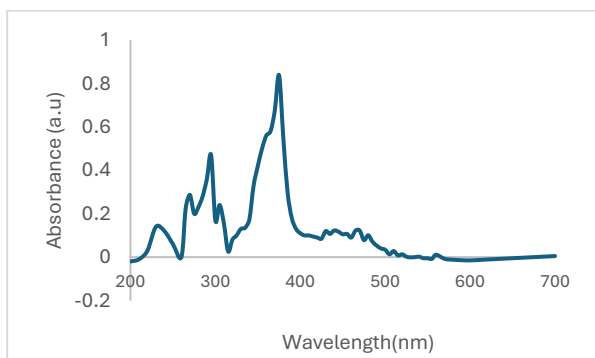


Fig. 2. UV–Vis absorption spectrum of silver nanoparticles synthesized using ethanolic saffron extract, showing a characteristic SPR peak around 425 nm, confirming the formation of silver nanoparticles.

The XRD analysis results clearly show that silver nanoparticles with a well-defined crystalline structure have been successfully formed. In the diffraction pattern, peaks appear at around 38° , 44° , 64° , and 77° , which correspond to the (111), (200), (220), and (311) crystal planes, respectively. These peaks are consistent with the face-centered cubic (FCC) structure of metallic silver, indicating that silver ions have been reduced to metallic silver during the synthesis process. The high intensity of the (111) peak suggests that this crystal plane is dominant

in the sample. Additionally, the relatively sharp peaks indicate the crystalline nature of the nanoparticles, while their broadening reflects the small size of the particles at the nanoscale. The particle size was estimated using the Scherrer equation and was found to be approximately 20 nm.

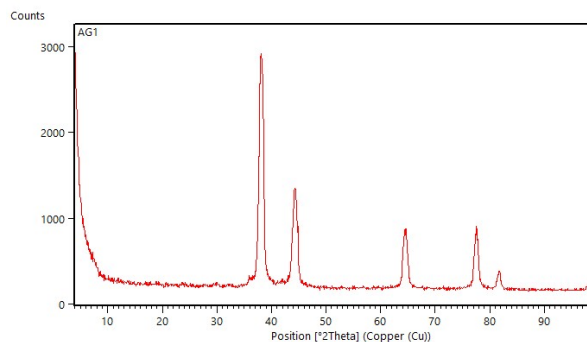


Fig. 3. XRD pattern of the synthesized silver nanoparticles, showing characteristic diffraction peaks corresponding to the (111), (200), (220), and (311) planes of face-centered cubic (FCC) silver, confirming their crystalline structure.

In the SEM image, the first thing that stands out is the irregular and aggregated structure of the particles. The nanoparticles appear as clusters with undefined shapes and rough edges, suggesting that after formation, they have stuck together and formed larger agglomerates. This is quite common in green synthesis and usually happens due to interactions between particles as well as the drying process.

In terms of particle shape, it is difficult to clearly distinguish individual nanoparticles at this magnification. However, some particles appear to have a tendency toward semi-spherical shapes, while others are more irregular. This variation indicates that the growth of the particles was not strictly controlled in the presence of saffron extract compounds, which act as both reducing and stabilizing agents at the same time. Silver nanoparticles (AgNPs) were biosynthesized using *Moringa oleifera* leaf extract, resulting in predominantly spherical nanoparticles with sizes ranging from 10 to 25 nm [19].

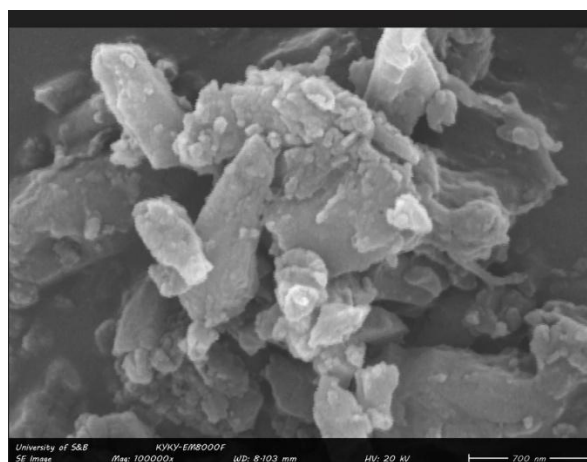


Fig. 4. SEM image of the synthesized silver nanoparticles, showing irregular morphology, noticeable aggregation, and a relatively broad size distribution of the particles.

The FT-IR analysis shows that the functional groups present in the saffron extract undergo noticeable changes during the synthesis process, indicating that these compounds actively participate in the formation of silver nanoparticles. In the region around 3400 cm^{-1} , a broad band is observed, which corresponds to -OH groups commonly found in phenols and alcohols. Changes in the intensity or position of this peak after synthesis suggest that these groups are involved in the reduction of silver ions to metallic silver. In the range of $2920\text{--}2850\text{ cm}^{-1}$, peaks related to C-H stretching vibrations are visible, confirming the presence of organic compounds in the extract. Slight variations in this region are likely due to interactions between these compounds and the nanoparticle surface.

Another significant peak appears around 1625 cm^{-1} , which is associated with carbonyl (C=O) groups or C=C bonds in aromatic structures. Changes in this peak indicate that these groups also contribute to the reduction process and may be involved in binding to the nanoparticle surface. Additionally, peaks observed in the range of $1380\text{--}1050\text{ cm}^{-1}$ correspond to C-O and C-O-C vibrations, which are characteristic of phenolic and flavonoid compounds. Changes in this region suggest that these compounds play a role in stabilizing the nanoparticles and preventing their aggregation.

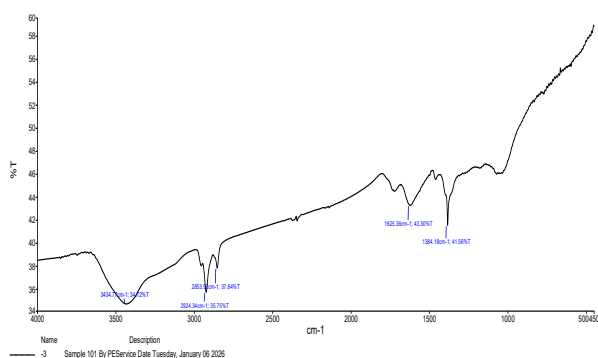


Fig. 5. FT-IR spectrum of the synthesized silver nanoparticles using ethanolic saffron extract, indicating the presence of functional groups involved in the reduction and stabilization processes of silver nanoparticles.

CONCLUSION

In this study, silver nanoparticles were successfully synthesized using a green method with an ethanolic extract of saffron (*Crocus sativus* L.). The presence of rich bioactive compounds especially phenols and flavonoids enabled the extract to both reduce silver ions and prevent nanoparticle aggregation without the need for harmful chemical agents. The color change of the solution from colorless to yellowish-brown also served as a simple and visible indication of nanoparticle formation. The results of different analyses clearly supported these findings. In the UV-Vis spectrum, a distinct peak appeared in the range of approximately $400\text{--}420\text{ nm}$, confirming the formation of silver nanoparticles. FT-IR analysis showed that functional groups such as -OH , C=O , and C-O played an important role in the reduction and stabilization processes. XRD analysis confirmed the crystalline structure of silver

with a face-centered cubic (FCC) arrangement, while SEM images revealed that the particles were not uniform in shape and showed some degree of aggregation. The synthesized nanoparticles, due to their structural and surface properties, have promising potential in areas such as antimicrobial, pharmaceutical, and antioxidant applications. However, further investigations are still needed for practical use. For future work, studies on antibacterial and antifungal activity, cytotoxicity evaluation, and additional biological assessments under in vitro and in vivo conditions are recommended. These studies can provide a clearer understanding of the efficiency and safety of these nanoparticles and help pave the way for their practical applications.

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