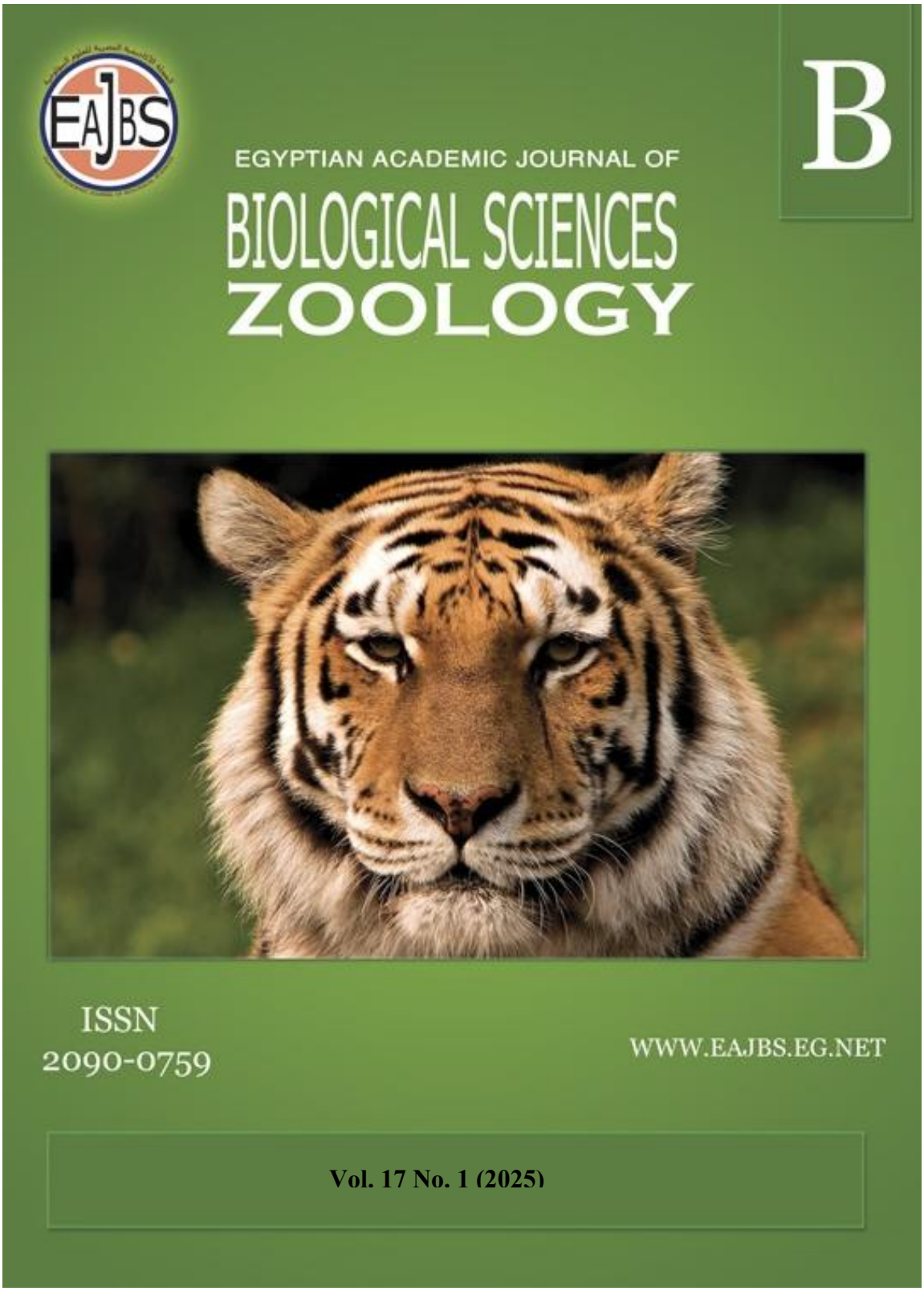
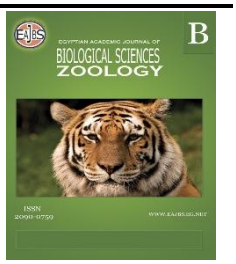


Citation: *Egypt. Acad. J. Biolog. Sci. (B. Zoology) Vol. 17(1) pp:167-176 (2025)*
DOI: 10.21608/EAJBSZ.2025.419842

Egypt. Acad. J. Biolog. Sci., 17(1):167-176(2025)
Egyptian Academic Journal of Biological Sciences
B. Zoology
ISSN: 2090 – 0759

http://eajbsz.journals.ek b.eg/



Effect of Green Silver Nanoparticles Combined with *Moringa oleifera* leaf Extract on the liver and Pancreas in Streptozotocin-Induced Diabetic Rats

Mohammed F. Al Khuzaee [1], Salim M. El Hamidy [1,2], Muhamed A. El Nobey [1, *], Abdulkader M. Shaikh Omar [1,2], Khaled Al Hunduwan [1] and Abdulelah A. Al Harthi[1]
[1]Department of Biological Sciences, Faculty of Science, King Abdulaziz University, Jeddah, Saudi Arabia.
[2]Princess Doctor Najla Bint Saud Al Saud Distinguished Research Centre for Biotechnology, King Abdulaziz University, Jeddah, Saudi Arabia.
***E-mail:** melnobey@stu.kau.edu.sa

ARTICLE INFO
Article History
Received:12/2/2025
Accepted:29/3/2025
Available:4/4/2025

Keywords:
Diabetic Mellitus, Green Silver Nanoparticles, Liver Function, *Moringa oleifera* leaf, Streptozotocin, Pancreas.

ABSTRACT

Diabetes mellitus (DM) is a global health challenge, with diabetic liver injury being a critical complication driven by oxidative stress, hyperglycemia, and inflammation. This study evaluates the synergistic therapeutic potential of green silver nanoparticles (Ag NPs) synthesized with *Moringa oleifera* leaf extract to mitigate liver dysfunction in streptozotocin (STZ)-induced diabetic rats. Ag NPs were characterized using transmission electron microscopy (TEM) to determine their morphology and size. Fifty adult albino rats were divided into five groups: Group I (control), Group II (diabetic), Group III (diabetic treated with Ag NPs), Group IV (diabetic treated with *M. oleifera*), and Group V (diabetic receiving combination treatment). Biochemical parameters, including glucose, HbA1c, insulin, ALT, AST, albumin, total protein, total bilirubin, GSH, SOD, CAT, and MDA, were assessed alongside histological examinations of the liver and pancreas. STZ-induced diabetic rats exhibited significant hyperglycaemia, elevated HbA1c levels, impaired liver function, and oxidative stress. Treatment with Ag NPs and *M. oleifera* leaf extract improved glucose metabolism, restored antioxidant defences, and normalized liver biomarkers. The combined treatment demonstrated superior efficacy, significantly reducing serum glucose, HbA1c, ALT, and AST levels while enhancing insulin, GSH, and SOD levels. Histological analysis confirmed the amelioration of STZ-induced hepatic and pancreatic structural damage. These findings underscore the therapeutic potential of green silver nanoparticles and *M. oleifera* leaf extract as a novel intervention targeting oxidative stress, inflammation, and liver dysfunction in diabetes.

INTRODUCTION

Diabetes mellitus is a global metabolic disorder that poses significant health challenges due to its chronic implications, affecting multiple organs, including the liver (Mohamed *et al*., 2016; Chung *et al*., 2020; Yameny, 2024). Among these complications, diabetic liver injury is a critical concern, driven by persistent hyperglycaemia, oxidative stress, and inflammation (Rohm *et al*., 2022; Zhang, 2024). These factors lead to structural and functional hepatic impairments, contributing to the progression of diabetes-related complications (Alfieri *et al*., 2024). Despite advancements in therapeutic strategies, effective treatments targeting both oxidative stress and inflammation in diabetic liver damage remain limited (Bae *et al*., 2023; Weinberg Sibony *et al*., 2024).

Nanotechnology has emerged as a promising field in biomedical research, offering innovative solutions for disease management (Reddy Gangadi, 2024). Green silver nanoparticles (Ag NPs), synthesized through eco-friendly and cost-effective methods, have

garnered attention for their potent antioxidant, anti-inflammatory, and antimicrobial properties (Srivastava *et al*., 2024). Unlike conventionally synthesized nanoparticles, green Ag NPs utilize plant extracts as reducing and stabilizing agents, minimizing toxicity and environmental impact (Villagrán *et al*., 2024; Zulfiqar *et al*., 2024).

Moringa oleifera is a medicinal plant widely recognized for its hepatoprotective properties (Pareek *et al*., 2023). Its leaf extract contains flavonoids, phenolics, and vitamins, which exhibit antioxidant and anti-inflammatory effects (Chiș *et al*., 2023; Chatzimitakos *et al*., 2024). Combining green Ag NPs with *M. oleifera* extract offers a synergistic strategy to neutralize reactive oxygen species (ROS), reduce inflammation, and protect against diabetic liver injury (Virk *et al*., 2023; Muhammad *et al*., 2023). This study investigates the effects of Ag NPs, *M. oleifera* extract, and their combination on liver function and histology in STZ-induced diabetic rats. By exploring this innovative therapeutic approach, this research aims to provide insights into novel strategies for managing diabetic liver complications.

MATERIALS AND METHODS

Characterization of Ag NPs:

The morphology and relative dimensions of Ag NPs were analysed using a transmission electron microscope (TEM) (JEOL Ltd., Tokyo, Japan). An aqueous suspension of nanoparticles was applied to a carbon-coated copper grid, dried, and subsequently examined.

Induction of Diabetes:

Diabetes mellitus was induced in rats fasted for 24 hours through a single intraperitoneal (IP) injection of STZ (50 mg/kg) dissolved in cold 0.1% M citrate buffer (pH 4.5). Following the injection, the rats were provided with sucrose (15 g/L) in their drinking water for 24 hours to reduce the risk of hypoglycaemia-induced mortality. Diabetes was confirmed by measuring fasting blood glucose (FBG) levels three days after STZ administration. Rats with FBG levels exceeding 300 mg/dL were classified as diabetic and included in the experiment

Experimental Design:

Fifty rats were obtained from the Animal Section of the King Fahad Medical Research Center at King Abdulaziz University and were classified into five groups (n = 10) as follows:

- **Group I**: Non-Diabetic Control received 0.5 mL citrate buffer intraperitoneally (IP).
- **Group II:** Diabetic Control received STZ (50 mg/kg, IP).
- **Group III**: Diabetic rats received STZ (50 mg/kg, IP) and were treated with Ag NPs (0.2 mg/kg, orally).
- **Group IV**: Diabetic rats received STZ (50 mg/kg, IP) and were treated with *M. oleifera* (0.2 mg/kg, orally).
- **Group V**: Diabetic rats received STZ (50 mg/kg, IP) and were treated with a combination of Ag NPs (0.2 mg/kg, orally) and *M. oleifera* (0.2 mg/kg, orally).

Glucose Metabolism:

Following blood collection, blood glucose levels were measured using the method described by Middleton and Griffiths (1957). Plasma insulin levels were quantified using the ELISA method with an insulin ELISA kit. Glycosylated haemoglobin (HbA1c) levels were determined using the method of Bannon (1982) with a commercial diagnostic kit.

Liver Function Biomarkers Assessment:

Serum alanine aminotransferase (ALT) and aspartate aminotransferase (AST) levels were measured using the method of Reitman and Frankel (1957). Albumin (ALB) levels were determined using the method of Busher (1990). Total protein (TP) was assessed using the method of Peters Jr. (1968). Lactate dehydrogenase (LDH) levels were measured using the method of Stevens et al. (1983). Total bilirubin (TB) was determined according to the method of Doumas et al. (1985).

Antioxidant Biomarker Assessment:

The antioxidant parameters were examined as follows: blood samples were collected and analyzed using the method of Misra and Fridovich (1972) for SOD and CAT, the method of Draper and Hadley (1990) for MDA, and the protocol of Moron et al. (1979) for GSH.

Histopathological Examination:

Liver and pancreatic tissues were fixed in 10% formalin, dehydrated through a graded series of alcohol concentrations, cleared with xylene, and embedded in paraffin wax.

For histological examination, 5 µm sections were cut and stained with Harris' haematoxylin and eosin.

Statistical Analysis:

Data were expressed as the mean ± standard deviation (SD). Statistical analysis was performed using one-way ANOVA, followed by Tukey's post hoc test for multiple comparisons, with $P < 0.05$ considered statistically significant. All analyses were conducted using GraphPad Prism software (version 9.0; GraphPad, USA).

RESULTS

Transmission Electron Microscopy for Ag NPs:

Figure 1, confirms the successful synthesis of Ag NPs. (A) The low-magnification TEM image reveals uniformly dispersed nanoparticles with a spherical morphology. (B) The high-resolution TEM image shows well-defined lattice fringes, indicating the crystalline nature of the Ag NPs. (C) The line profile analysis highlights distinct interplanar spacings, consistent with the characteristic crystallographic planes of silver nanoparticles. These results validate the structural integrity and crystalline properties of the synthesized Ag NPs.

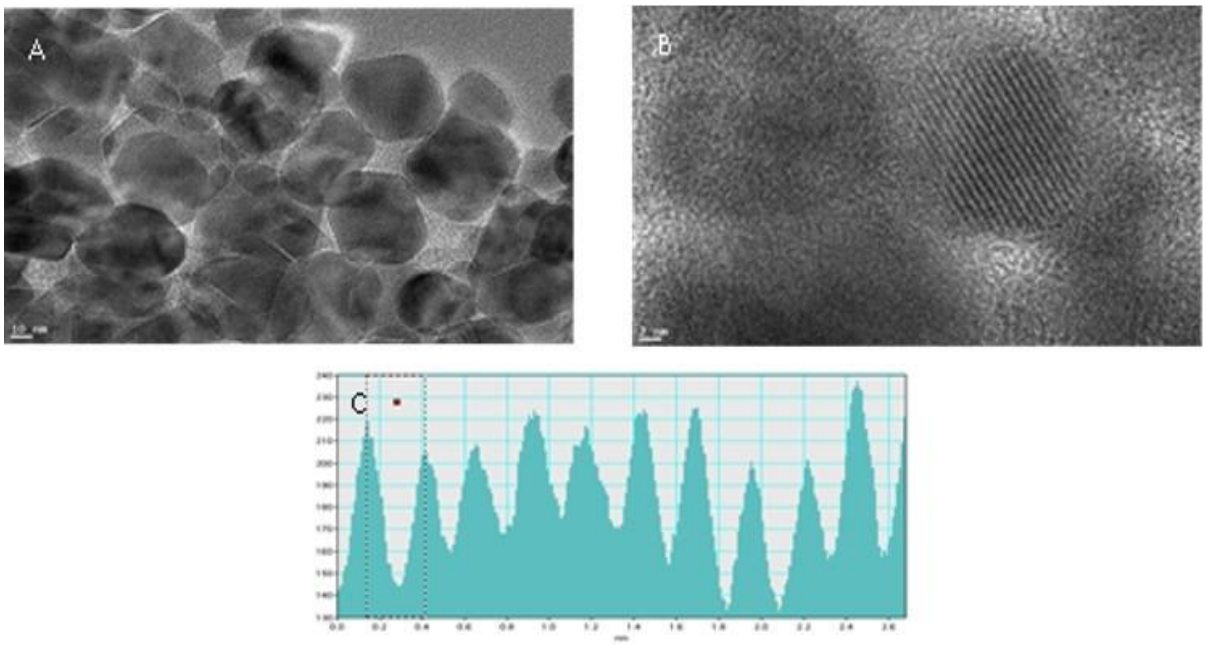


Fig. 1. Transmission Electron Microscopy (TEM) characterization of Ag NPs. (A) TEM image showing the morphology and uniform distribution of Ag NPs. (B) High-resolution TEM image revealing lattice fringes, confirming the crystalline structure of Ag NPs. (C) Line profile analysis illustrating the interplanar spacing characteristics of Ag NPs.

***Moringa. Oleifera* Attenuates Glucose Metabolism Disorders Caused by DM:**

Figure 2, illustrates the effects of Ag NPs, M. oleifera leaf extract, and their combination treatments on serum glucose, insulin, and HbA1c levels in diabetic and control groups. The STZ-induced diabetic group exhibited a significant increase in glucose and HbA1c levels, accompanied by a sharp decrease in insulin levels compared to the control group ($P < 0.001$). Treatment with M. oleifera leaf extract significantly reduced glucose and HbA1c levels while restoring insulin levels compared to the diabetic group ($P < 0.001$). The combination of M. oleifera and Ag NPs further enhanced these effects, showing a superior reduction in glucose and HbA1c levels and an increase in insulin levels compared to individual treatments. Treatment with Ag NPs alone demonstrated significant improvements, although less pronounced than the combination treatment. These findings suggest that M. oleifera and Ag NPs synergistically mitigate hyperglycemia and improve glucose metabolism in diabetic rats.

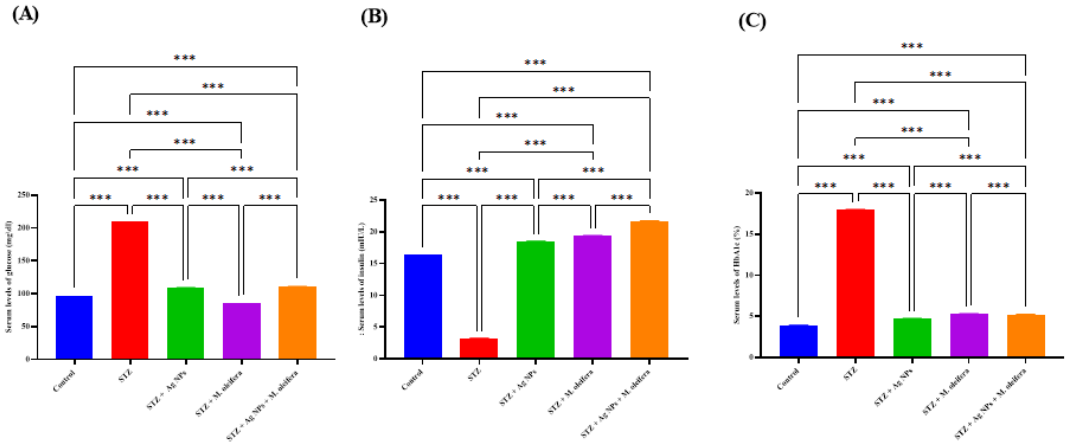


Fig. 2. Effects of M. oleifera and Ag NPs on serum glucose, insulin, and HbA1c levels in STZ-induced diabetic rats. (A) Serum glucose levels, (B) insulin levels, and (C) HbA1c levels. Results are expressed as the mean ± standard deviation (SD). Statistical significance was set at $P < 0.05$.

***M. oleifera* ameliorates Hepatic Impairment Caused by DM**:

Figure 3, demonstrates that the diabetic group exhibited significant liver damage, as indicated by elevated serum levels of ALT and AST and decreased levels of albumin, total protein, and total bilirubin compared to the control group ($P < 0.001$). Treatment with Ag NPs, *M. oleifera* leaf extract, and their combination showed substantial improvements, with the combined Ag NPs + M. oleifera treatment demonstrating the most pronounced normalization of biomarker levels, suggesting synergistic hepatoprotective effects.

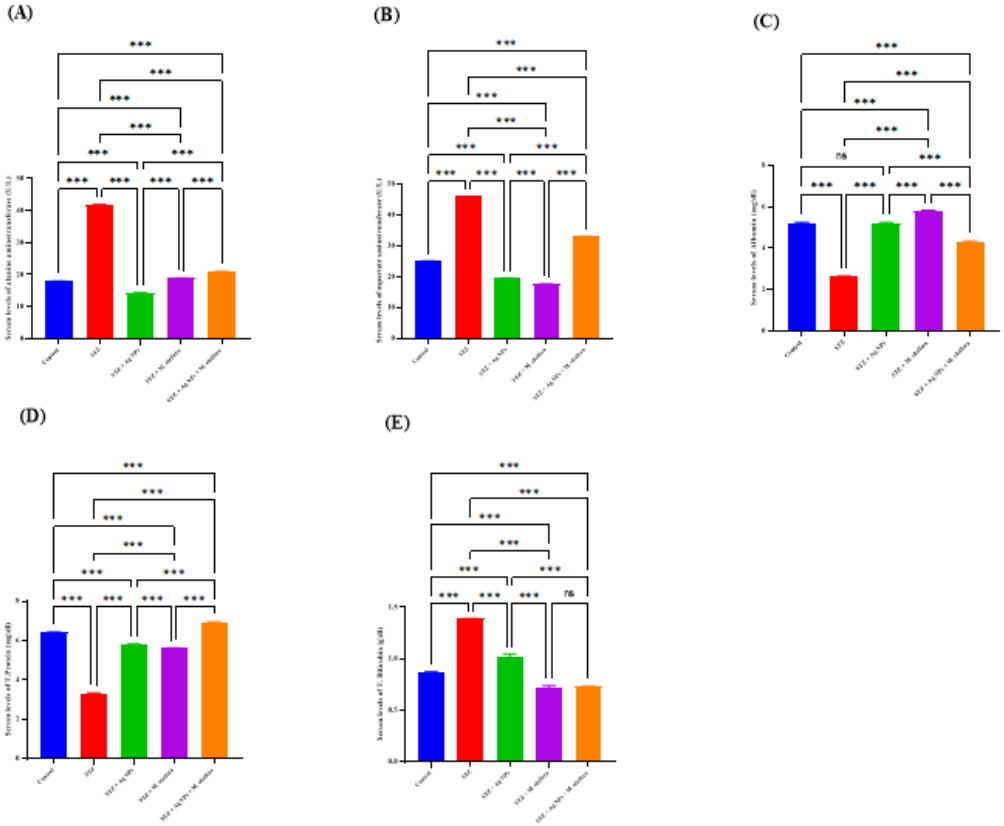


Fig. 3. Effect of Ag NPs, M. oleifera, and their combination on liver function biomarkers in STZ-induced liver damage. The graphs illustrate the serum levels of liver function biomarkers, including (A) ALT, (B) AST, (C) albumin, (D) total protein, and (E) total bilirubin across experimental groups. Results are expressed as the mean ± standard deviation (SD). Statistical significance was set at $P < 0.05$.

Ag NPs and *M. oleifera* mitigate the Diabetic impairments in Antioxidant Biomarkers:

Figure 4, illustrates the effects of Ag NPs and M. oleifera on STZ-induced oxidative stress and antioxidant biomarkers. The STZ-treated group exhibited a significant depletion in serum reduced GSH levels and decreased activities of SOD and CAT, along with a marked increase in MDA, a lipid peroxidation marker. These changes indicate elevated oxidative stress and impaired antioxidant defense. Treatment with Ag NPs and *M. Oleifera*, particularly in combination, reversed these effects by significantly increasing GSH levels, restoring SOD and CAT activities, and reducing MDA levels. The combination of Ag NPs and M. oleifera was more effective than individual treatments, demonstrating a synergistic effect in mitigating oxidative damage and enhancing antioxidant capacity.

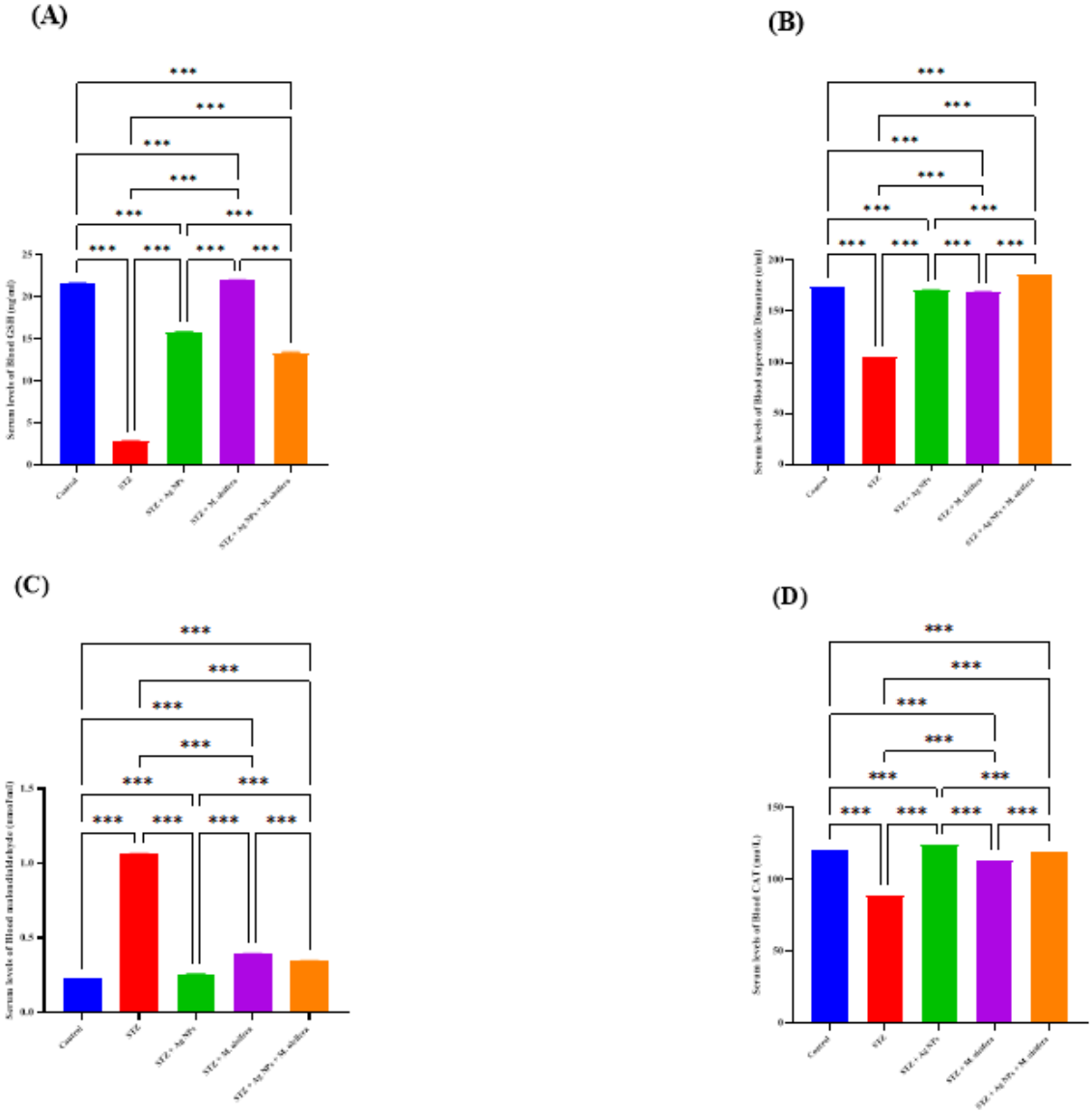


Fig. 4. Effect of Ag NPs, M. oleifera, and their combination on antioxidant biomarkers in STZ-induced liver damage. The graphs illustrate the serum levels of antioxidant biomarkers, including (A) GSH, (B) SOD, (C) MDA, and (D) CAT. Results are expressed as the mean ± standard deviation (SD). Statistical significance was set at $P < 0.05$.

Histological Examination:

Figure 5, illustrates the histological examination of pancreatic sections, showing distinct variations across experimental groups. (A) The control group exhibits normal pancreatic architecture with well-organized acinar cells and intact connective tissue. Meanwhile, the islets of Langerhans (IL) appear lightly stained. (B) Diabetic rats show severe degeneration, including islet atrophy, necrosis, vacuolation, and loss of α- and β-cells, along

with widened interlobular ducts. (C) Diabetic rats treated with silver nanoparticles (Ag NPs) display mild structural improvements, although some vacuolation and cell atrophy persist. (D) Treatment with M. oleifera extract results in better islet preservation, reduced acinar atrophy, and improved distinction between endocrine and exocrine regions. (E) The combination of Ag NPs and M. oleifera leads to significant islet regeneration and reduced vacuolation, suggesting enhanced pancreatic recovery.

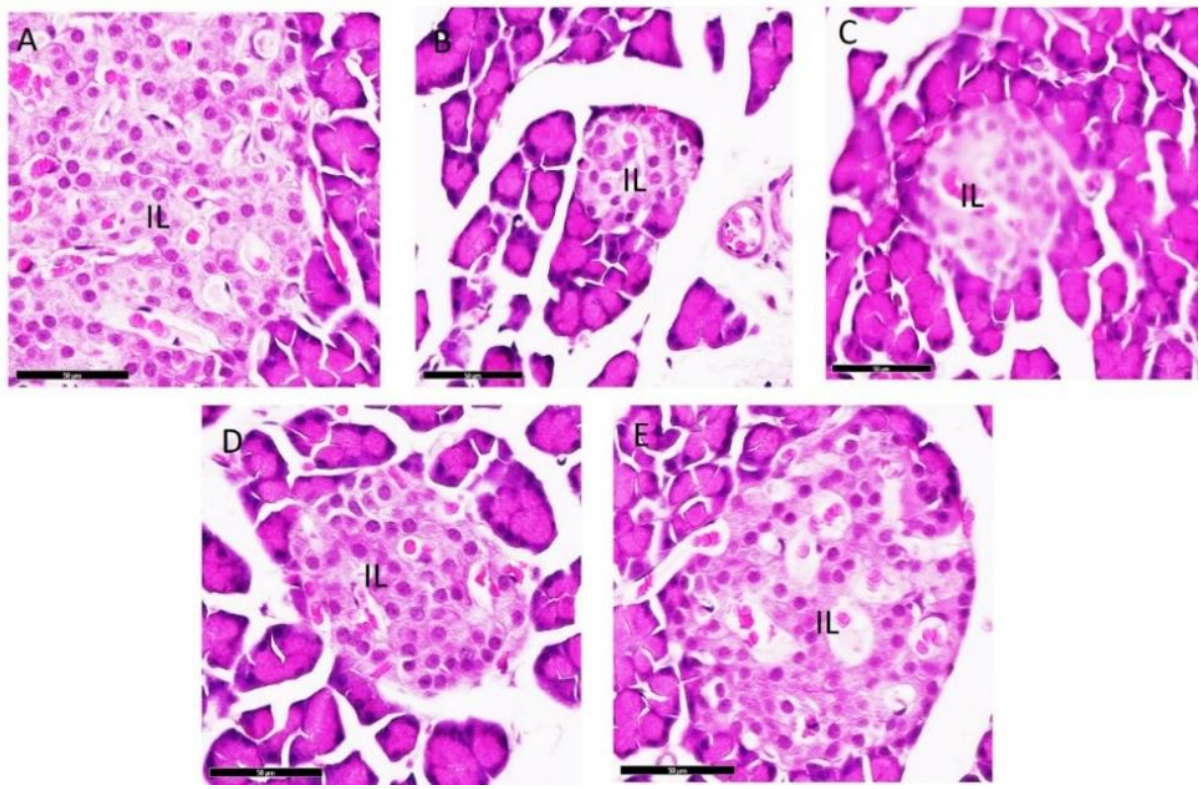


Fig. 5. Photomicrograph of pancreatic tissues stained with H&E (scale bar: 50 µm, 400× magnification). (A) Control group, (B) Diabetic group (received STZ), (C) Diabetic + Ag NPs, (D) Diabetic + *M. oleifer*a leaf extract, (E) Diabetic + Ag NPs + *M. oleifer*a leaf extract. IL: Islet of Langerhans.

Figure 6, illustrates the histological analysis of liver tissues stained with hematoxylin and eosin, revealing significant differences among the experimental groups. The control group (A) exhibits normal hepatic architecture, with intact central veins (CV), sinusoidal spaces (S), and well-organized hepatocytes (HC). The diabetic group (B) shows severe structural damage, including disrupted sinusoidal spaces, swollen hepatocytes, and inflammation. Treatment with Ag NPs (C) and M. oleifera leaf extract (D) demonstrates partial restoration of liver architecture, with reduced hepatocyte swelling and improved sinusoidal spaces. Treatment with the combination (E) displays a near-normal hepatic structure, highlighting a synergistic effect in restoring liver histology.

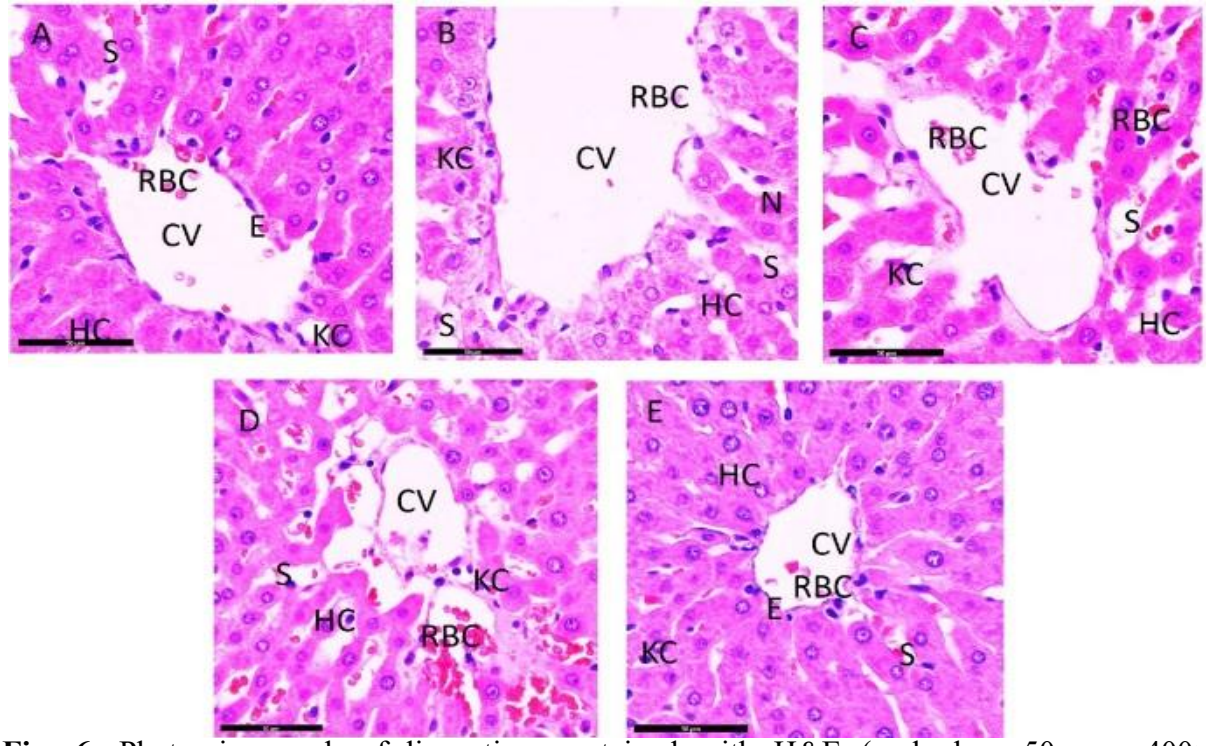


Fig. 6. Photomicrograph of liver tissues stained with H&E (scale bar: 50 µm, 400× magnification). (A) Control group, (B) Diabetic group (received STZ), (C) Diabetic + Ag NPs, (D) Diabetic + *M. oleifera* leaf extract, (E) Diabetic + Ag NPs + *M. oleifer*a leaf extract. CV: Central vein; RBC: Red blood cell; HC: Hepatic cell; KC: Kupffer cell; S: Sinusoid; Black arrow: Binucleated hepatocyte; E: Endothelial cell.

DISCUSSION

This study elucidates the synergistic therapeutic potential effects of green silver nanoparticles (Ag NPs), *M. oleifera* leaf extract, and their combination in alleviating hepatic dysfunction and oxidative stress in STZ-induced diabetic rats. The finding highlights their combined efficacy in reducing hyperglycemia, restoring liver function biomarkers, and ameliorating histopathological changes in hepatic tissue. Diabetic liver injury is primarily driven by hyperglycemia-induced oxidative damage, leading to increased ALT, AST, and MDA levels and impaired antioxidant defenses such as reduced GSH, SOD, and CAT (Zeng *et al.,* 2021; Zhang *et al.,* 2022).

In this study, the administration of Ag NPs and *M. oleifera* leaf extract significantly improved glucose metabolism, indicated by reduced serum glucose and HbA1c levels and elevated insulin levels. Notably, the combined treatment exhibited superior efficacy compared to individual treatments, demonstrating a synergistic effect in mitigating hyperglycemia (Kalakotla *et al.,* 2022; Zeng *et al.,* 2021).

Furthermore, the restoration of liver function biomarkers, including albumin, total protein, and total bilirubin levels, suggests a pronounced hepatoprotective effect, particularly in the combined treatment group (Zeng *et al.,* 2021). Histopathological analysis further validated these biochemical improvements, revealing that the combined treatment more effectively preserved hepatic and pancreatic architecture than individual therapies. The therapeutic benefits are likely attributed to the bioactive compounds in M. oleifera, such as flavonoids and phenolics, which possess potent antioxidant and anti-inflammatory properties (Arora & Arora, 2021; Chiș *et al.,* 2023). Additionally, the crystalline structure of Ag NPs, as confirmed by TEM, contributes to their efficacy by enhancing antioxidant defense mechanisms and reducing lipid peroxidation (Hassan Afandy *et al.,* 2023; Wypij *et al.,* 2021). While these findings are interesting, several limitations should be addressed. The study duration was relatively short, and further long-term studies are necessary to determine the safety, optimal dosage, and molecular mechanisms underlying these effects.

Conclusion

The current study highlights the combined treatment of Ag NPs and *M. oleifera* leaf extract as a promising therapeutic strategy for diabetic liver complications. By addressing oxidative stress, hyperglycemia, and inflammation, this novel approach demonstrates superior efficacy in restoring liver function, reducing oxidative damage, and improving pancreatic and hepatic histology compared to individual treatments. These findings provide a strong foundation for further research into the molecular mechanisms and clinical applications of Ag NPs and *M. oleifera* in diabetes management.

Declarations:

Ethical Approval: The experiments were processed using the animal ethical rules of King Abdulaziz University's Animal Care and Use Committee (ACUC). Furthermore, all tests adhered to the Arrive standards and EU Directive 2010/63/EU regarding animal research.

Competing interests: The authors declare that there is no conflict of interest.

Author's Contributions: Mohammed F. Al Khuzaee and Muhamed A. El Nobey contributed equally to this work and share the first authorship. Muhamed A El Nobey wrote the manuscript, and Salim M El Hamidy revised it**.**

Funding: This study received no funding from any sources.

Availability of Data and Materials: All data sets are available in the manuscript and supplementary file.

Acknowledgments: Not applicable

REFERENCES

Alfieri, C. M., Molinari, P., Cinque, F., Vettoretti, S., Cespiati, A., Bignamini, D., Nardelli, L., Fracanzani, A. L., Castellano, G., & Lombardi, R. (2024). What Not to Overlook in the Management of Patients with Type 2 Diabetes Mellitus: The Nephrological and Hepatological Perspectives. *International Journal of Molecular Sciences*, *25(14), 7728.*

Arora, S., & Arora, S. (2021). Nutritional significance and therapeutic potential of Moringa oleifera: The wonder plant. *Journal of food biochemistry*, e13933 .

Bae, C ,.Lee, Y., & Ahn, T. (2023). Therapeutic treatments for diabetes mellitus-induced liver injury by regulating oxidative stress and inflammation. *Applied Microscopy*, *53(1), 4.*

Bannon, Pierre .1982. Effect of pH on the elimination of the labile fraction of glycosylated hemoglobin. *Clinical chemistry,* 28(10):2183-2183.

Bowers, G. N., Jr, & McComb, R. B. (1966). A continuous spectrophotometric method for measuring the activity of serum alkaline phosphatase. *Clinical chemistry,* 12(2), 70–89.

Busher, J.T. (1990) Serum Albumin and Globulin. In: Walker, H.K., Hall, W.D., Hurst, J.W., Eds., Clinical Methods: The History, Physical, and Laboratory Examinations, Chapter 101, 3rd Edition, Butterworths, Boston. https://www.ncbi.nlm.nih .gov/books/ NBK204/.

Chatzimitakos, T. G., Athanasiadis, V., Kotsou, K., Mantiniotou, M., Kalompatsios, D., Makrygiannis, I., Bozinou, E., & Lalas, S. I. (2024). Optimization of Pressurized Liquid Extraction (PLE) Parameters for Extraction of Bioactive Compounds from Moringa oleifera Leaves and Bioactivity Assessment. *International Journal of Molecular Sciences* ,25(9): 4628.doi: 10.3390/ijms25094628.

Chiș, A., Noubissi, P. A., Pop, O. L., Mureșan, C. I., Fokam Tagne, M. A., Kamgang, R., Fodor, A., Sitar-Tăuț, A. V., Cozma, A., Orășan, O. H., Hegheș, S. C., Vulturar, R., & Suharoschi, R. (2023). Bioactive Compounds in Moringa oleifera: Mechanisms of Action, Focus on Their Anti-Inflammatory Properties. *Plants*, *13(1), 20.* DOI:10.3390/plants13010020

Chung, W., Promrat, K., & Wands, J. (2020). Clinical implications, diagnosis, and management of diabetes in patients with chronic liver diseases. *World Journal of Hepatology*, *12*, 533 .557 .

Doumas, B. T., Kwok-Cheung, P. P., Perry, B. W., Jendrzejczak, B., McComb, R. B., Schaffer, R., & Hause, L. L. (1985). Candidate reference method for determination of total bilirubin in serum: development and validation. *Clinical chemistry,* 31(11), 1779–1789.

Draper, H. H., & Hadley, M. (1990). Malondialdehyde determination as index of lipid peroxidation. *Methods in enzymology,* 186, 421–431. https://doi.org/10.1016/0076-6879(90)86135-i.

Hassan Afandy, H., Sabir, D. K., & Aziz, S. B. (2023). Antibacterial Activity of the Green Synthesized Plasmonic Silver Nanoparticles with Crystalline Structure against Gram-Positive and Gram-Negative Bacteria. *Nanomaterials*, 13(8):1327.doi: 10.3390/ nano13081327.

Kalakotla, S., P., G., Banu, A. N., & Shaik, S. (2022). Development of Plant-Mediated Silver Nanoparticles & Their Pharmacological Evaluation .DOI:10.21203/rs.3.rs-1224222/v1

Middleton, J., & Griffiths, W. (1957). Rapid colorimetric micro-method for estimating glucose in blood and CSF using glucose oxidase. *British medical journal*, *2*(5060), 1525–1527.

Misra, H. P., & Fridovich, I. (1972). The role of superoxide anion in the autoxidation of epinephrine and a simple assay for superoxide dismutase. Journal of Biological chemistry, 247(10), 3170-3175.

Mohamed, J., Nafizah, A. H .N., Zariyantey, A. H., & Budin, S. B. (2016). Mechanisms of Diabetes-Induced Liver Damage: The role of oxidative stress and inflammation. *Sultan Qaboos University medical journal*, *16 2*, e132-141 .

Moron, M. S., Depierre, J. W., & Mannervik, B. (1979). Levels of glutathione, glutathione reductase and glutathione S-transferase activities in rat lung and liver. *Biochimica et biophysica acta (BBA)-general subjects*, *582*(1), 67-78.

Muhammad, S., Ali, A., Shah, J., Hamza, M., Kashif, M., Ajat Khel, B. K., & Iqbal, A. (2023). Using Moringa oleifera stem extract for green synthesis, characterization, and anti-inflammatory activity of silver oxide nanoparticles. *Natural and Applied Sciences International Journal (NASIJ) .4*(1), 80-97.

Pareek, A., Pant, M ,.Gupta, M. M., Kashania, P., Ratan, Y., Jain, V., Pareek, A., & Chuturgoon, A. (2023). Moringa oleifera: An Updated Comprehensive Review of Its Pharmacological Activities, Ethnomedicinal, Phytopharmaceutical Formulation, Clinical, Phytochemical, and Toxicological Aspects. *International Journal of Molecular Sciences*, 24(3):2098.doi: 10.3390/ijms24032098.

Peters T., Jr (1968). Proposals for standardization of total protein assays. *Clinical chemistry,* 14(12), 1147–1159.

Reddy Gangadi, R. (2024). Emerging Nanobiotechnology Solutions for Infectious Disease Diagnosis and Treatment. *International Journal of Science and Research (IJSR)* .

Reitman, S. and Frankel, S. (1957) A Colorimetric Method for the Determination of Serum Glutamic Oxalacetic and Glutamic Pyruvic Transaminases. *American Journal of Clinical Pathology,* 28, 56-63.

Rohm, T., Meier ,D. T., Olefsky, J. M., & Donath, M. Y. (2022). Inflammation in obesity, diabetes, and related disorders. *Immunity*, *55 1*, 31-55 .

Srivastava, A., Trivedi, S., Agarwal, N., Pandey, P. K., & Solanki, V. S. (2024). A review on multifarious medicinal potential of green synthesized silver nanoparticles. *World Journal of Advanced Research and Reviews .23*(1), 2384-2402.

Stevens, J. F., Tsang, W., & Newall, R. G. (1983). Measurement of the enzymes lactate dehydrogenase and creatine kinase using reflectance spectroscopy and reagent strips. *Journal of Clinical Pathology,* 36(12), 1371–1376. https://doi.org/10. 1136/jcp. 36.12.1371.

Villagrán, Z., Anaya-Esparza, L. M., Velázquez-Carriles, C. A., Silva-Jara, J. M., Ruvalcaba-Gómez, J. M., Aurora-Vigo, E. F., Rodríguez-Lafitte, E., Rodríguez-Barajas, N., Balderas-León, I., & Martínez-Esquivias, F. (202 .(4Plant-Based Extracts as Reducing, Capping, and Stabilizing Agents for the Green Synthesis of Inorganic Nanoparticles. *Resources*.*13*(6), 70; **https://doi.org/10.3390/resources13060070**

Virk, P., Awad, M. A., Saleh Abdu-llah Alsaif, S., Hendi, A. A., Elobeid, M. A., Ortashi, K. M. O., Qindeel, R., El-khadragy, M., Yehia, H. M., Ferkry Serag EL-DIN, M., & Ali Salama, H. (2023). Green synthesis of Moringa oleifera leaf nanoparticles and an assessment of their therapeutic potential. *Journal of King Saud University - Science* . *35*(3), 102576.

Weinberg Sibony, R., Segev, O., Dor, S., & Raz, I. (2024). Overview of oxidative stress and inflammation in diabetes. *Journal of Diabetes*, *16(10), e70014.*

Wypij, M., Jędrzejewski, T., Trzcińska-Wencel, J., Ostrowski, M., Rai, M., & Golińska, P. (2021). Green Synthesized Silver Nanoparticles: Antibacterial and Anticancer Activities, Biocompatibility, and Analyses of Surface-Attached Proteins. *Frontiers in Microbiology*, *12, 632505.* https://doi.org/10.3389/fmicb.2021.632505

Yameny, A. A. (2024). Diabetes Mellitus Overview 2024. *Journal of Bioscience and Applied Research*, *10(3), 641-645*.

Zeng, F., Luo, J., Han, H ,.Xie, W., Wang, L., Han, R., Chen, H., Cai, Y., Huang, H., & Xia, Z. (2021). Allopurinol ameliorates liver injury in type 1 diabetic rats through activating Nrf2. *International Journal of Immunopathology and Pharmacology*, *35, 20587384211031417.*

Zhang, X. (2024). Modified Buyang Huanwu Decoction alleviates diabetic liver injury via inhibiting oxidative stress in db/db mice. *American Journal of Translational Research* .

Zhang, Y., Tan, X., Cao, Y.-y., An, X., Chen, J., & Yang, L. (2022). Punicalagin Protects against Diabetic Liver Injury by Upregulating Mitophagy and Antioxidant Enzyme Activities. *Nutrients*, 14(14):2782.doi: 10.3390/nu14142782.

Zulfiqar, Z., Khan, R. R. M., Summer, M., Saeed, Z., Pervaiz, M., Rasheed, S., ... & Ishaq, S. (2024). Plant-mediated green synthesis of silver nanoparticles: synthesis, characterization, biological applications, and toxicological considerations: a review. *Biocatalysis and Agricultural Biotechnology, 57*, 103121.