

Thermodynamic and Kinetic Study of Silver Nanoparticles Binding with BSA Using UV–Visible and Conductivity Methods

Ashoke Hazra*

Received: 06 April 2025 / Received in revised form: 17 July 2025, Accepted: 19 July 2025, Published online: 25 July 2025

Abstract

The thermodynamic and kinetic behavior of green-synthesized silver nanoparticles (AgNPs) interacting with bovine serum albumin (BSA) was studied using UV–Visible spectroscopy and conductivity techniques. AgNPs prepared from *Azadirachta indica* leaf extract showed a surface plasmon resonance (SPR) band at 430 nm. Binding constants (K) were measured at five temperatures (298–318 K). Van't Hoff analysis provided ΔH and ΔS , while ΔG was evaluated at each temperature. The interaction is spontaneous ($\Delta G < 0$) and shows an endothermic signature from the derived ΔH . Conductivity and UV–Visible spectroscopy (not shown) support complex formation and changes in protein environment upon binding. We investigated how temperature affects the binding between the molecules. Our examination of the thermodynamic data verified that the contact is endothermic—that is, it takes in heat from its environment—and occurs spontaneously. We also kept an eye on electrical conductivity variations. Complex formation was confirmed by conductivity studies.

Keywords: BSA, Conductivity, kinetics, Silver nanoparticles, Thermodynamics, UV–Visible spectroscopy

Introduction

Nanoparticle–protein interactions are critical in nanobiotechnology, influencing distribution, stability, and bioactivity (Hazra *et al.*, 2025). Silver nanoparticles (AgNPs) are widely studied for their optical, catalytic, and antimicrobial properties (Jaiswal *et al.*, 2022). Bovine serum albumin (BSA) serves as a model protein due to its structural similarity to human serum albumin and well-characterized binding properties (Kandiah & Chandrasekhar, 2021).

Thermodynamic and kinetic studies provide insights into spontaneity, enthalpic, and entropic contributions of AgNP–protein interactions (Llusco *et al.*, 2020; Pedroso *et al.*, 2022; Tian *et al.*, 2022). Protein adsorption may alter conformation, affecting nanoparticle stability and bioactivity (Kiarashi & Yasamineh, 2024). UV–Vis spectroscopy and conductivity measurements are effective tools to study these interactions (Yu *et al.*, 2022; Ebrahimi *et al.*, 2023).

Ashoke Hazra*

Department of Chemistry, A.K.P.C. Mahavidyalaya, Bengai, Hooghly, West Bengal, India.

*E-mail: hazra.ashoke@gmail.com

This study investigates the binding of green-synthesized AgNPs with BSA. With the help of Van't Hoff plots, thermodynamic parameters (ΔH , ΔS , ΔG) were derived, and kinetic parameters were evaluated using pseudo-first-order analysis (Talabani *et al.*, 2021; Omer *et al.*, 2022; Svobodova-Sedlackova *et al.*, 2022; de Carvalho-Silva *et al.*, 2023; Wirwis & Sadowski, 2023; Hermanto *et al.*, 2024; Mansoor *et al.*, 2024).

Materials and Methods

Silver nitrate (AgNO_3), BSA, and neem leaves were used. Neem extract was obtained by boiling 10 g of dried leaves in 100 mL of distilled water. 1 mM AgNO_3 solution was mixed with the extract (1:1) at 40°C, producing a brown color indicative of AgNP formation.

UV–Vis spectra were recorded from 300–700 nm (formation was confirmed by SPR at ~430 nm). Conductivity was measured using a digital conductivity meter. BSA (1×10^{-4} M) was titrated with AgNPs at temperatures 25–55°C. The binding constants (K) were determined using the Benesi–Hildebrand equation, while the thermodynamic parameters were derived from van't Hoff analysis. Conductivity measurements were performed using a properly calibrated conductivity meter. The thermodynamic parameters were further evaluated through linear regression of the van't Hoff plots ($\ln K$ versus $1/T$).

Results and Discussion

UV–Visible Spectroscopy

The surface plasmon resonance (SPR) band observed at 430 nm is a characteristic optical signature confirming the successful formation of silver nanoparticles (AgNPs). This spectral band is the result of a phenomenon where incoming light energizes the free-moving electrons on the nanoparticle's surface, causing them to oscillate in a synchronized, wave-like motion. The position and intensity of the SPR peak depend on particle size, shape, and the surrounding dielectric environment. A distinct red shift in the SPR peak upon the addition of bovine serum albumin (BSA) indicates a change in the local refractive index around the AgNPs. This spectral shift provides clear evidence of BSA adsorption onto the nanoparticle surface, leading to the formation of AgNP–BSA complexes. The protein molecules interact with AgNPs through electrolyte.



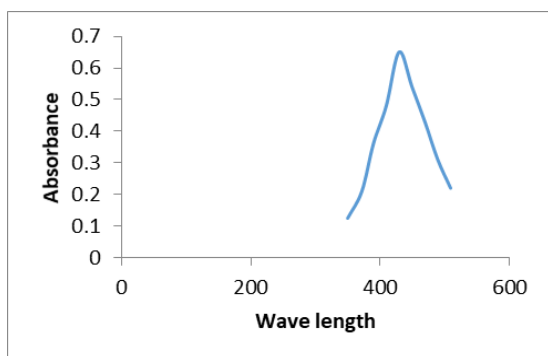


Figure 1. UV-Visible absorption spectrum of AgNP-BSA complex.

The SPR band at 430 nm confirmed AgNP formation. A red shift upon BSA addition indicated nanoparticle-protein complex formation.

Conductivity Study

The electrical conductivity of a solution generally increases with temperature due to enhanced ionic mobility. As the temperature rises, the viscosity of the solvent decreases, allowing ions to move more freely and carry charge efficiently. In systems containing bovine serum albumin (BSA) and silver nanoparticles (AgNPs), this temperature-dependent conductivity reflects both ionic and colloidal interactions. The diffusion of ions over the surfaces of nanoparticles is facilitated by the higher temperature-induced increase in kinetic energy. As a result, interparticle resistance decreases and the electrostatic double layer around AgNPs becomes more dynamic. BSA molecules, acting as stabilizing agents, adsorb onto the AgNP surfaces through electrostatic and hydrophobic interactions. This adsorption modifies the surface charge distribution, influencing the zeta potential and ionic conduction pathway. Elevated temperature may induce conformational changes in BSA, altering its binding affinity to AgNPs. Such changes can enhance charge transfer between the protein corona and the nanoparticle surface. The overall result is an increase in conductivity due to improved ion transport and interfacial electron exchange. This temperature-driven enhancement demonstrates the coupling between protein-nanoparticle interactions and electrochemical behavior (Rudayni *et al.*, 2022; Spirito *et al.*, 2022; Sugimori *et al.*, 2022; Bulusu & Cleary, 2023; Dorn *et al.*, 2024; Mao *et al.*, 2024).

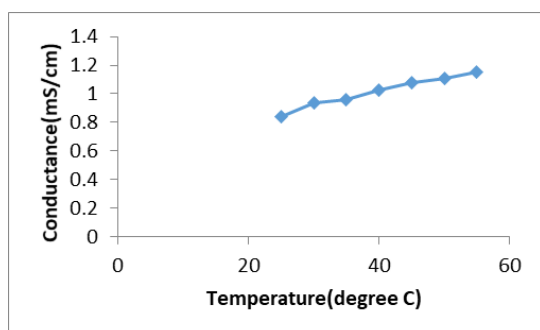


Figure 2. Conductivity variation with temperature for AgNP-BSA complex.

Conductivity increased with temperature, suggesting enhanced ion mobility and electrostatic interaction between BSA and AgNPs.

Different Thermodynamic Parameters

Table 1. Data on Binding constants and Change in Gibbs free energy at various temperatures

Temperature (K)	Binding Constant K (M ⁻¹)	ln K	ΔG (kJ/mol)
298	25,000	10.127	-25.091
303	32,000	10.373	-26.134
308	41,000	10.621	-27.200
313	50,000	10.820	-28.158
318	60,000	11.002	-29.090

Van't Hoff analysis (linear regression of ln K vs 1/T) yields:

$$\Delta H = 34.66 \text{ kJ mol}^{-1}$$

$$\Delta S = 200.64 \text{ J mol}^{-1} \text{ K}^{-1}$$

Thermodynamic Study at Different Temperatures

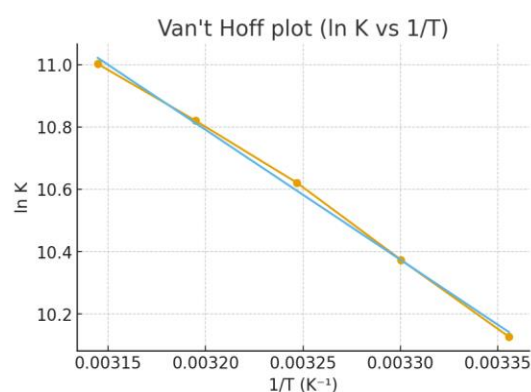


Figure 3. Van't Hoff plot (ln K vs 1/T).

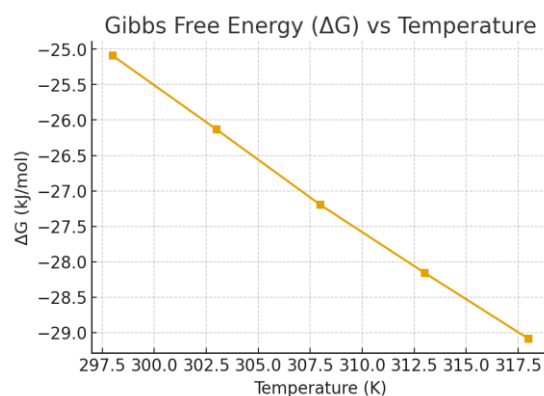


Figure 4. ΔG vs Temperature plot.

The positive enthalpy change ($\Delta H = 34.66 \text{ kJ mol}^{-1}$) shows that the binding process is endothermic. The positive entropy value ($\Delta S = 200.64 \text{ J mol}^{-1} \text{ K}^{-1}$) indicates an increase in disorder during binding, likely caused by the release of water molecules from the surface of the nanoparticles or the protein. The negative ΔG values at all temperatures confirm that the binding occurs spontaneously.

Overall, these thermodynamic results suggest that hydrophobic interactions play a major role in the binding process.

Conclusion

By measuring how strongly silver nanoparticles (AgNPs) bind to BSA at various temperatures, we determined the key thermodynamic forces at play. Our analysis revealed that the binding is spontaneous, with ΔH , ΔS , and ΔG values characteristic of an interaction governed by hydrophobic effects. The accompanying plots provide a graphical summary of these calculations. BSA binds spontaneously to green-synthesized AgNPs via hydrophobic and electrostatic interactions. UV–Vis and conductivity studies confirm stable AgNP–BSA complex formation, relevant for biomedical and nanotherapeutic applications.

The present study demonstrates a successful investigation of the thermodynamic and kinetic behavior of biosynthesized silver nanoparticles (AgNPs) interacting with bovine serum albumin (BSA). UV–Visible spectroscopy confirmed AgNP formation through a distinct surface plasmon resonance (SPR) band around 430 nm. A red shift and peak broadening upon BSA addition indicated stable nanoparticle–protein complex formation. Conductivity measurements revealed enhanced ionic mobility and electrostatic stabilization in the AgNP–BSA system. The binding constant increased with temperature, suggesting an endothermic and spontaneous interaction. The thermodynamic study showed positive ΔH and ΔS values, meaning that hydrophobic and electrostatic forces mainly drive the interaction. The negative ΔG values at all temperatures confirm that the binding between AgNPs and BSA happens spontaneously. The Van't Hoff plot exhibited linearity, validating the equilibrium nature of the binding process. The kinetic data supported a pseudo-first-order mechanism consistent with controlled diffusion and surface adsorption. The stability of the AgNP–BSA complex enhances the potential biocompatibility of silver nanoparticles. Such interactions are crucial for understanding nanoparticle behavior in biological systems and drug delivery contexts (Ruchin *et al.*, 2022; Kiedrowicz *et al.*, 2023; Kulkarni *et al.*, 2023; AlShammasi *et al.*, 2024; Ravoori *et al.*, 2024).

Overall, the combined UV–Visible and conductivity approaches provided complementary insights into the thermodynamic stability and kinetic control of AgNP–BSA conjugation.

Acknowledgments: The author gratefully acknowledges the guidance, support, and facilities provided by the institution, A.K.P.C. Mahavidyalaya, and colleagues throughout this work. The author is also thankful to Mr. Arnab Hazra, 3rd year Mechanical Engg student of NIT Durgapur, West Bengal, for drawing the necessary graphs and helping with data analysis for this study.

Conflict of interest: None

Financial support: None

Ethics statement: None

References

- AlShammasi, H., Alkhaldi, G., Alharbi, L., & Alyami, M. (2024). Shifts in parental perception of pediatric patients needing dental rehabilitation under general anesthesia post-pandemic. *Turkish Journal of Public Health Dentistry*, 4(1), 29–35. doi:10.51847/jAv4cm0TTn
- Bulusu, A., & Cleary, S. D. (2023). Comparison of dental caries in autistic children with healthy children. *Annals of Journal of Dental & Medical Assistance*, 3(2), 14–19. doi:10.51847/wa2pZXE4RJ
- de Carvalho-Silva, J., Fernandes da Silva, M., Silva de Lima, J., Souza Porto, T., Bezerra de Carvalho Jr, L., & Converti, A. (2023). Thermodynamic and kinetic investigation on *Aspergillus ficuum* tannase immobilized in calcium alginate beads and magnetic nanoparticles. *Catalysts*, 13(9), 1304.
- Dorn, G. A., Poznyakovsky, V. M., Danko, N. N., Vladimirovna, P. E., & Tokhiriyon, B. (2024). A study of assessing the impact of pantothenic acid, embryotoxicity, and teratogenicity. *International Journal of Veterinary Research & Allied Sciences*, 4(2), 5–13. doi:10.51847/tvX0TaYc7D
- Ebrahimi, B., Amini, S., Tofighi, M. R., & Ghasemzadeh, H. (2023). Synthesis and characterization of an albumin nanoparticle with intrinsic antibacterial property. *Artificial Cells, Nanomedicine, and Biotechnology*, 51(1), 106–116. doi:10.1080/21691401.2023.2185292
- Hazra, A., Malik, A., Banerjee, R., Chakraborty, A., & Khatun, S. (2025). Synthesis of ZnO nanoparticles and interaction with protein and surfactants. *Journal of Biochemical Technology*, 16(2), 11–15. doi:10.51847/HEDzHaWVT
- Hermanto, D., Ismillayli, N., Fatwa, D. H., Zuryati, U. K., Muliasari, H., & Wirawan, R. (2024). Bio-mediated electrochemical synthesis of silver nanoparticles using green tea (*Camellia sinensis*) leaves extract and their antibacterial activity. *South African Journal of Chemical Engineering*, 47(1), 136–141.
- Jaiswal, S., Manas, A., Pandey, A. K., Priya, S., & Sharma, S. K. (2022). Engineered nanoparticles-protein interactions influence protein structural integrity and biological significance. *Nanomaterials*, 12(7), 1214. doi:10.3390/nano12071214
- Kandiah, M., & Chandrasekhar, K. N. (2021). Green synthesis of silver nanoparticles using *Catharanthus roseus* flower extracts and the determination of their antioxidants, antimicrobial and photocatalytic activity. *Journal of Nanotechnology*, 2021(1), 5512786. doi:10.1155/2021/5512786
- Kiarashi, M., & Yasamineh, S. (2024). Albumin nanoparticles are a promising drug delivery system in dentistry. *Journal Name*, 23, 122.
- Kiedrowicz, M., Dembowska, E., Banach, J., Safranow, K., & Pynka, S. (2023). Evaluating periodontal health in type 2 diabetics with chronic complications. *Annals of Orthodontics & Periodontics Specialty*, 3, 19–27. doi:10.51847/axdaRLPSoJ
- Kulkarni, S., Zope, S., Suragimath, G., Varma, S., & Kale, A. (2023). The influence of female sex hormones on

- periodontal health: a regional awareness study. *Annals of Orthodontics & Periodontics Specialty*, 3, 10–18. doi:10.51847/v4EFMh6WEf
- Llusco, A., Grageda, M., & Ushak, S. (2020). Kinetic and thermodynamic studies on synthesis of Mg-doped LiMn₂O₄ nanoparticles. *Nanomaterials*, 10(7), 1409.
- Mansoor, S., Asimullah Khan, Nabi, G., Hussian, S., Javid, Z., & Akbar Jan, F. (2024). Synthesis, characterization, kinetic and thermodynamic study of photocatalytic degradation of malachite green dye using MnO₂ nanoparticles as catalyst. *Chemistry Africa*, 7(3), 1575–1583.
- Mao, J., Zhang, T., Zhang, G., Wang, M., Liu, C., & Wang, M. (2024). The impact of cold storage on the survival and viability of parasitoid bee pupae and whole insects. *International Journal of Veterinary Research & Allied Sciences*, 4(2), 20–26. doi:10.51847/d0aYiMtX37
- Omer, M., Khan, B., Khan, I., Alamzeb, M., Zada, F. M., Shah, R., Algarni, M., & Simal-Gandara, J. (2022). Equilibrium, kinetic and thermodynamic studies for the adsorption of metanil yellow using carbonized pistachio shell-magnetic nanoparticles. *Water*, 14(24), 4139.
- Pedroso, R., Pedreiro, E., Gremiao, M. P. D., & Chorilli, M. (2022). Curcuminoids loaded in BSA nanoparticles. *Molecules*, 27(9), 2758. doi:10.3390/molecules27092758
- Ravoori, S., Sekhar, P. R., Pachava, S., Pavani, N. P. M., Shaik, P. S., & Ramanarayana, B. (2024). Perceived stress and depression among oral cancer patients: a hospital-based cross-sectional study. *Turkish Journal of Public Health Dentistry*, 4(1), 1–5. doi:10.51847/FoK9xAl1JW
- Ruchin, A., Alekseev, S., Khapugin, A., & Esin, M. (2022). The fauna and diversity of ground beetles (Coleoptera, Carabidae) in meadow ecosystems. *Entomological Letters*, 2(1), 1–11. doi:10.51847/xiIPPVCe56
- Rudayni, H. A., Basher, N. S., Al-Keridis, L. A., Ibrahim, N. A., & Abdelmageed, E. (2022). Exploring the effectiveness of *Ocimum basilicum* extracts in mosquito larvae management. *Entomological Letters*, 2(1), 12–18. doi:10.51847/upImR4jWMM
- Spirito, F. D., Iacono, V. J., Alfredo, I., Alessandra, A., Sbordone, L., & Lanza, A. (2022). Impact of COVID-19 awareness on periodontal disease prevention and management. *Asian Journal of Periodontics & Orthodontics*, 2, 16–26. doi:10.51847/t8D9TJGOCU
- Sugimori, T., Yamaguchi, M., Kikuta, J., Shimizu, M., & Negishi, S. (2022). The biomechanical and cellular response to micro-perforations in orthodontic therapy. *Asian Journal of Periodontics & Orthodontics*, 2, 1–15. doi:10.51847/Z9adSJ59rj
- Svobodova-Sedlackova, A., Huete-Hernandez, S., Calderón, A., Barreneche, C., Gamallo, P., & Fernandez, A. I. (2022). Effect of nanoparticles on the thermal stability and reaction kinetics in ionic nanofluids. *Nanomaterials*, 12(10), 1777.
- Talabani, R. F., Hamad, S. M., Barzinjy, A. A., & Demir, U. (2021). Biosynthesis of silver nanoparticles and their applications in harvesting sunlight for solar thermal generation. *Nanomaterials*, 11(9), 2421.
- Tian, J., Shi, Z., & Wang, G. (2022). Thermodynamic and kinetic binding behaviors of human serum albumin to silver nanoparticles. *Materials (Basel)*, 15(14), 4957. doi:10.3390/ma15144957
- Wirwis, A., & Sadowski, Z. (2023). Green synthesis of silver nanoparticles: optimizing green tea leaf extraction for enhanced physicochemical properties. *ACS Omega*, 8(33), 30532–30549.
- Yu, G., Cheng, Y., & Duan, Z. (2022). Study of the electrical conductivity characteristics of micro and nano-ZnO/LDPE composites. *Molecules*, 27(12), 3674. doi:10.3390/molecules27123674