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DESIGN AND EVALUATION OF ASTRAGALUS MEMBRANACEUS-LOADED NANOEMULSION FOR THE TREATMENT OF DIABETES

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ABSTRACT

Background: Diabetes mellitus is a long-term metabolic disease that is typified by oxidative stress, persistent hyperglycaemia, and related problems such as dyslipidemia and increasing pancreatic β-cell dysfunction. Conventional treatments are accessible, but prolonged use can result in negative side effects and insufficient metabolic control. Despite their promise for multitargeted therapy, herbal bioactives frequently have low bioavailability and poor water solubility. The current work aimed to enhance the antidiabetic activity and drug delivery efficiency by developing and refining an Astragalus membranaceus-loaded nanoemulsion (AM-NEs). **Materials and Methods:** The castor oil content, Smix ratio (Tween-80 and PEG 400), and sonication time were varied to optimise AM-NEs using a Box-Behnken design. In vitro drug release, morphology, entrapment efficiency, zeta potential, particle size, and polydispersity index were all evaluated for the optimised formulation. Over 21 days, body weight, fasting blood glucose, and lipid profile were measured in streptozotocin-induced diabetic rats to determine the antidiabetic efficacy. **Results and conclusion:** The optimised nanoemulsion showed entrapment efficiency of 83.72 ± 3.85%, zeta potential of −35 mV, PDI of 0.272 ± 0.029, and particle size of 162.76 ± 8.47 nm. According to Korsmeyer-Peppas kinetics, in vitro experiments revealed a cumulative drug release of 67.89% for 24 hours. Morphological investigation verified that the droplets were uniformly distributed and spherical. When compared to diabetic controls, the in vivo study showed a significant, dose-dependent improvement in body weight, a notable decrease in fasting blood glucose, and a normalisation of the lipid profile. In diabetic rats, the nanoemulsion significantly improved lipid metabolism and glycaemic control, suggesting increased pharmacological performance and bioavailability. These results imply that AM-NEs are a promising nanocarrier-based phytopharmaceutical strategy for managing diabetes, which calls for additional mechanistic and clinical research for translational use.

KEYWORD: Diabetes, Astragalus membranaceus, nanoemulsion, Hyperglycaemia.

INTRODUCTION

Diabetes mellitus (DM) is the most common endocrine disorder that affects more than 100 million people worldwide (6% population). It is caused by a deficiency or ineffective production of insulin by the pancreas, which results in an increase or decrease in the concentration of glucose in the blood. It is found to damage many of the body's systems, particularly blood vessels, eyes, kidneys, heart and nerves.[1] Diabetes mellitus has been classified into two types, i.e. insulin insulin-dependent diabetes mellitus (IDDM, Type I) and non-insulin-dependent diabetes mellitus (NIDDM, Type II). Type I diabetes is an autoimmune disease characterised by a local inflammatory reaction in and around islets that is followed by selective destruction of insulin-secreting cells, whereas Type II diabetes is characterised by peripheral insulin resistance and impaired insulin secretion.[2] The presence of DM shows increased risk of many complications such as cardiovascular diseases, peripheral vascular diseases, stroke, neuropathy, renal failure, retinopathy, blindness, amputations, etc.[3]

It is estimated that 366 million people had DM in 2011; by 2030 this would have risen to 552 million. The number of people with type 2 DM is increasing in every country with 80% of people with DM living in low- and middle-income countries. DM caused 4.6 million deaths in 2011.[4] It is estimated that 439 million people would have type 2 DM by the year 2030. The incidence of type 2 DM varies substantially from one geographical region to the other as a result of environmental and lifestyle risk factors.[5]

Astragalus membranaceus has been used as a valuable medicinal plant, especially in traditional Chinese medicine (TCM), also known as Huang Qi, in Chinese, Astragalus membranaceus is the dry root of Astragalus membranaceus.[6,7] A. membranaceus dried root (Astragali Radix) has been consumed over 2000 years due to health-promoting effects. Traditional Chinese medicine recommended A. membranaceus to treat various gastrointestinal disorders, including intestinal inflammation, chronic phlegmatic disorders, chronic diarrhea, and stomach ulcer, anti-inflammation, antitumor, antioxidant, anti-allergy, hypoglycaemic properties and treatment of various diseases.[8-10] The main reported chemical constituents of A. membranaceus were triterpenes, polysaccharides, flavonoids, and saponins.[11] The presence of compounds such as terpenoids and flavonoids that usually occur in free or glycosidic form is of relevant pharmacological interest due to the bioactivities attributed to these classes of compounds.

Nanoemulsion are defined as isotropic, thermodynamically stable transparent or translucent systems of oil and water which stabilize by surfactant with a droplet size usually in the range of 5 to 200 nm. Nanoemulsion having various advantages over the macroemulsion are as follows i.e., Nanoemulsions have a much higher surface area and free energy than macroemulsions that make them an effective transport system. This system does not show the problems of inherent creaming, flocculation, coalescence and sedimentation, which are commonly associated with macroemulsions. Nanoemulsions can be developed by spontaneous emulsification method to enhance the solubility and bioavailability of poorly water soluble drugs. These are non-toxic non-irritant hence can be easily applied to skin and mucous membranes. The use of nanoemulsion as delivery systems can improve the efficacy of a drug, allowing the total dose to be reduced and thus minimizing side effects.[12]

The main disadvantage of currently available drugs is that they have to be given throughout the life and produce side effects. Medicinal plants and their bioactive constituents can be used for treatment of DM throughout the world especially in countries where access to the conventional anti-DM agents is inadequate (3). Various experimental models are also available to screen antidiabetic activity of plant.[13]

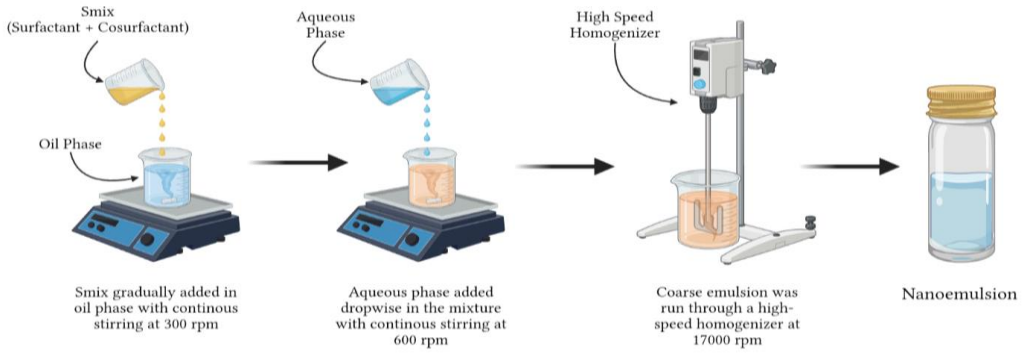
MATERIAL AND METHODS

Chemicals: Astragalus membranaceus (purity: 99%) was provided by sigma Aldrich, USA, acetic acid were obtained from Sigma-Aldrich (St. Louis, MO, USA). Polyoxyethylene sorbitan monolaurate (Tween-20) was purchased from Lobachem, (Mumbai, India), Acetonitrile was obtained from Lobachem, (Mumbai, India). Other reagents were a Analytical grade.

Animals: Adult Wister rats of either sex (200 – 250 gm) were used for the study, the rat were housed in the polypropylene cages under normal laboratory conditions (Temperature 25±1°C and relatively humidity 50±5°C) with 12 h light/dark cycle. Rats were fed with standard food pellets with water libitum. Experiment protocol was approved by IAEC.

Formulation of Nano emulsion

The Astragalus membranaceus nano emulsions were produced using a low-energy emulsification technique and aqueous titration method, The measured amount of the drug was incorporated into the oil and mixed until full solubilization occurred. Subsequently, the surfactant and co-surfactant mixtures in specified ratios were added into the appropriate tubes, followed by the gradual addition of the aqueous phase with continuous vortex mixing to produce a clear, transparent, and homogeneous Nano emulsion. The transparent or slightly brown NEs were assessed for droplet size, polydispersity index (PDI), and zeta potential utilizing a Zetasizer (Nano-ZS90, M).



2.5.1. Centrifugation test

All formulations were centrifuged at 5000 rpm for 20 minutes and were examined for indications of phase separation, creaming, or cracking. Homogeneous stable formulations have been selected for subsequent studies.

2.5.2. Heating-cooling cycle

NEs from the centrifugation test performed a heating–cooling cycle to examine the impact of temperature fluctuations on the formulation's stability. Test samples were kept for 48 hours, initially at 35 °C and subsequently at 4 °C, representing a single treatment cycle. Formulations that exhibited no indications of creaming, cracking, or phase separation, and demonstrated no significant alterations in droplet size, polydispersity index, and surface charge after three heating-cooling cycles, were advanced to the next stage of evaluation.

Optimization of AM Nano Emulsion

The Box–Behnken Design of Design-Expert is a superior tool for experimental design was utilized for optimization, particularly for analysing quadratic response surfaces and constructing second-order polynomial models. (14,15). BBD was utilized in this analysis to evaluate the impact of independent variables on specified dependent variables and to refine the formulation parameters of AM-NEs. (16,17) The chosen independent variables comprised the drug: Oil (X1), Smix (X2), and sonication time (X3), along with their corresponding low (1), middle (0), and high (1) values established in previous trials. Given the limitations on Nano emulsion formulations, particle size (Y1) and entrapment efficiency (Y2) were designated as dependent variables. The software Stat Ease Design Expert version 12.0.5.0 produced a design matrix consisting of 17 experimental runs. Table 1 delineates the variables and their corresponding levels employed in the factorial design.[18,19]

Factor	Low (–1)	Medium (0)	High (+1)
X_1: Black Seed Oil Concentration (% w/v)	2	4	8
X_2: Surfactant–Co-Surfactant (smix) %	5	10	20
X_3: Sonication time (min)	5	15	20
Particle size	Minimum		
Entrapment Efficiency	Maximum		

Characterization: (Article 3)

2.3.2. Particle size, zeta potential and (Article 23 NLCs)

The particle size (PS), polydispersity index (PDI), and zeta potential (ZP) of optimized AM-NEs were determined using the differential light scattering (DLS) technique on a particle size analyser (Beckman Coulter, Delsa Nano C, CA) at 25±1°C. Optimized AM – Nes was diluted with purified water to achieve a suitable concentration for this analysis. (20) All measurements were taken in triplate, and the results are expressed in means.

2.4. Entrapment efficiency (EE) and drug loading (DL)

Concentration of unentrapped free drug was used to measure entrapment efficiency (EE%). Drug-loaded sample was taken in a microcentrifuge tube and centrifuged at 50,000 rpm at 25°C for 2 hours, using Sorval mX 150 microcentrifuge (Thermo scientific, USA) and analyzed by HPLC. It was measured by calculating the amounts of non encapsulated NFT in aqueous surfactant solution, against the total amount of drug added to the formulation using following formula.[21]

In Vitro drug release: In vitro release studies were performed using modified Franz diffusion cell. Dialysis membrane having pore size 2.4 nm. The membrane was soaked in double distilled water for 12 h before mounting in a modified Franz diffusion cell. AM - NEs dispersion (1 mL) was placed in the donor compartment, and the receptor compartment was filled with 1% SLS in phosphate buffer, pH 7.4 (12 mL). During the experiments, the solution in receptor side was maintained at 37 ± 0.5°C and stirred at 800 rpm with Teflon-coated magnetic stirring bars. At fixed time intervals, 100 µL of the sample was withdrawn from receiver compartment through side tube and analysed by HPLC. Data obtained from in vitro release studies were fitted to various kinetic equations.[22,23] to find out the mechanism of drug release. The kinetic models used were zero-order equation, first-order equation, Higuchi release, and Weibull equation.

2.3.1. MORPHOLOGY

The microstructure of AM-NEs was observed using transmission electron microscope (TEM, JEM-1200EX, JEOL, Tokyo, Japan). Initially, samples diluted with double-distilled water were deposited on a film-coated copper grid, following by stained with 1% aqueous solution of phosphotungstic acid, ultimately the superfluous phosphotungstic acid on the samples was wiped off by filer paper and the sample was allowed to dry before examined under the TEM.

Evaluation of antidiabetic activity

4.2.13. Assessment of antidiabetic activity of the Astragalus membranaceus

For the assessment of the antidiabetic activity of *Astragalus membranaceus*, blood samples were collected from the tail vein of overnight-fasted rats on the 0th day

(prior to the initiation of treatment) and subsequently on the 3rd, 10th, 17th, 24th, and 30th days of the experimental period. Blood glucose levels were determined immediately using an Accu-Chek Active glucometer. The body weight of each rat was measured gravimetrically on the 0th and 30th days to monitor changes during the study. In addition, lipid profile parameters, including total cholesterol, triglycerides (TG), high-density lipoprotein (HDL), and low-density lipoprotein (LDL), were estimated for all four experimental groups to evaluate the extract's effect on dyslipidaemia associated with diabetes.[24] The detail of the biochemical parameter are given below.

Fasting Blood Glucose

Blood samples were obtained from the tail vein of overnight-fasted animals on Day 0, Day 3, Day 7, Day 14, and Day 21. Blood glucose levels were estimated immediately using an Accu-Check Active® glucometer. All measurements were carried out in triplicate to minimize random error, and the mean value was used for analysis.

4.2.13.1. Body Weight Measurement

Body weights were recorded on Day 0 and Day 21 using a digital electronic balance with a precision of ±0.1 g. The gravimetric method was followed to ensure accuracy, and the percentage change in body weight over the study period was calculated for each animal.

4.2.13.3. Lipid profile

Blood was collected on the 28th day from retro orbital plexus of overnight fasting rats and different lipid parameters in blood like Triglycerides (TGs), Total Cholesterol (TC), low density lipoprotein (LDL), and high-density lipoprotein (HDL) was estimated using commercial kits procured from Bayer Diagnostics & Siemens Healthcare, India with the aid of a clinical chemistry analyser Chem-7 (Erba, Manheim, Germany).

Statistical Analysis

The data were presented as the mean ± standard deviation (SD) from a minimum of three independent repetitions. The analyzed groups were considered substantially distinct when $p < 0.05$. A one-way analysis of variance (ANOVA) was conducted utilizing Design-Expert version 12.0 software (Stat-Ease, Minneapolis, MN, USA).

RESULTS

Formulation and Optimization of Nano Emulsion:

Experimental design employed for the development and optimization of AM-NEs The optimization of Astragalus membranaceus (AM-NEs) was conducted utilizing the Box-Behnken Design (BBD). The present study utilized the ultrasonication technique to adjust the ratios of Castor oil (oil phase), the Smix ratio (Tween-80: surfactant; PEG 400: cosurfactant), and the duration of ultrasonication. The criteria analysed for optimization included Particle size (PS), and Entrapment Efficiency (EE) of the formulated AM-NEs, as outlined in Table 2. The selection of independent variables was predicated on preliminary experimental results. The optimization results indicated a Particle size range of 137.7 nm to 756.37 nm and Entrapment Efficiency range of 34.04% to 93.95% (Table 2). The quadratic model demonstrated the optimal fit for each response, including particle size and Entrapment Efficiency, as evidenced by superior R-squared values relative to alternative models. Polynomial equations (quadratic) for each response were generated utilizing the software as detailed below.

RUN	LIPID	SMIX	SONICATION	PS	EE
1	2	5	12.5	142±0.43	80±0.54
2	8	5	12.5	131±1.31	75±0.67
3	5	5	5	127±0.54	85±0.90
4	2	12.5	20	132±0.12	79±0.32
5	5	12.5	12.5	119±0.76	80±0.12
6	5	20	5	165±0.98	70±0.24
7	8	12.5	20	178±1.43	87±0.97
8	2	20	12.5	198±0.65	79±1.32
9	2	12.5	5	213±0.89	80±0.56
10	5	12.5	12.5	126±0.65	73±0.78
11	8	20	12.5	159±0.32	77±0.87
12	5	5	20	189±0.87	79±0.32
13	8	12.5	5	213±o.54	87±0.92
14	5	12.5	12.5	123±0.56	78±0.12
15	5	20	20	214±1.56	84±0.43
16	5	12.5	12.5	145±1.76	68±0.46
17	5	12.5	12.5	135±0.89	73±0.69

A, B, and C denote the coded values for Castor oil (oil phase), the Smix ratio (Tween-80 and PEG 400), and the duration of ultrasonication (in minutes). A positive sign indicates a beneficial effect, while a negative sign signals a detrimental influence on the dependent variables or responses. The lack of fit for all replies was found to be

insignificant (P > 0.05) at a 95% confidence level. Moreover, the other parameters of the quadratic model were highly statistically significant, with P < 0.0001, indicated by elevated F-values and a sufficient precision value exceeding 3.0. This suggests that the model fit well and provided sufficient signal.

Impact of independent variables on globule size

The globule size of AM-NEs varied from 137.7 nm to 756.37 nm. The influence of the variables on globule size is elucidated in Polynomial Equation (01), accompanied by 3D and contour plots that depict these effects (Fig. 2A). The oil phase exhibited a significant impact on the size of nanoemulsion globules. Raising the oil fraction from 2.0% to 10.0% led to increased globule diameters attributable to particle aggregation. The Smix factor significantly influenced the outcome: an initial reduction in globule size was noted when concentration increased from 5.0% to 20.0%, followed by a subsequent increase. The first decrease in size may be ascribed to diminished interfacial tension between phases, which impedes particle agglomeration.[34] Ultrasonication promotes the disintegration of globules. Prolonging the ultrasonication duration leads to a decrease in the globule size of the synthesized nanoemulsion, attributable to the fragmentation of globules into smaller dimensions.

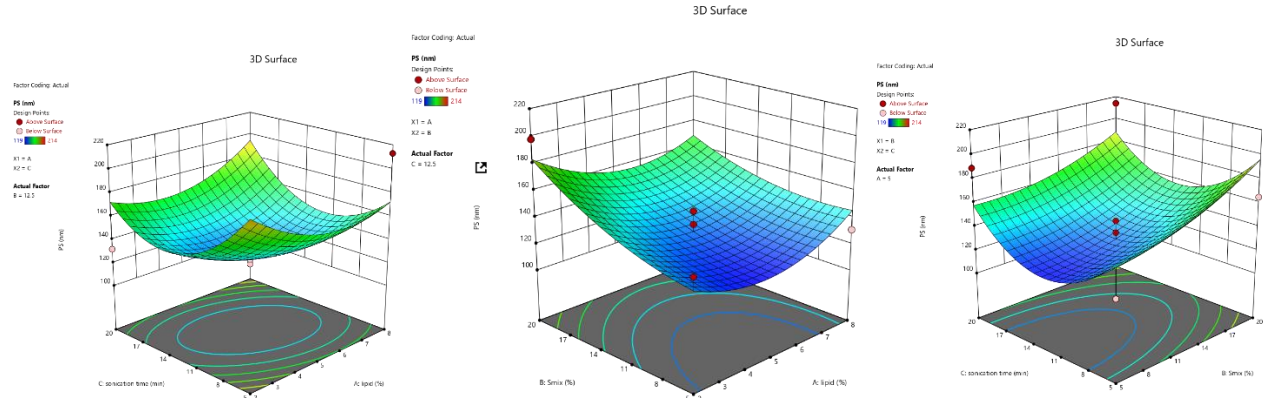


Fig: Effect of Independent on Particle size.

Impact of independent variables on Entrapment Efficiency

Influence of Independent Variables The EE% of AM-NEs fluctuated according to the formulation variables, as demonstrated in the polynomial model and associated response surfaces. Augmenting the oil concentration enhanced the encapsulation efficiency percentage by facilitating medication solubilization within the dispersion phase. The Smix ratio had a biphasic effect: EE% first rose due to enhanced interfacial stabilization, then decreasing at elevated levels where surplus surfactant promoted drug partitioning into mixed micelles. Sonication length had a moderate impact, with ideal times facilitating effective entrapment, but extended sonication diminished EE% due to interfacial rupture and drug leakage into the aqueous phase.

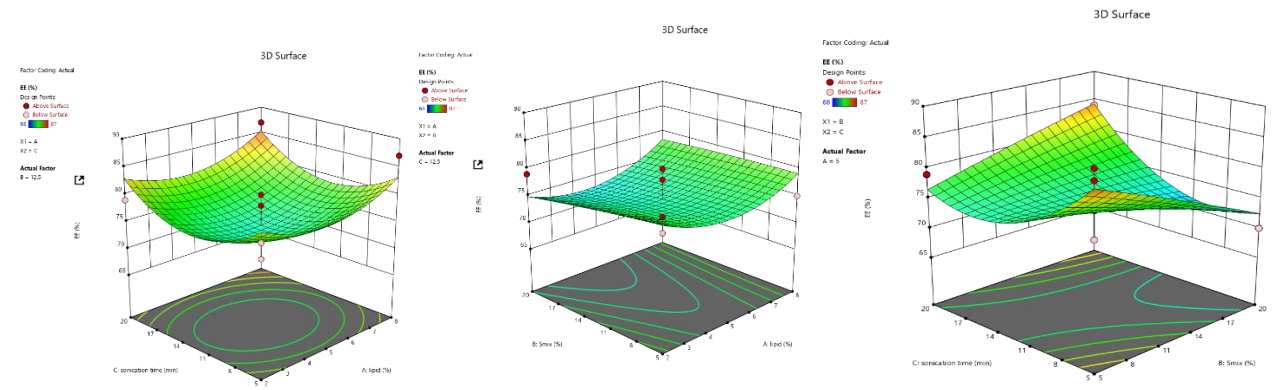
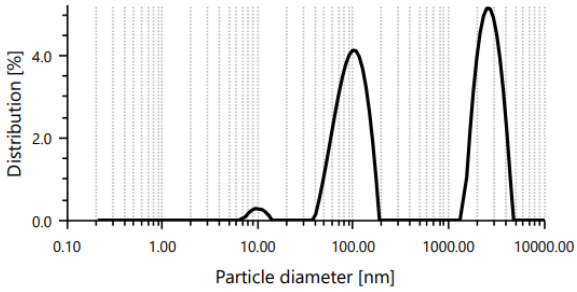


Fig: Effect of Independent factor on Entrapment Efficiency.

Characterisation

Particle size, PDI and zeta potential

Optimized AM- NEs formulation presented particle size of 162.76 ± 8.47 nm, PDI of 0.272 ± 0.029. The particle size and size distribution curve of the optimized AM-NEs formulation is depicted in Fig. 2A. The zeta potential curve of optimized formulation is shown in Fig. 2 B. The observed zeta potential of telmisartan loaded NLC was −35 mV, ensuring that the particles in the formulation are stable. The zeta potential values greater than 30 mV or less than −30 mV suggest full electrostatic stabilization.[28-30]



Result

Hydrodynamic diameter	162.76 nm	**Mean intensity**	336.1 kcounts/s
Polydispersity index	24.8 %	**Absolute intensity**	111697.6 kcounts/s
Diffusion coefficient	3.0 µm²/s	**Intercept g1²**	0.5987
Transmittance	63.9 %	**Baseline**	1.203

Particle size distribution peaks (intensity)

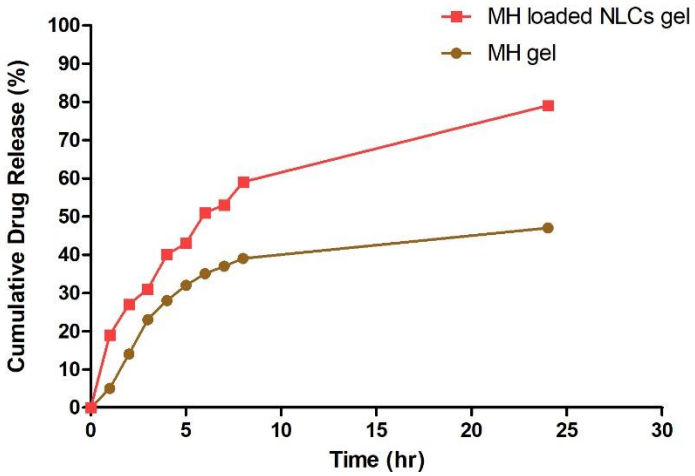
Peak name	Size [nm]	Area [%]	Standard deviation [nm]
Peak 1	2711	49.65	668.8
Peak 2	98.89	48.70	33.60
Peak 3	10.03	1.65	1.59

Entrapment Efficiency

The entrapment efficiency of the optimized telmisartan loaded NLC formulation was found to be 83.72 ± 3.85%

In Vitro drug Release

A release study based on *in vitro* examination was conducted to determine the release profile of AM from NE using a dialysis membrane at 37.0 ± 1.0 ◦C in pH 7.4 PBS. The average % cumulative release of AM after 24 h from AM-NEs was found to be 67.89 % ± 6.33, respectively (Fig. 6A). Korsemeyer-Peppas kinetics release model was found to be suitable for our optimized AM-NEs, with an r2 value of 0.9831.



Morphological studies: (NLC 23)

An exact morphology (size, shape, and internal structure) of developed nanoparticles could be analyzed by transmission electron microscopy. The optimized AM-NEs formulation had an almost spherical surface with a uniform size distribution, as seen in TEM images. Fig. 2 (d) shows a TEM photomicrograph of the optimized AM-NEs formulation.

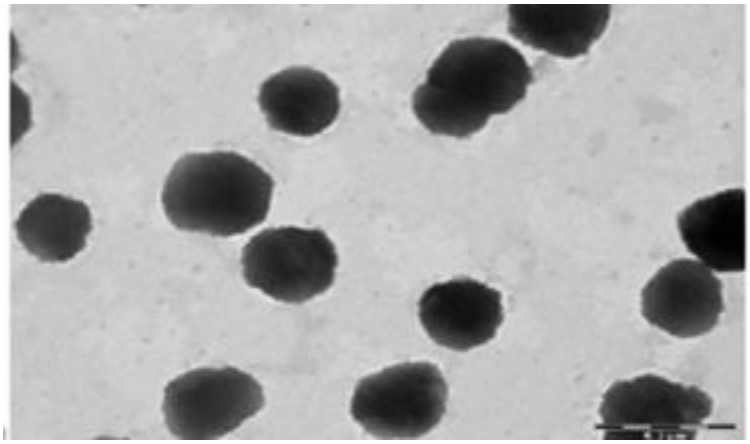


Fig: TEM of Astragalus membranaceus loaded Nano Emulsion

Evaluation of antidiabetic activity

5.1.8. Evaluation of Antidiabetic activity* of *Astragalus membranaceus

5.1.8.1. Effect on body weight

In the present study, diabetic control rats exhibited a marked loss in body weight together with increased food and water intake, reflecting the classical symptoms of diabetes. Treatment with AM-NEs extract for 21 days significantly improved gradual rise in body weight when compared with the diabetic control group. The effect was found to be dose-dependent, with the higher dose showing a greater impact. The standard drug Metformin produced gradually rise in body weight along with better results as compared to AM-NEs. These findings suggest that AM-NEs helps to correct metabolic disturbances in diabetes. The detail was given below in table no.

Table 5.5: Effect of AM-NEs on body weight in Experimental Diabetic Rats.

Group	Initial body Weight (Day 0)	Final body Weight (Day 21)	Body Weight change
Normal Group	159.41±0.71	192.5±2.57	23.1
Control Group	198±0.44	181.5±0.954	-15.4
Test-I (250mg/Kg)	171.8±0.74	152.5±1.155	-9.2
Test-II (500mg/Kg)	180.5±1.35	193.82±1.15	13.2
Standard Group	174.5±1.89	202.35±1.57	25.4

The diabetic control group showed a significant loss in body weight (–19.5 g) during the 21-day experimental period, reflecting the catabolic effect of uncontrolled diabetes. In contrast, treatment with AM-NEs improved body weight in a dose-dependent manner. The AM-NEs produced a notable increase in body weight (+13.54 g), compared to the diabetic control. The standard drug Metformin showed the maximum improvement (+23.35 g), restoring body weight close to the normal group. These findings confirm that AM-NEs helps counteract diabetes-induced weight loss, supporting its role in improving metabolic balance.

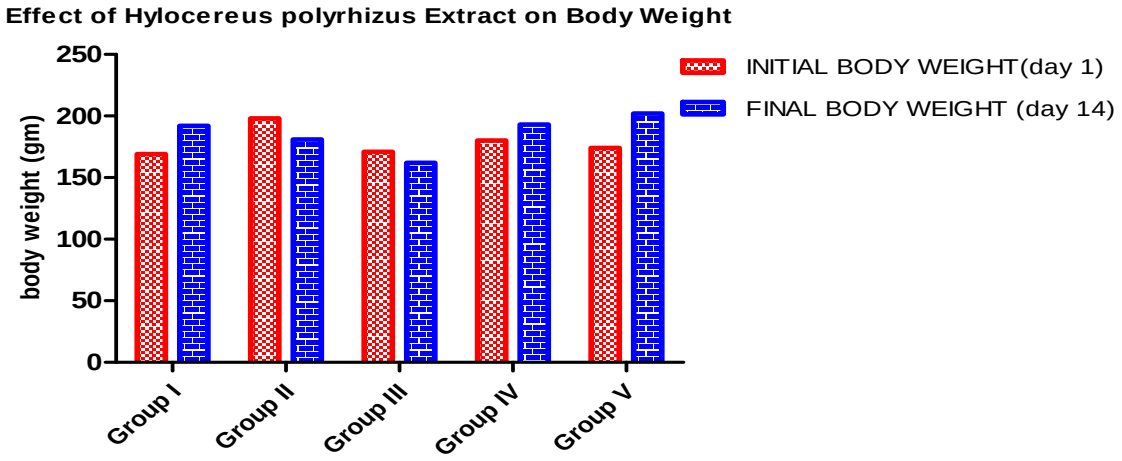


***Figure 5.2:* Effect of *H. polyrhizus extract* in body weight of STZ induced diabetes in rats. Values are expressed as mean ± SEM (number of animals, n = 4).**

5.1.8.2. Effect on blood glucose level

Fasting blood glucose level is an important parameter for evaluating the antidiabetic activity of AM-NEs. In the present study, treatment with AM-NEs produced a significant, dose-dependent reduction in fasting blood glucose levels in streptozotocin-induced diabetic rats. The standard group treated with Metformin also showed a highly significant reduction in blood glucose levels when compared with the diabetic control group. These findings indicate that AM-NEs may be effective in the management of diabetes (Table 5.6).

Table 5.6: Effect of AM-NEs on blood glucose level in Experimental Diabetic Rats.

Sr. No.	Groups	Blood glucose level in mg/dl				
		0^{th} day	3^{th} day	7^{th} day	14^{th} day	21^{th} day
1.	Normal control	125±2.58	119±0.98	123±1.34	115±1.34	124±1.35
2.	Diabetic control	495±1.54	482±2.55	454±2.34	425±2.35	397±0.57
3.	Test 1 (250 mg/kg)	481±3.55	412±3.24	335±0.57	257±1.54	148±1.54
4.	Test 2 (500mg/kg)	478±1.24	408±3.14	312±1.54	233±3.14	125±2.05
5.	Standard group	458±2.55	381±3.24	289±3.21	203±2.58	108±1.94

Treatment with the standard drug Metformin significantly reduced blood glucose levels ($P < 0.001$) and restored them close to the normal range. Similarly, AM-NEs produced a gradual and dose-dependent decrease in fasting blood glucose on days 0, 3, 7, 14, and 21 when compared with the diabetic control group. The reduction was statistically significant at ($P < 0.05$, $P < 0.01$, and $P < 0.001$) depending on the treatment day and dose. These findings suggest that the extract of AM-NEs possesses significant antihyperglycemic activity and may serve as a promising natural excipient for the development of antidiabetic formulations.

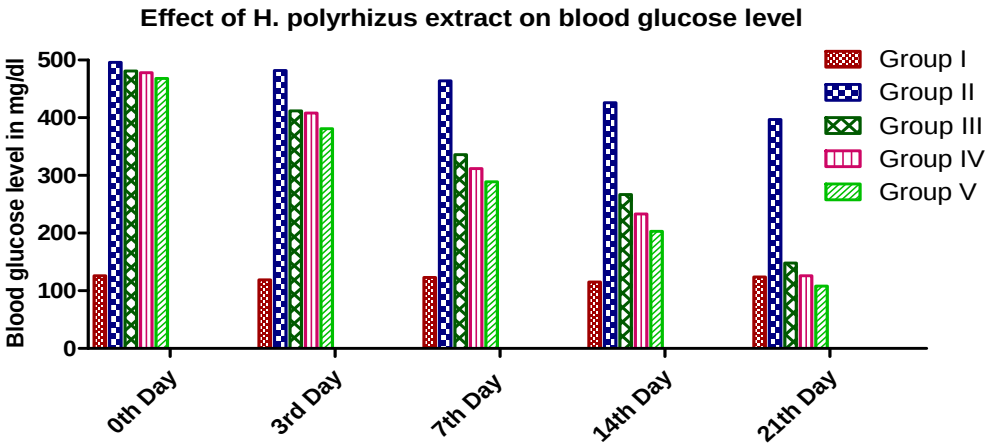


Figure 5.3: **Effect of *H. polyrhizus extract* in blood glucose level of STZ induced diabetes in rats. Values are expressed as mean ± SEM (number of animals, n=4).**

5.1.8.3. Effect on Lipid profile in diabetic rats

Lipid profile is a crucial parameter in evaluating the antihyperlipidemic effect of therapeutic agents, as dyslipidemia is one of the major complications associated with diabetes mellitus. These alterations reflect a typical diabetic dyslipidemic state, characterized by hypercholesterolemia, hypertriglyceridemia, and low HDL levels, which predispose to atherosclerosis and cardiovascular complications. The antihyperlipidemic activity of AM-NEs may be attributed to its rich phytochemical composition, particularly flavonoids, phenolic compounds, and dietary fibers, which are known to regulate lipid metabolism by enhancing LDL clearance, inhibiting cholesterol absorption, and improving reverse cholesterol transport.

Table 5.7 *Effect of AM-NEs on Lipid Profile in Experimental Diabetic Rats.*

Sr. no.	Groups	Total cholesterol (mg/dl)	Triglyceride (mg/dl)	LDL (mg/dl)	HDL (mg/dl)
I	Normal	82.35±2.5	58.2±1.5	47.2±1.8	48.5±2.9
II	Diabetic control	124±3.17	104.5±2.7	93.5±2.3	28.2±1.2
III	Test-I (250mg/kg)	113±1.1	98.2±1.3	80.2±2.5	34.1±1.3
IV	Test-II (500mg/kg)	104±2.5	89.5±2.5	57.3±2.8	39.4±1.81
V	Standard	92±4.15	53.5±4.31	55±3.23	45.45±2.4

In the present study, streptozotocin-induced diabetic rats (diabetic control group) showed a marked increase in total cholesterol (133 ± 3.17 mg/dl), triglycerides (118.5 ± 2.7 mg/dl), and LDL (104.5 ± 2.3 mg/dl), along with a significant reduction in HDL (28.2 ± 1.2 mg/dl), compared to the normal group. Treatment with AM-NEs significantly improved the lipid profile in a dose-dependent manner. These findings indicate that AM-NEs possesses both antidiabetic and antihyperlipidemic potential and may help in correcting dyslipidemia associated with diabetes (Table 5.7).

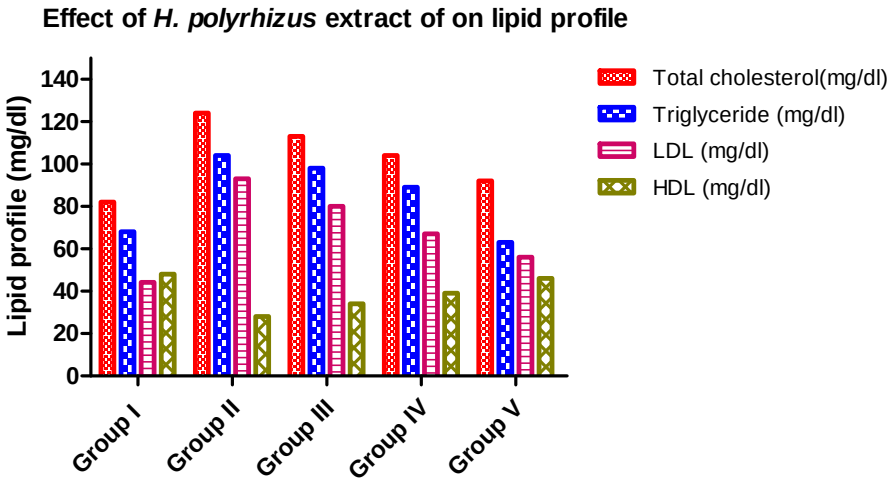


Fig 5.4: Effect of AM-NEs in lipid profile of STZ-induced diabetes in rats. Values are expressed as mean ± SEM (number of animals, n = 4).

CONCLUSION

The present study successfully developed and optimized a nanoemulsion system for the efficient delivery of *Astragalus membranaceus* bioactives using a systematic experimental design approach. The optimized formulation exhibited desirable physicochemical characteristics, including nanoscale particle size, high entrapment efficiency, uniform morphology, and satisfactory electrostatic stability, indicating effective formulation robustness. In vivo evaluation in streptozotocin-induced diabetic rats revealed significant improvement in metabolic parameters, including reduction in fasting blood glucose levels, restoration of body weight, and correction of diabetes-associated dyslipidemia in a dose-dependent manner. These findings suggest enhanced bioavailability and therapeutic efficiency of the encapsulated phytoconstituents.

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