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Antidiabetic Effect of Glycyrrhizin glabra extract and Glycyrrhiza glabra Silver Nanoparticle in Female Rats

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KEY WORDS : Diabetes mellitus; Glycyrrhiza glabra; Silver nanoparticle; Alloxan; Nicotinamide

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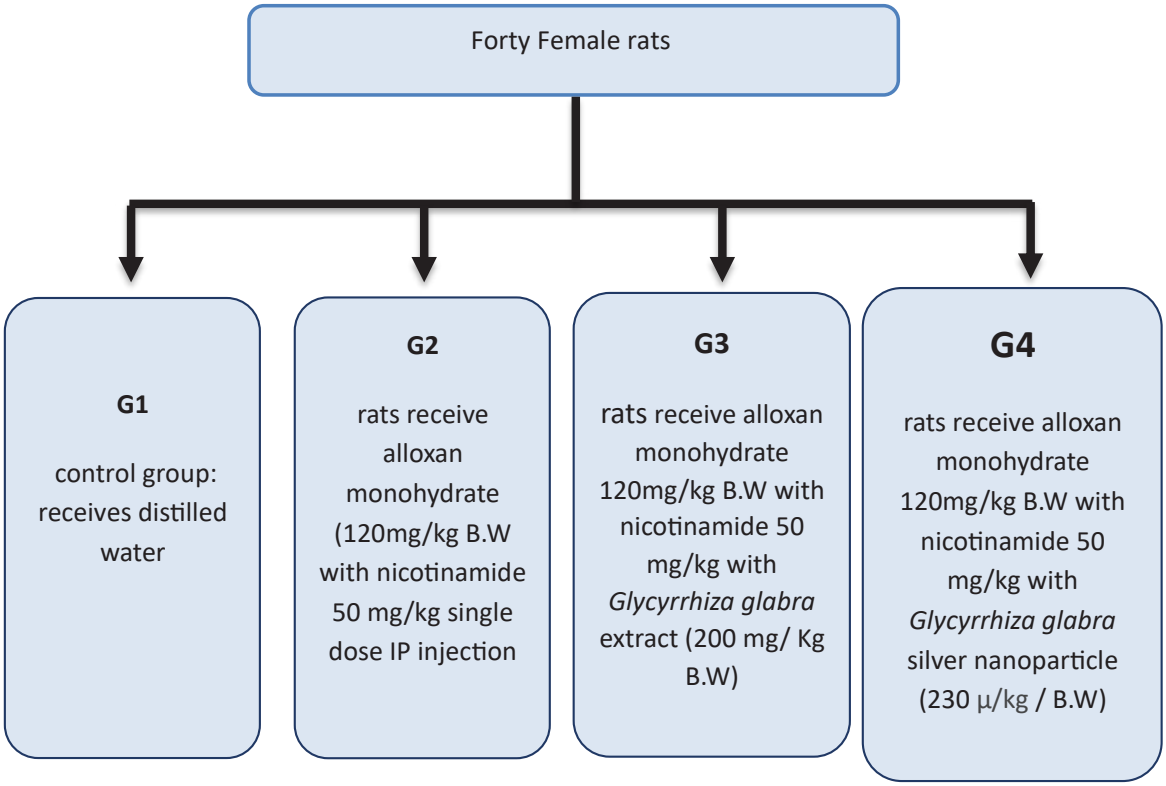
ABSTRACT

Objective: To study the antidiabetic effect of *Glycyrrhiza glabra* ethanolic extract and *Glycyrrhiza glabra* silver nanoparticle against induced diabetes mellitus (DM) by alloxan and nicotinamide in adult female Rats. **Methods:** The current study was performed over 36 days in which the *G. glabra* extract (200 mg/ Kg) and *G. glabra* extract loaded on Silver nanoparticle (230 µg/ Kg) were given to alloxan and nicotinamide (120mg/kg, 50 mg/kg, respectively, single dose, IP) induced diabetic rats (10 rats per group). The antidiabetic effect has been evaluated biochemically**. Results:** The results of induction of DM for 36 days showed a significant increase (P<0.05) in serum Glucose concentration with a significant decrease (P<0.05) in serum insulin concentration in DM induced (G2) group when compared with the control group (G1). and showed a significant decrease (P<0.05) in the serum glucose concentration and a significant increase (P<0.05) of serum insulin concentration in (G3) group (received *G. glabra* extract) and (G4) group (*G.glabra* loaded on silver nanoparticle) in comparison with G2. **Conclusion:** This study concluded that *Glycyrriza glabra* extract and *Glycyrrhiza glabra* Silver nanoparticle have a significant antidiabetic effect induced by alloxan and nicotinamide.

GRAPHICAL ABSTRACT



Introduction

Diabetes is considered one of the top 10 causes of death worldwide, with approximately 1.6 million people dying from it annually[1]. Diabetes mellitus (DM) is a metabolic disorder caused by reduced insulin activity and/or secretion. As the disease progresses, complications like nephropathy, retinopathy, and cardiovascular issues can arise. There are two main subtypes of DM: type I and type II. Type I DM is typically treated with insulin replacement therapy, while type II DM is managed using oral hypoglycemic drugs. The drug therapy for type II DM includes insulin secretagogues, biguanides, insulin sensitizers, alpha-glucosidase inhibitors, incretin mimetics, amylin antagonists, and sodium-glucose co-transporter-2 (SGLT2) inhibitors[2].

A study found that individuals between the ages of 40 and 49 with type 2 diabetes could lose an average of ten years of life. People with diabetes are 2-3 times more likely to develop coronary heart disease compared to those without diabetes[3].

Medicinal plants can be prescribed for long-term use in diabetes because the risk of serious side effects of natural preparations is significantly lower than with the use of medicines[4-7]. Licorice, a plant with the scientific name Glycyrrhiza glabra L., has been used for medicinal purposes since ancient times. Its main component is glycyrrhizin, which gives it a sweet taste and is used as a sweetener and flavouring agent in various foods8. Licorice has been a key ingredient in traditional Chinese medicine for over a thousand years. It contains numerous active chemical compounds, including over 20 triterpenoids and 300 flavonoids, which have been found to possess various pharmacological properties[9]. These properties include anti-inflammatory, antitumor, antimicrobial, antiviral, and immunoregulatory effects. Licorice has also shown potential in protecting and treating the cardiovascular, respiratory,

endocrine, digestive, and nervous systems[10]. Additionally, licorice and its bioactive compounds exhibit several beneficial effects, such as anti-inflammatory, antioxidant, anti-atherogenic, and anti-platelet actions, making it a promising agent for preventing atherosclerosis. Experimental and clinical studies have demonstrated licorice's ability to reduce cholesterol accumulation in macrophages and improve traditional cardiovascular risk factors[11]. Some studies have specifically focused on the beneficial effects of licorice extract and silver nanoparticles derived from Glycyrrhiza glabra in female rats.

Some herbal drugs are drawbacked by their limitations in the pharmacokinetics properties of these medications[12,13]. Glycyrrhizin are characterized by low bioavailability due to weak absorption, therefore, glycyrrhizin loaded nanoparticle provides improved pharmacokinetic properties[14-16]. The physicochemical properties of nanoparticles play a crucial role in enhancing the bioavailability of therapeutic agents, whether administered systemically or locally. Research studies have shown that nanoparticles can significantly improve the delivery and effectiveness of therapeutic drugs in the body[17,18]. However, it is important to note that these properties can also impact cellular uptake, biological distribution, penetration into biological barriers, and ultimately the therapeutic effects[19,20]. Consequently, there is a growing need for the development of silver nanoparticles (AgNPs) with controlled structures that are uniform in size, morphology, and functionality. These controlled structures are essential for a wide range of biomedical applications[21–25].

AgNPs typically ranging from 1 to 100 nanometers in size, possess a large surface area to volume ratio, high stability, their small size allows for easy penetration into cells and tissues, making them ideal for targeted drug delivery[24]. Additionally, silver nanoparticles have the ability to bind to a variety of drugs, enhancing their stability and bioavailability[25]. Furthermore, AgNPs have demonstrated potential in enhancing the efficacy of traditional chemotherapy drugs by promoting their selective accumulation in tumor tissues while minimizing their toxicity to healthy cells[26]. The surface of silver nanoparticles can also be modified to improve their biocompatibility, reduce their toxicity, and control their release kinetics[24]. These properties make silver nanoparticles a promising tool in the development of novel drug delivery systems that can improve therapeutic outcomes and reduce side effects[27-30].

Silver nanoparticles have gained significant attention in the field of diabetes research due to their unique properties and potential therapeutic applications[31]. One of the key reasons why silver nanoparticles are being explored in the context of diabetes is their antimicrobial activity[32,33]. Diabetes patients are often prone to infections, and silver nanoparticles have shown promising results in inhibiting the growth of various pathogens, including bacteria and fungi[33]. Additionally, silver nanoparticles possess excellent biocompatibility and are easily synthesized, making them an attractive option for biomedical applications. In the case of diabetes, silver nanoparticles have been studied for their potential in wound healing. Diabetes-related foot ulcers and chronic wounds can be challenging to treat, and silver nanoparticles have demonstrated the ability to accelerate wound closure, reduce inflammation, and prevent infection[32]. Moreover, silver nanoparticles have also been investigated for their potential to regulate glucose levels[31]. Researchers have explored the use of silver nanoparticles in developing glucose biosensors, which can accurately monitor blood glucose levels and aid in the management of diabetes[33]. These biosensors utilize the unique optical and electrical properties of silver nanoparticles to detect glucose molecules, providing a non-invasive and convenient method for diabetes monitoring[32]. Furthermore, silver nanoparticles have been studied for their antioxidant and anti-inflammatory properties, which are beneficial in mitigating oxidative stress and inflammation associated with diabetes. Overall, the utilization of silver nanoparticles in diabetes research holds great promise and could potentially revolutionize the management and treatment of this chronic condition[31-33].

One important aspect to consider when utilizing silver nanoparticles for biomedical purposes is their biocompatibility, or the ability to interact with liv-

ing organisms without causing adverse effects[34-36]. In this context, glycyrrhiza, a plant species commonly known as licorice, has been explored for its potential as a biocompatible material when combined with silver nanoparticles[11,32]. Glycyrrhiza possesses several bioactive compounds, such as flavonoids and glycyrrhizic acid, which have been shown to exhibit various beneficial properties, including anti-inflammatory, antioxidant, and antimicrobial effects[32,34-37]. These properties make glycyrrhiza an attractive candidate for enhancing the biocompatibility of silver nanoparticles. Studies have shown that incorporating glycyrrhiza into silver nanoparticle formulations can improve their biocompatibility by reducing cytotoxicity and promoting cell viability[34]. Furthermore, glycyrrhiza has demonstrated the ability to enhance the stability and dispersibility of silver nanoparticles, which are crucial factors for their successful application in biomedical settings[37,38].

The present study aimed to use glycyrrhizin-loaded silver nanoparticles to improve the hypoglycemic properties of glycyrrhizin in rodent diabetic models due to the biocompatibility of silver nanoparticles as being used by previous studies[19-24].

Materials and methods

Plant Materials and Extraction: Mature specimens of wild P. oleracea (roots and stems) were obtained from ⁉ Markets in Baghdad province. The plant was authenticated officially by the Ministry ⁉of Agriculture, State Board for Seed Certification and Testing, located in Abu Ghraib, ⁉Baghdad, under certification Iraq number 3273, dated 6th November 2022⁉.

Synthesis of AgNPs-loaded Glycyrriza glabra: Preparation of green synthesis silver nanoparticles by using Glycyrrhiza glabra ethanolic extracts26. Five ml of each extract was sprayed into 95 ml of 10 mM silver nitrate $AgNO_3$ solution (which prepared by dissolving 1.69 g $AgNO_3$ into 1 L deionized water) separately dropwise with a flow rate of 0.2 ml/min under ultrasonic conditions, with an ultrasonic power of 100 W and a frequency of 42 kHz. After sonication for 20 min, the solutions were stirred at 800 rpm at 25°C for 30 min, then kept in dark bottles at 25°C for 24 h. After 24 h the reaction mixture was purified by centrifugation for 10 min at 10000 rpm to get clear supernatant[26]. The final colloid samples were stored in dark bottles at 4°C. During 5 days the color of the solutions was changed from clear yellowish to dark greenish brown this color change indicates the formation of green synthesis silver nanoparticles. The ratio of extract and AgNO3 was optimised based on maximal yield through series of experiment to reach the best ratio giving maximum yield.[27,28]

Physicochemical Characterization

Visual Observation or Color Modification of Biosynthesized AgNPs: The initial stage in the increased manufacture (or fabrication) of biogenic AgNPs utilizing aqueous extract glycyrrhiza glabra (GG) with AgNO3 is to adjust (or change) the particle colour (GG AgNPs). Thus, the present research found that when the glycyrrhiza glabra extract was progressively added to the colourless silver nitrate solution, the colour of the combination changed to yellowish after 20 minutes. The colour then began to alter after an hour to brown then to dark brown after 48 hours, remaining constant at dark brown. The mixture was centrifuged, and the suspension was washed five times (Figure 1 and Figure 2).

Characterization of glycyrrhiza glabra silver nanoparticle:

UV-visible spectroscopy: UV–visible spectroscopy is a primary step in the confirmation of the synthesis of glycyrrhiza glabra silver nanoparticles. The absorbances of glycyrrhiza glabra and glycyrrhiza glabra silver nanoparticles were measured. The absorbance at wavelengths was calculated for glycyrrhiza glabra and glycyrrhiza glabra silver nanoparticles. In the glycyrrhiza glabra, the highest absorbance value was 2.223 at wavelengths 265nm. On the other hand, the optical density of glycyrrhiza glabra silver nanoparticle reached the highest absorbance value was 3.763 at wavelengths 231 and 270. The optical density wave of wavelength reached 3.068 at wavelengths

327, respectively. Indicate the formation of the nanomaterial and the successful loading of the GGE on chitosan nanoparticles (Figure 3, Table 1).

Fourier transformation infrared spectroscopy (FTIR): The FT-IR spectra of dried powdered GGE (reduced by Mentha Spicata were obtained at various peak ranges ranging from 500 to 4000 cm. FT-IR spectra with distinct peak bands at the band (3772.76) show an excited O-H stretch free hydroxyl bond, while the peak at (3408.22), shows an N-H stretch bond. Furthermore, the C-H stretch found in peak (2924.09 and 2866.22), C=N stretch found in peak (2384.02), 1726.29, 1618.28, 1369.46, 1232.51, 1064.71, 829.39, 482.20).

The presence of a wide and strong absorption band of glycyrrhiza glabra silver nanoparticle around (3404.36 $cm^{-1)}$ was attributed to the free hydroxyl stretching in alcohols and phenols. The stretching vibration of the hydroxyl group (O- H) of phenols and alcohols was assigned to the band at (3390.86cm^{-1}). The band between (2956.87 cm-1) and (2927.94 cm^{-1}) refers to C-H stretching in alkanes. The N-H bending in 1 amine is due to the peak at (1620.21 cm^{-1}). The peak at (1516.05cm^{-1}) is due to nitro compound N- O asymmetric stretch bending. The C-H rock bending in alkanes was responsible for the peak at (1357.89 cm^{-1}). The peak at (1062.07 cm^{-1}) was ascribed to aliphatic amine C-N wag stretch bending. The peak at (831.32cm^{-1} and 777.31cm^{-1}) on the other hand, is attributable to C-CL stretch bending in alkyl halides. Finally, the -C-C-H: C-H bending in alkynes is ascribed to the peaks at (615.29 cm^{-1}) and (547.78 cm^{-1}). These findings suggest the existence of functional groups in biosynthesized AgMSNPs.

Scanning electron microscopy (SEM) analysis: The morphology of glycyrrhiza glabra silver nanoparticles was investigated using a scanning electron microscope (SEM), glycyrrhiza glabra silver nanoparticles have a soft spherical surface appearance with a diameter range (of 13.40-69.76 nm), and have a relatively homogeneous morphology. The increase in dissolved ion concentration caused by liquid evaporation can reduce the electrostatic repulsive force, facilitating agglomeration (Figure 5).

The Scanning Electron Microscope analysis revealed the silver particles present in Nano size. These particles were (circular or spherical) form as some cluster aggregation with smooth edges were observed under SEM and below (100 nm) in range. The distribution particle size marginally varied with the variations in duration time. The particle numbers elevated with increasing duration because of the change in the reducing process.

Atomic Force Microscopy (AFM): The particle size ranged from (2 to 98) nm but the highest concentration of nanoparticles was between (65.3 to 70.7nm) in volume (Figure 6). The 3D image of glycyrrhiza glabra silver nanoparticle revealed a population of homogeneous particles with a regular surface shape (Figure 7). The images of AFM demonstrated smart interaction among glycyrrhiza glabra silver nanoparticles, leading to the formation of well-discrete aggregates.

Energy Dispersive X-ray (EDX): The elemental analysis of GGAgNPs via EDX which shows the presence of (O, Ag, Na, N, C and K) in concentration (5.6, 7.3, 2.9, 3.4, 4.6 and 3.6) % respectively. EDX examination reveals a significant signal at 1.7 keV and 2.2 keV due to the presence of carbon and oxygen respectively, confirming the presence of GGAgNPs.

X-ray diffraction (XRD): The main peak of 2θ value at (25.7°, 33.4, 37.6, 51.6,60 and 71.2) referred to the active compound in GGE while the value at (38.7, 52.3 and 78.1) referred to GGAgNPs intensity Level. The glycyrrhiza glabra peak, which shows its main peak of 2θ value at (22.8596°) and an intensity level at (736.5503 cont.), change indicates the difference in the crystal structure between these two materials where GGAgNPs was more crystalline than AgONPs, as well as the AgONPs diffraction peak, which was previously found at (38.7, 52.3 and 78.1), has shifted to a higher value (33.4°) in this study, which may be related to the interaction of AgO loaded with GGE from GGAgNPs. The XRD results suggested that the Particle size effects are generally responsible for the widening of peaks in crystalline XRD patterns. Wider peaks indicate lower particle sizes and reflect the influence of experimental circumstances on particle

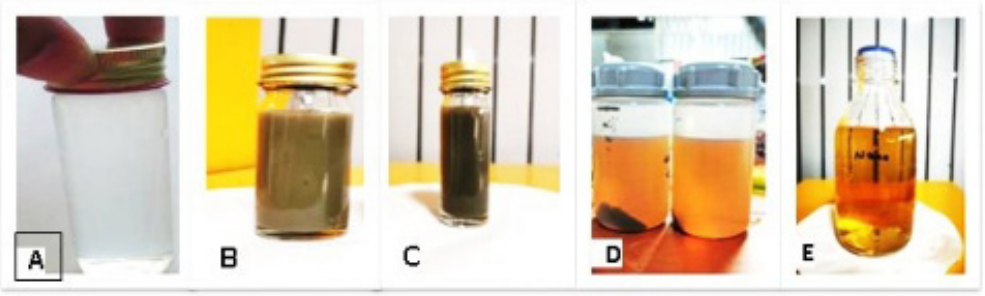


Figure 1. *Colour change during synthesis of Silver Nanoparticles: A-$AgNO_3$only, B-Change in colour from white to dark brown when put the glycyrrhiza glabra extract drop by drop. C-Change in colour after 2 hours from reaction time. D-Depositions of particles(centerfuge and wash).The GGAgNPs after tow days.*

Figure 2.*The dark brown colour of prepared biogenic Silver nanoparticles(AgNPs) after 48 hours of incubation.*

structures. Small crystals have a limited number of levels of reflection with low intensity, while large crystals have a large number of these levels with high intensity.

Animals: The study was conducted based on the reviewed and approved ethical approval received from the local committee in animal use and care, College of Veterinary Medicine, University of Baghdad (Approval Number 297 on 1st Feb 2022).

The groups of 40 rats were subdivided into:

G1: control group: received distilled water.

G2: diabetic group: this group receive alloxan monohydrate (120mg/kg of body weight) with nicotinamide 50 mg/kg single dose IP injection[29].

G3: diabetic rats-*Glycyrrhiza glabra* extract: diabetic rats received alloxan monohydrate 120mg/kg of body weight with nicotinamide 50 mg/kg with *Glycyrrhiza glabra* extract (200 mg/ Kg body weight)[30].

G4: diabetic rats-*Glycyrrhiza glabra* loaded on silver nanoparticle: diabetic rats receive alloxan monohydrate 120mg/kg of body weight with nicotinamide 50 mg/kg with *Glycyrrhiza glabra* loaded on silver nanoparticle (230 µg / Kg body weight)[30].

Blood samples were collected after 36 days of the treatment to measure the concentration of serum

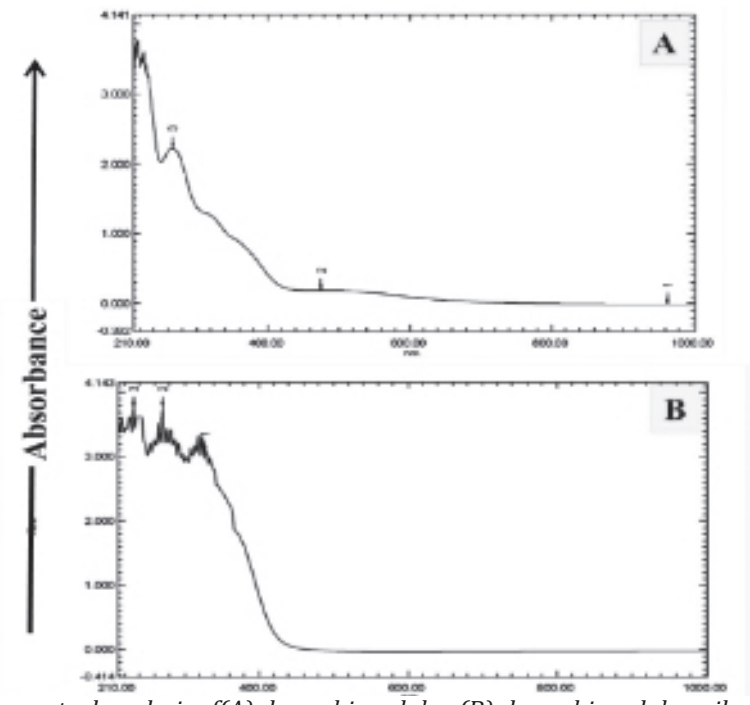


Figure 3. *UV-visible spectral analysis of(A)glycyrrhiza glabra(B)glycyrrhiza glabra silver nanoparticle.*

Table 2. UV-visible spectral analysis results of glycyrrhiza glabra and Glycyrrhiza glabra silver nanoparticle

Glycyrrhiza glabra			
N0.	P/V	Wavelength	Absorbance
1	↑	962	-0.013
2	↑	475	0.185
3	↑	265	2.223
Glycyrrhiza glabra silver nanoparticle			
N0.	P/V	Wavelength	Absorbance
1	↑	327	3.068
2	↑	270	3.763
3	↑	231	3.763

glucose concentration and insulin concentration

Results

Glucose concentration in the serum of female rats in (G2) exhibited a significant increase (P<0.05) with mean value (305.8±22.4) when compared with other treated groups and control group, whereas (G3 and G4 group) exhibited a significant decline (P<0.05) in glucose concentration with mean value (128.8±6.55, 115±5.37) respectively in comparison with (G2) with no significant variance (P<0.05) when compared with the control group, in addition, there was no statistical differences (P<0.05) in glucose concentration between (G3) and (G4 group). The insulin concentration showed a significant decrease

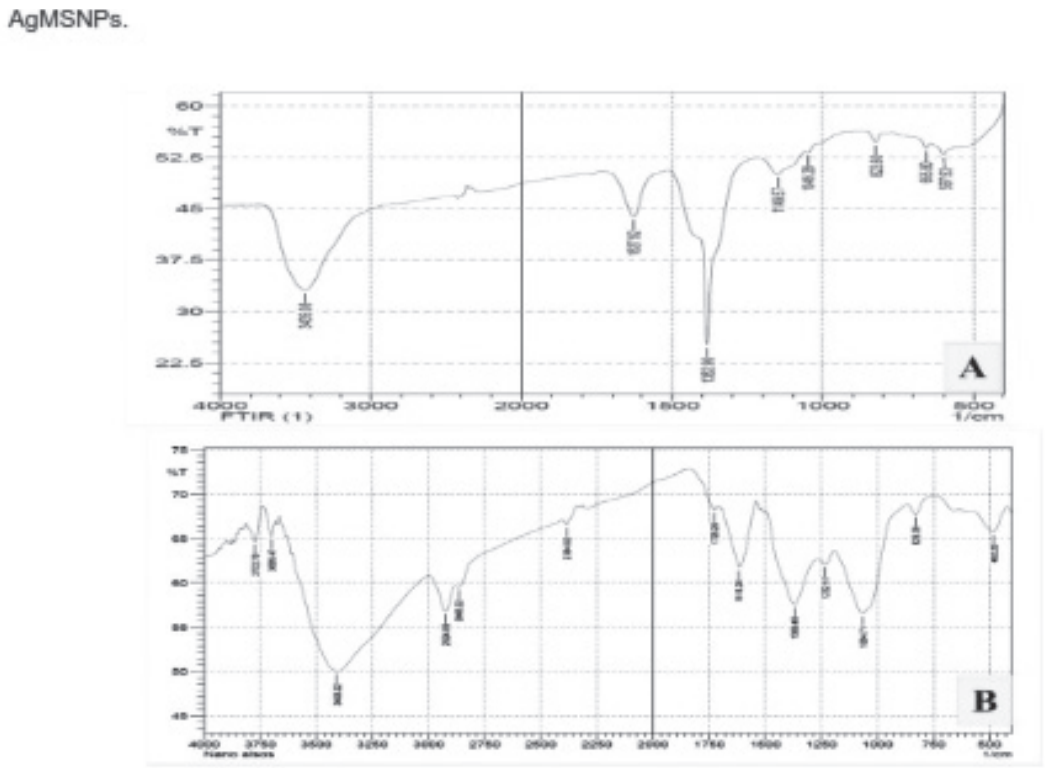


Figure 4. *Fourier transformation infrared spectroscopy(FTIR) of (A) glycyrrhiza glabra (B) Glycyrrhiza glabra silver nanoparticle*

(P<0.05) in (G2) with a mean value (3.16±0.41) as compared with other treated groups and control groups. whereas, there were no statistical differences (P<0.05) in insulin concentration between (G3) and (G4 group) with mean value (7.8±0.84, 7.52±0.88) respectively. While the insulin concentration of (the G3 and G4 groups) showed a significant increase (P<0.05) in mean value (7.8±0.84, 7.52±0.88) as compared with all other treated groups with no significant difference (P<0.05) as compared with the control group as in table (Figure 10).

Discussion

In the present study, *Glycyrrhiza* and its nanoparticle produced a significant anti-hyperglycemic effect in nicotinamide–alloxan–induced diabetic rats. Glycyrrhiza extracts its anti-hyperglycemic effect by increasing either insulin secretion by B-cells or the transportation of glucose from the bloodstream to peripheral tissue. In this study nicotinamide–alloxan dosing resulted in significantly increased fasting blood glucose level, which might be due to insulin secretion in diabetic rats[39] hyperglycemia in diabetes causing different complications. Therefore, the objective of all diabetic treatment is to maintain the blood glucose concentration within the normal Range. The anti-hyperglycemic effect of glycyrrhiza may inhibit sodium-glucose co-transporter-1 (S-Glut-1) mediated glucose transport across the intestine. Also, they enhance glucose-stimulated insulin secretion and induce m RNA levels of insulin receptor Substrate[40.]

In addition, as islet volume and several islet cells (including B-Cells) increase in glycyrrhiza-treated diabetic vats concerning diabetes control rats, the

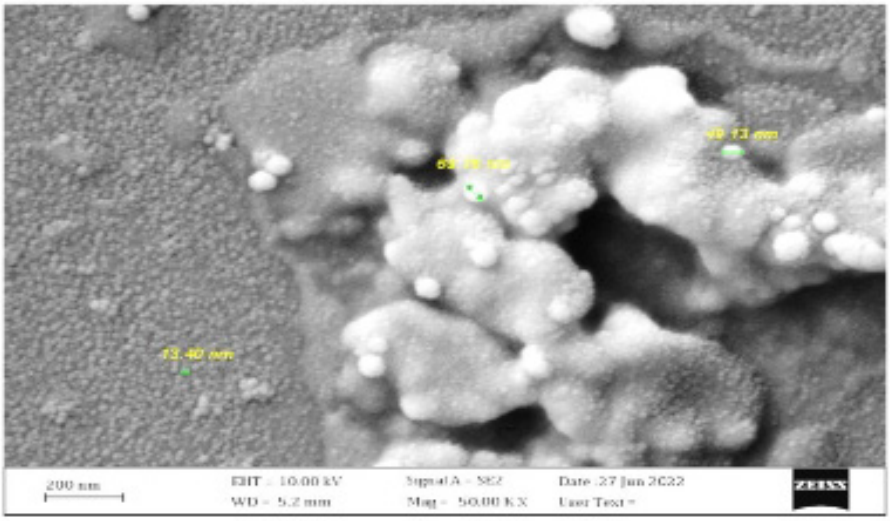


Figure 5. *Scanning electron microscopy(SEM) analysis for glycyrrhiza glabra silver nanoparticle*

possible mode of action of *Glycyrrhiza* is the regeneration or sensitization of Pancreatic B-cells that elevate serum insulin and thereby rectify hyperglycemic condition[41]. Insulin is a hormone produced by the pancreas that regulates blood glucose levels and stores glucose in insulin-sensitive organs such as fat, skeletal muscle, and the liver. Insulin resistance, which is the inability of these tissues to respond to insulin, is a key factor in type 2 diabetes. As a result, glucose uptake from the blood becomes difficult, leading to increased insulin production by the pancreas to lower glucose levels[42].

At a later stage, the pancreatic B-cells cannot compensate for the high demand for insulin, therefore, this eventually leads to B-cell dysfunction and defect in insulin, Secretion and hyperglycemia occur. Studies have shown that glycyrrhiza glabra could improve insulin sensitivity in diabetic rodents[43].

A study found that glycyrrhiza glabra reduced insulin resistance in diabetic rodents, indicating that it could improve insulin sensitivity. Otherwise, impaired fasting blood glucose is another indicator of insulin resistance and is characterized by another increase in hepatic insulin resistance[44]. In addition, high fasting serum insulin levels are another sign of insulin resistance45 and glycyrrhiza glabra, was found to decrease fasting serum insulin levels in diabetes rodents[46].

Insulin receptor (IR) is a transmembrane receptor which can be activated by insulin and insulin-like growth factor (IGF).IR and insulin receptor substrate (IRS) Play a vital role in regulating glucose homeostasis in the Pancreas and insulin-sensitive tissues[47]. IRs act as a Secondary messenger to transmit signals from insulin to downstream Intracellular pathways, thus enhancing insulin sensitivity and the transcription of insulin-related genes48. *Glycyrrhiza glabra* was found to increase IR mRNA expression and its phosphorylation to enhance insulin sensitivity[49].

Also, another study found that it enhanced insulin sensitivity through the phosphorylation of IRS-1. And IRS-2 in HFD induced diabetic in mice[50].

Moreover, protein tyrosine phosphatase 1B (PTP1B), the main enzyme involved in IR desensitization, regulates Insulin levels[51]. It is a negative regulator of the insulin signaling pathway, and its inhibitors have become an attractive strategy to treat T2 DM[52] Interestingly glycyrrhiza glabra was identified as Competitive PIP1B inhibitors[53]. Insulin-like growth

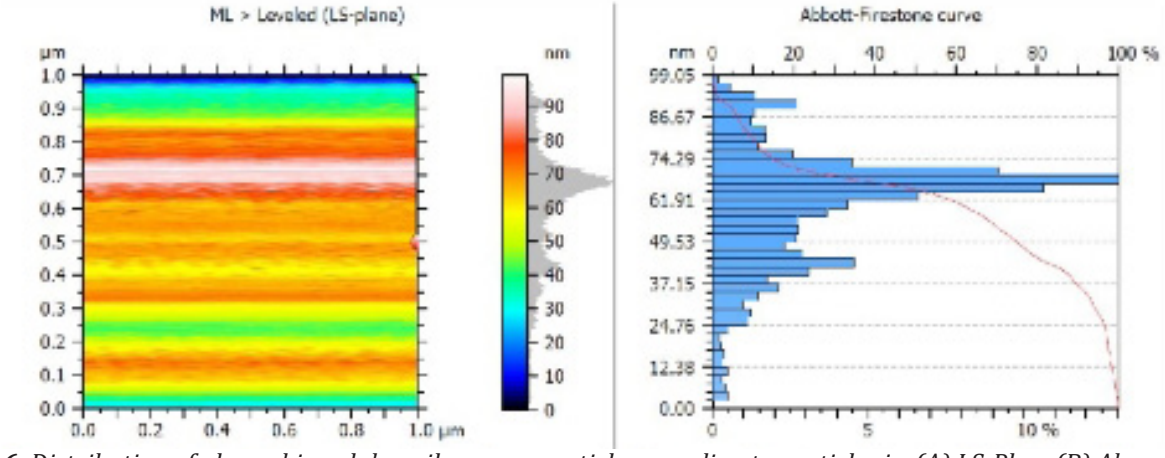


Figure 6. *Distribution of glycyrrhiza glabra silver nanoparticle according to particle size(A) LS-Plan. (B) Abbott-Firestone curve.*

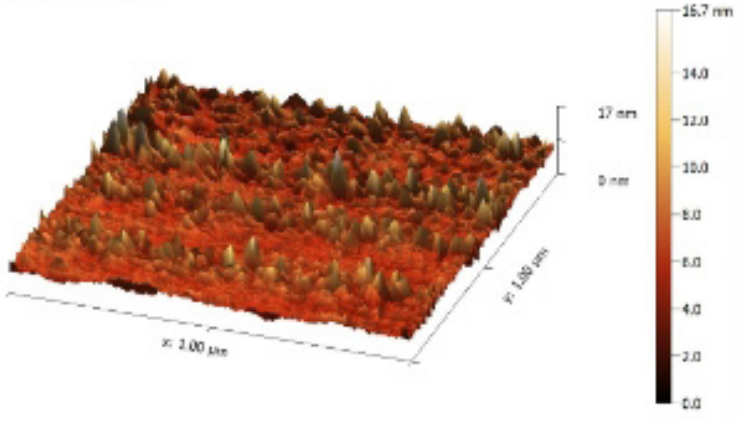


Figure 7. *A three-dimensional image of glycyrrhiza glabra silver nanoparticle revealed a population of homogeneous particles with a regular surface shape. Energy Disperesive X-ray (EDX): The elemental analysis of GGAgNPs via EDX which shows the presence of (O, Ag, Na, C and K) in concentration (5.6, 7.3, 2.9, 3.4, 4.6 and 3.6)% respectively*

factor - 1 (IGF -1) IS a growth hormone that can enhance glucose uptake and reduce hepatic glucose production, thereby improving insulin sensitivity[54].

Insulin and IGS-1 regulate many Signaling Pathways, including Ras/mitogen-activated Protein Kinase (MAPK), and phosphoinositide de 3-kinase (PI3K)/AKt pathway[55]. The activation of Ras is involved in the development of T2DM, and the activation of Ras genes improves insulin sensitivity[56].

In addition, the PI3K /Akt pathway was identified as one of the vital pathways that are associated with insulin resistance (87-D) *Glyayshiza glabra* was shown. To improve glucose uptake and reverse insulin resistance by targeting Ras proteins and activating PI3K /AKt Pathway[57]. Moreover, It was found that It improves insulin Sensitivity via suppressing advanced glycation end products (AGE) and receptors of AGE. (RAGS) axis which contributed to insu-

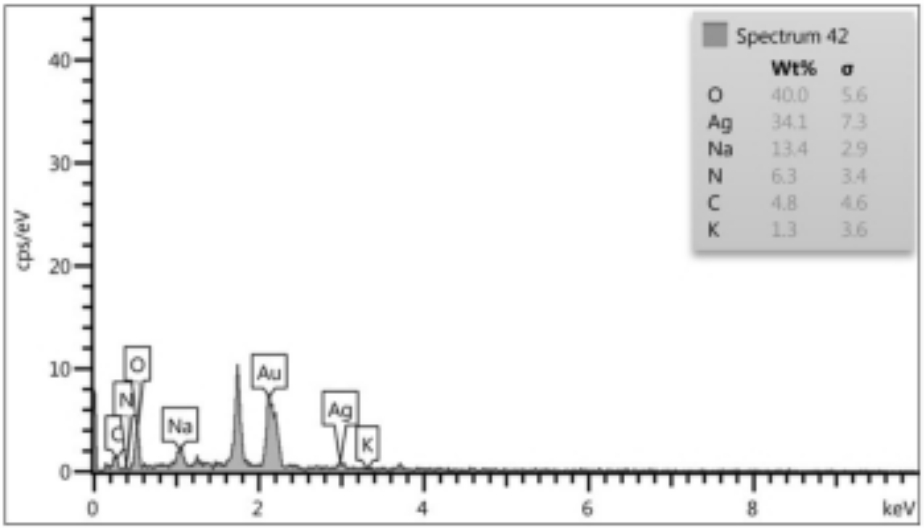


Figure 8. *Spectrum of Elemental Analysis of Glycyrrhiza glabra silver nanoparticle by EDX.*

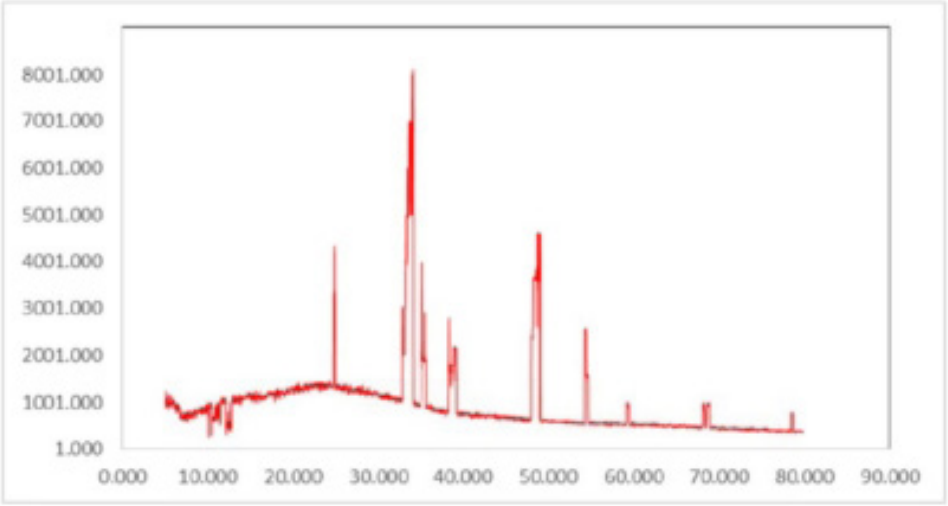


Figure 9. *The X-ray diffraction patterns of Glycyrrhiza glabra silver nanoparticle*

lin resistance58. The inhibition of pro-inflammatory mediators was suggested to be one of the strategies for preventing insulin resistance and the pathogenesis of diabetes[57]. Glyaynhiza glabra shown to reduce fasting plasma TNF-Alpha and IL-6 the pro-inflammatory mediator's levels to enhance insulin–responsive Pathways[59].

However, other studies found that *Glycyrhiza glabra* could reduce the M RNA expressions of the enzymes Phosphoenolpyruvate carboxykinase (PEPCK) and glucose 6-phosphatase to suppress gluconeogenesis and upregulate Pyravate dehydrogenase (PDase) and Glycogen Synthase Kinase 3B (GSK-3B) mRNA expressions to increase glycogen

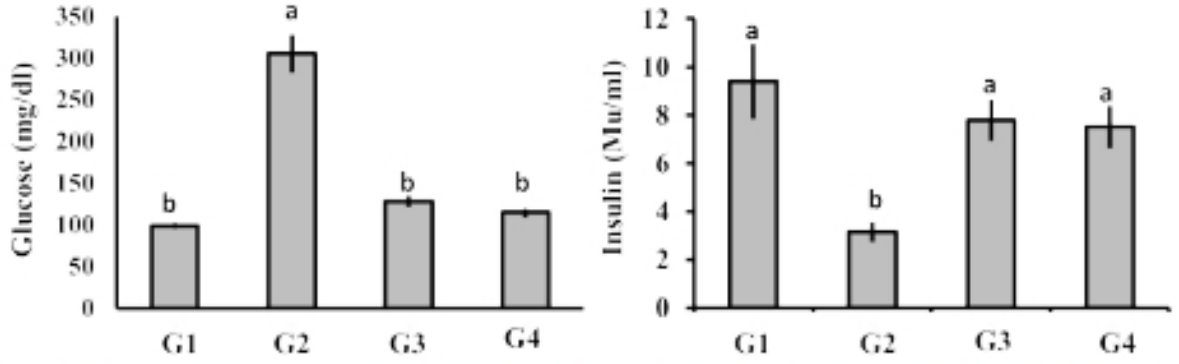


Figure10. *Antidiabetic effect of glycyrrhiza glabra extract and glycyrrhiza glabra extract loaded on silver nanoparticle on serum glucose level(mg/dl) and insulin concentration (μ/ml) in diabetic female rats. Different letters reveal significant differences between groups at level (P<o.o5). G1=Control negative group, G2=Alloxan with nicotinamide treated group. G3= Glycyrrhiza glabra ethanolic extract-treated group. G4=Glycyrrhiza glabra extract loaded on silver nanoparticle (n=5).*

synthesis in the liver[48]. Similarly, it reduced PEPck activities in the liver and kidney of normal rats[50] and alleviated the activities of PEPck and glucose 6-phosphatase in the liver and Kidney in rats with metabolic Syndrom[46], Furthermore, *Glgeyrhiza glabra* was found to promote GLUT4 expression by targeting Ras protein to regulate MAPK Pathway[57]. Due to the poor bio viability of *Glgeyrhiza glabra*, it has been formulated as nanoparticles. The loaded nanoparticles lowered fasting blood glucose levels in nicotinamide plus streptozotocin (STZ)-induced T2DM rats. Interestingly, the dosages used in *Glyegrrhiza glabra*. Loaded nanoparticles, were only a quarter of the dosages of Pure *Glycyrrhiza glabra* form[60,61]. Moreover, a combination of -*Glycenhiza glabra* -loaded nanoparticles and thymoquinone-loaded nanoparticles was applied, displaying better anti-diabetic activities than when administrated separately in nicotinamide and STZ-induced T2DM rats, including decreased blood glucose level[62]. Moreover, these nanoparticles were shown to accumulate in the liver after 2 h of treatment, reduce blood glucose levels and inhibit hepatic gluconeogenesis in rats[63].

Conclusion

The research concluded that both the extract and the nanoparticle derived from Glycyrrhiza glabra exhibit a notable antidiabetic effect, particularly when induced by alloxan and nicotinamide. This implies that these substances have the potential to be used as therapeutic agents in the management of diabetes mellitus. The antidiabetic effect observed in the study suggests that Glycyrrhiza glabra, commonly known as licorice, may hold promise as a natural remedy for individuals with diabetes. Further research and clinical trials are necessary to explore the mechanisms underlying this effect and to determine the optimal dosage and administration methods. Nonetheless, these findings shed light on the potential of Glycyrrhiza glabra extract and Glycyrrhiza glabra Silver nanoparticle as potential treatments for diabetes, offering hope for individuals seeking alternative and natural approaches to managing this chronic condition. □

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