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Formulation And Evaluation Of Optimized Transferosomal Gel Of Repaglinide And Its Patches For Enhanced Transdermal Delivery For Antidiabetic Activity

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Abstract
The current study aimed to develop and optimize a transferosomal gel and transdermal patch formulation of repaglinide to enhance its transdermal delivery and antidiabetic efficacy. Transferosomes were formulated using the reverse phase evaporation method and optimized through a full factorial design considering particle size and entrapment efficiency as critical parameters. The optimized formulation (NTF7) displayed a particle size of 266.9 nm, a zeta potential of 33.2 mV, and high entrapment efficiency of 97.8%, indicating excellent stability and effective drug encapsulation. In vitro drug release from the transferosomes and the Carbopol 934-based transferosomal gel (NTFG7) showed sustained release of 97.95% and 96.54%, respectively, over 12 hours, following Korsmeyer-Peppas kinetics indicative of a non-Fickian, anomalous release mechanism. Transdermal patches (NTFG7 P) prepared from the optimized gel also exhibited controlled release (94.78% over 12 hours) and favorable physicochemical properties. In vivo studies on STZ and HFD-induced diabetic rats demonstrated significant glucose-lowering effects with both gel and patch formulations, with NTFG7 P achieving greater glycemic control and negligible skin irritation (PDII = 0.33). Histopathological analysis revealed protective effects on pancreatic tissues. The developed transferosomal gel and patch systems represent promising, patient-friendly alternatives to oral repaglinide, offering improved bioavailability, sustained release, and enhanced therapeutic outcomes in diabetes management.
Keywords: *Repaglinide, Transferosomes, Transdermal Patch, Antidiabetic Activity, Nanocarrier, Sustained Release*

1. INTRODUCTION

Complications such as cardiovascular disease, neuropathy, and nephropathy are rising in frequency and severity[1], making diabetes mellitus, a chronic metabolic illness defined by persistent hyperglycemia, a worldwide health problem. Worldwide, 537 million persons were living with diabetes in 2021, "and that figure is expected to increase dramatically by 2045, according to the International Diabetes Federation (IDF) [2].. Among various therapeutic strategies, oral administration of anti-diabetic drugs remains the conventional approach. However, this route often encounters significant limitations, including poor bioavailability, extensive first-pass metabolism, gastrointestinal side effects, and variable absorption. These limitations necessitate the exploration of novel drug delivery systems to enhance therapeutic efficacy while minimizing adverse effects[3]**.** Repaglinide, a short-acting oral insulin secretagogue, is commonly used in the management of type 2 diabetes mellitus. Despite its therapeutic benefits, its clinical use is restricted due to low and inconsistent oral bioavailability (~56%) resulting from hepatic first-pass metabolism [4]**.**Consequently, alternative routes such as transdermal delivery have garnered interest. Transdermal systems offer several advantages including avoidance of first-pass metabolism, controlled drug release, improved patient compliance, and reduced dosing frequency [5]. Nonetheless, the major challenge in transdermal delivery is the formidable barrier posed by the stratum corneum, the outermost layer of the skin. To overcome this obstacle, vesicular carriers such as liposomes, ethosomes, and more recently, transferosomes, have emerged as promising nanocarrier systems. Transferosomes are ultradeformable vesicles composed of phospholipids and edge activators, such as surfactants, that enhance their flexibility and penetration through narrow pores in the skin. Their deformability enables them to squeeze through the intercellular spaces of the skin, thereby facilitating deeper penetration and improved systemic absorption of the encapsulated drug [6]. The optimized formulation was then incorporated into a Carbopol 934-based gel matrix to develop a suitable dermal delivery system. Comprehensive physicochemical characterization, in vitro release studies, and kinetic modeling were carried out to evaluate the performance of the formulation. Furthermore, in vivo antidiabetic efficacy was assessed in streptozotocin (STZ) and high-fat diet (HFD)" induced diabetic Wistar rats. The study also involved histopathological

examination to observe the effects of the formulation on pancreatic tissues. Through this multidisciplinary approach, the research aims to provide a viable alternative to oral repaglinide therapy, offering a more consistent and patient-friendly delivery route. By combining nanotechnology and transdermal delivery principles, the transferosomal gel system holds promise for improving therapeutic outcomes in diabetes management.

2. MATERIAL AND MATERIAL

2.1 Chemicals and Instruments required

The chemicals used in the study included Phosphotidylcholine (CDH), Andrographolide (Sigma), Cholesterol "(CDH), Tween 80 (Loba Chemie), Chloroform and Methanol (Merck), Diethyl ether (Rankem), Dimethyl sulfoxide (DMSO, CDH), and freshly prepared distilled water. Various instruments and glassware were utilized during the formulation and evaluation processes. These included a digital balance (Electrolab the Elico Model-LI612 digital pH meter; the LAB-HOSP sonicator; and the S-4800 TYPE II scanning electron microscope, manufactured by Hitachi High Technologies Corporation of Japan. A centrifuge and mechanical stirrer were acquired from Remi Instruments, Mumbai, and a Zetasizer (Malvern Instruments Ltd.) was used for particle size and zeta potential analysis. Also used in the investigation were a stability chamber (CHM-10 S, Remi Lab, Mumbai), an infrared spectrophotometer (Affinity-1S, Shimadzu, Kyoto, Japan), and an ultraviolet spectrophotometer (Shimadzu UV-1700, Japan).

2.2 Formulation of Transferosomes

2.1Trial formulation of transferosomes

2.1.1Method

Transferosomes were created using the reverse phase evaporation method. Soya lecithin and cholesterol were collected as lipids in a clean beaker. Tween 20 was dissolved in a solvent mixture of diethyl ether and chloroform, maintained at room temperature for 24 hours. Repaglinide (10mg) was dissolved in PBS 7.4 pH buffer saline solution, hydrated with phosphate buffer saline, and sonicated for 2 minutes to obtain a transferosomal suspension [7].

Table 1. Composition and Particle Size of Trial Transferosomal Formulations

S.N	Formulation Code	Phosphotidylcholine (mg)	Cholesterol (mg)	Organic Solvent Ratio (DCM:Methanol)	PBS Buffer (pH 7.4, ml)	Particle Size (nm)
1	NTF1	30	30	1:1	15	230.4
2	NTF2	50	25	1:1	15	649.0
3	NTF3	25	50	1:1	15	661.1
4	NTF4	30	30	2:1	15	443.8
5	NTF5	50	25	2:1	15	314.2
6	NTF6	25	50	2:1	15	560.2
7	NTF7	30	30	1:2	15	690.2
8	NNTF7	50	25	1:2	15	438.2
9	NTF9	25	50	1:2	15	550.7
10	NTF10	30	30	3:1	15	221.5
11	NTF11	50	25	3:1	15	332.1
12	NTF12	25	50	1:3	15	549.7
13	NTF13	30	30	1:3	15	360.2

2.1.2 Particle size

In order to determine the size of the droplets, a Malvern Particle Sizer and Zeta Potential Analyzer (Malvern Instruments Ltd., UK) was employed at room temperature. Dissolved in one milliliter of deionized water was the transferosome suspension [8].

2.1.3 Entrapment Efficiency

The entrapment efficiency of repaglinide in transfersomes was assessed using a centrifugation method. The prepared transfersomal dispersion was centrifuged at 14,000 rpm for 30 minutes, and 1 ml of the resulting supernatant was collected and diluted with PBS (pH 7.4). The amount of unentrapped

repaglinide in the supernatant was then quantified using a UV spectrophotometer after appropriate dilution.

%Entrapment Efficiency = (Total Drug- Unentrapped drug) / Total Drug X 100

Table 2: Entrapment efficiency of the trial formulation

S.N	Formulation code (trial formulation)	Entrapment efficiency (%)
1	NTF1	84.1
2	NTF2	92.6
3	NTF3	83
4	NTF4	95.8
5	NTF5	69.9
6	NTF6	85.4
7	NTF7	96.2
8	NNTF7	93.8
9	NTF9	87.5
10	NTF10	71.2
11	NTF11	91.7
12	NTF12	89.5
13	NTF13	95.8

2.2 Optimization of formulation parameters

Design of experiments (DOE), first introduced by Ronald A. Fisher, helps plan experiments systematically to draw objective conclusions. In this study, a two-factor, three-level full factorial design was applied using DOE++ software. Polynomial equations were generated to link independent variables with outcomes like particle size and entrapment efficiency. The desirability function was then used to optimize the formulation by balancing multiple responses to achieve the desired characteristics[9].

Table 3: DOE run for the optimized formulation

Std	Rum	Phospholipid (mg)	Surfactant (%0	Sonication time (Min.)	Particle size	Entrapment efficiency
8	1	100	1.75	60	438.2	93.8
1	2	50	0.5	45	230.4	84.1
6	3	100	1.75	30	560.2	96.2
9	4	75	0.5	30	550.7	87.5
2	5	100	0.5	45	649	92.6
13	6	75	1.75	45	360.2	95.8
11	7	75	0.5	60	332.1	91.7
12	8	75	3	60	549.7	89.5
10	9	75	3	30	221.5	71.2
5	10	50	1.75	30	314.2	69.9
3	11	50	3	45	661.1	83
7	12	50	1.75	60	690.2	85.4
4	13	100	3	45	443.8	95.8

Table 4: Independent and dependent variables

Factor	Name	Units	Type	Sub Type	Minimum	Maximum	Coded Low	Coded High	Mean	Std. Dev.
A	Phospholipid	mg	Numeric	Continuous	50.00	100.00	-1 ↔ 50.00	+1 ↔ 100.00	75.00	20.41
B	Surfactant	%	Numeric	Continuous	0.5000	3.00	-1 ↔ 0.50	+1 ↔ 3.00	1.75	1.02
C	Sonocation time	Min.	Numeric	Continuous	30.00	60.00	-1 ↔ 30.00	+1 ↔ 60.00	45.00	12.25

2.2.1 Experimental design

We used a factorial design to look at the elements in a methodical way. The answers were evaluated using a statistical model that included interaction and polynomial variables, as shown in the equation.

Yi = b0 +b1X1 + b2X2 + b12X1X2 + b11X12 + b22X22.....

Where Yi (Y1, Y2) were the dependent variables,

Deformability Index and Particle size, b0 is the arithmetic mean response of the 13 runs, b1 and b2 are the estimated coefficients for the factors X1 and X2, respectively.

2.3 Optimized formulation preparation

The Transferosomes were created using a reverse phase evaporation method. Soya lecithin and cholesterol were used as lipids, followed by Tween 80 as a surfactant. The beaker was kept at room temperature for 24 hours until a thin film formed. Drug repaglinide was dissolved in PBS 7.4 pH buffer saline solution, hydrated using edge deactivator sodium deoxycholate, and sonicated for 60 minutes to obtain transferosomal suspensions with particle size in nm.

2.4 Characterisation of optimised transferosomes formulation

2.4.1 Particle size and Zeta potential

A Malvern Particle sizer and Zeta Potential Analyzer were used to measure the droplet size and zeta potential of the transferosomes at room temperature. Electrophoretic mobility and the Henry Equation were used to determine the zeta potential. The majority of zeta experiments are performed in aqueous systems; nevertheless, zeta potential will be present in any colloids distributed in a solvent with a high dielectric constant[10].

2.4.2 Entrapment efficiency

The drug was encapsulated in transfersomes using a centrifugation method. The supernatant was diluted with PBS and the unentrapped drug was determined using a UV spectrophotometer. The total amount of un-entrapped drug was determined from the free drug in the supernatant. Encapsulation efficiency was measured as the percentage of drug trapped.

%Entrapment Efficiency = (Total Drug- Unentrapped drug)/Total Drug X 100

2.4.3 SEM

Surface properties including surface smoothness and diameter homogeneity are crucial metrics for assessing Transferosomes. The transferosomes' surface morphology was examined using a scanning electron microscope.

2.4.4 In vitro drug release of transferosomes

The study used Hao's method with dialysis bags in isotonic buffer to monitor drug release over 12 hours. Data fitted to kinetic models showed gradual release, with calculated Higuchi (KH) and Korsmeyer–Peppas (Kp) rate constants.

2.5 Formulation of optimised transferosomal gel

Transferosomal gel was formulated with transferosomes, Carbopol 934, propylene glycol, and methyl paraben. After dispersing Carbopol in water, ingredients were mixed and stirred to form a smooth gel, with pH adjusted by triethanolamine.

Table 5: Formulation of Gels of transferosomal gel

Chemical required	**NTFG7**
Transferosomes	100 mg
Carbopol 934	0.25%
Triethanolamine	q.s (2-3 drops)
Propyl paraben	0.1%
Ethanol	0.5 %
Distilled Water	q.s

2.6 Characterisation of the optimized transferosomal gel

2.6.1 Visual characterisation

The physical appearance or visual characterisation of the gel was analysed by the visually and normal human senses. The organoleptic properties included colour, odor, and texture.

2.6.2 Physical characterisation

2.6.2.1 Homogeneity:

Physical appearance and homogeneity of the prepared gels were evaluated by visual perception.

2.6.2.2 pH

The pH of the transfersomal gel was measured at room temperature using a calibrated pH meter and a Calomel electrode after dispersing 1 g of gel in water.

2.6.2 Evaluation of Transferosomal Gel: Swelling Index, Viscosity, and In Vitro Drug Release

A gram of gel was placed on porous aluminum foil in 0.1 N NaOH to measure the swelling index, calculated as [(Wt – Wo)/Wo] × 100, where Wo is the initial weight and Wt is the swollen weight at time t. The viscosity was determined using a Brookfield Viscometer at 60 rpm, recording steady readings after 5 minutes. For in vitro drug release, transferosomal gels were tested using Hao's method in isotonic buffer, with samples analyzed over 12 hours to study release kinetics. Additionally, formulations (NTFG7) were stored at three temperatures and 75% relative humidity for 3 months, with periodic checks on drug content and physical stability.

2.7 Formulation of Transferosomes patches

Aluminum foil was used as a backing membrane in the solvent casting process to combine the potential transferosome formulations into a transdermal patch. Preliminary research was used to determine the concentration and content of the patch. Using propylene glycol (15 percent w/w of total polymer) as a permeation enhancer and polyethylene glycol 400 (30 percent w/w of total polymer) as a plasticizer, a transferosomal patch was created. Then, with careful stirring, a predetermined quantity of transferosome gel was added to the polymer solution to create a homogenous, homogeneous mixture. After the produced solution was transferred onto a petridish with aluminum foil serving as a backing membrane, it was placed in a desiccator to dry at 400 degrees until the solvent had completely evaporated[11].

2.7.1 Evaluation parameter

2.7.1.1 Tensile strength, Thickness, Folding endurance and Percentage Moisture Content

The % elongation and tensile strength were tested using a lab-made pulley system, while patch thickness was measured at three points with a Vernier caliper to ensure dose uniformity. Folding endurance was checked by repeatedly folding a 2×2 cm patch strip until it broke, indicating brittleness. Finally, patches were weighed before and after 24 h in a desiccator to calculate moisture content.

Percentage moisture content = [(Initial weight—Final weight)Final weight]×100.

2.8 Animals required

Wistar rats (150–200 g) were housed in clean polypropylene cages at 22 ± 2 °C with a 12-hour light/dark cycle and fed a standard pellet diet and water ad libitum throughout the study.

2.8.1 In-vivo anti-diabetic activity

Rats were first acclimatized, then fed a high-fat diet for four weeks to induce insulin resistance. After overnight fasting, they received intraperitoneal STZ to trigger partial beta-cell dysfunction. Fasting blood glucose was checked 48 hours later, and the high-fat diet was continued, with regular monitoring of food, water intake, and body weight.

2.8.2 Histopathological Examination

After being serially dehydrated in alcohol and cleaned in xylene, the pancreatic tissue sections that had been fixed with formalin were embedded in paraffin blocks. Following a routine procedure, the micro sections (4-5 microns thick) were cut, stained with hematoxylin and eosin (H and E), and checked for histological alterations[12].

2.9 Statistical Analysis

The findings are given as Mean ± SD (n=6). The Bonferroni t-test and one-way analysis of variance (ANOVA) were used for statistical analysis of the results. When comparing groups, a significance criterion of $P < 0.05$ was used.

3. RESULTS

3.1 ANOVA for Quadratic model

Table 6: Response 1: Particle Size

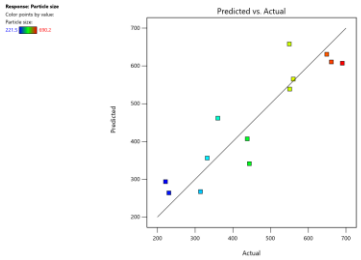
Source	Sum of Squares	df	Mean Square	F-value	p-value	
Model	2.608E+05	6	43459.30	4.96	0.0362	significant
A-Phospholipid	4767.76	1	4767.76	0.5440	0.4886	
B-Surfactant	1621.65	1	1621.65	0.1850	0.6821	
C-Sonocation time	16525.62	1	16525.62	1.89	0.2188	
AB	1.011E+05	1	1.011E+05	11.53	0.0146	
AC	62001.00	1	62001.00	7.07	0.0375	

BC	74747.56	1	74747.56	8.53	0.0266	
Residual	52588.36	6	8764.73			

3.1.1 Final Equation in Terms of Coded Factors

Particle Size +461+24.41A+14.24B+45.45C-158.97 AB-124.50 AC+136.70 BC

For certain amounts of each element, predictions on the response may be made using the equation expressed in terms of coded factors. By default, the factors' high and low values are denoted by the numbers +1 and -1, respectively. By comparing the factor coefficients, the coded equation may be used to determine the relative importance of the elements.



Graph 1: Countor plot showing the actual factor dependent variables against particle size, scatter plot showing the actual factor dependent variables against particle size

Table 71 : Response 2: Entrapment Efficiency

Source	Sequential p-value	Adjusted R^2	Predicted R^2	
Linear	0.0219	0.5201	0.4851	Suggested
2FI	0.3347	0.5749	0.1519	
Quadratic	0.4316	0.6206		
Cubic				Aliased

3.2 ANOVA for Linear model

Table 8: Response 2: Entrapment Efficiency

Source	Sum of Squares	df	Mean Square	F-value	p-value	
Model	584.04	3	194.68	5.34	0.0219	significant
A-Phospholipid	392.00	1	392.00	10.74	0.0096	
B-Surfactant	33.62	1	33.62	0.9215	0.3622	
C-Sonocation time	158.42	1	158.42	4.34	0.0669	
Residual	328.36	9	36.48			
Cor Total	912.40	12				

3.2.1 Final Equation in Terms of Coded Factors

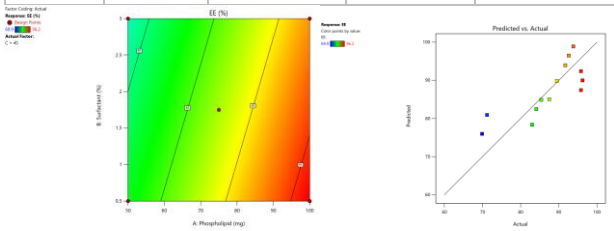
EE =+87.42+7.00A-2.05 B++4.45C

It is possible to anticipate the reaction for certain amounts of each element by using the equation in terms of coded factors. The factors' high and low values are by default coded as +1 and -1, respectively. By comparing the factor coefficients, one may determine the relative importance of the elements using the coded equation.

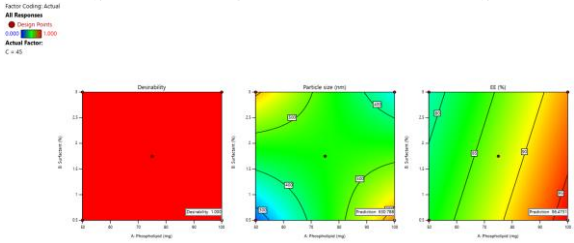
Table 9: actual and predicted value for independent variables

Run Order	Actual Value	Predicted Value	Residual	Internally Studentized Residuals	Externally Studentized Residuals	Cook's Distance	Influence on Fitted Value DFFITS	Standard Order
1	93.80	98.87	-5.07	-1.024	-1.027	0.127	-0.716	8
2	84.10	82.47	1.63	0.328	0.311	0.013	0.217	1
3	96.20	89.97	6.23	1.257	1.305	0.192	0.909	6

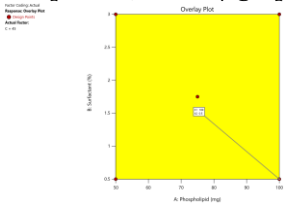
4	87.50	85.02	2.48	0.500	0.478	0.030	0.333	9
5	92.60	96.47	-3.87	-0.782	-0.763	0.074	-0.532	2
6	95.80	87.42	8.38	1.443	1.552	0.043	0.448	13
7	91.70	93.92	-2.22	-0.449	-0.428	0.024	-0.298	11
8	89.50	89.82	-0.3231	-0.065	-0.061	0.001	-0.043	12
9	71.20	80.92	-9.72	-1.962	-2.445	0.467	-1.704(1)	10
10	69.90	75.97	-6.07	-1.226	-1.266	0.182	-0.882	5
11	83.00	78.37	4.63	0.934	0.926	0.106	0.646	3
12	85.40	84.87	0.5269	0.106	0.100	0.001	0.070	7
13	95.80	92.37	3.43	0.692	0.670	0.058	0.467	4”



Graph 2: Counter plot showing the actual factor dependent variables against entrapment efficiency, Scatter plot showing the actual factor dependent variables against entrapment efficiency



Graph 3: a; overlay graph and b,c response surface plot for particle size and entrapment efficiency



Graph 4: overlay plot for the optimized dependent variables

Factors for the optimized formulation

Name	Goal	Lower Limit	Upper Limit	Lower Weight	Upper Weight	Importance
A:Phospholipid	is in range	50	100	1	1	3
B:Surfactant	is in range	0.5	3	1	1	3
C:Sonocation time	is in range	30	60	1	1	3
Particle size	none	221.5	690.2	1	1	3
EE	none	69.9	96.2	1	1	3

3.3 Optimized formulation preparation

Table 20: The optimized formulation of transferosomes

S.N	Optimized Formulation code	Repaglinide	Phosphotidylcholine (mg)	Cholesterol (mg)	Surfactant Tween 20	PBS buffer 7.4 Ph (ml)
1	NTF7	5mg	25 mg	50mg	1 ml	20 ml

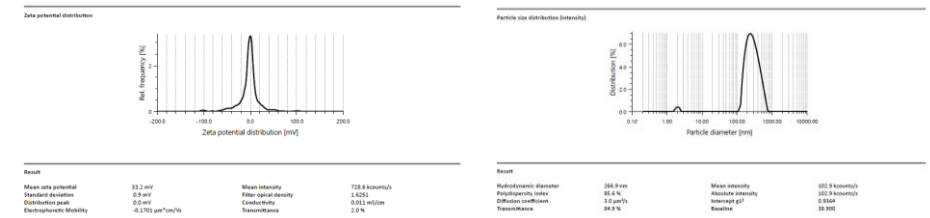
3.4 Characterisation of optimised transferosomes formulation

3.4.1 Particle size and Zeta potential

Table 31: showing the Particle Size Distribution, Polydispersity Index and Zeta potential of optimized formulation

S.N	Parameters	Formulation code NTF7
1	Particle sizer	266.9 nm
2	PDI	85.6 %
3	Zeta Potential	33.2 mv

The particle size and zeta potential of the transferosomes of the optimized formulation NTF7 was 266.9 nm and 33.2 mv. Zeta potential result shows that the formulation was stable, as the charges increases formulation stability increases.



Graph 4: zeta potential and Particle size of optimized formulation NTF7

3.4.2 Entrapment efficiency

Table 42: Entrapment efficiency of the optimised formulation NTF7

S.N	Parameters	Formulation code NTF7
1	Entrapment Efficiency	97.8 ±0.02

The entrapment efficiency of the transferosomes of the optimized formulation of NTF7 was 97.8 % that means the vesicles entrap the drug 97.8 percent and rest amount was the free drug content.

3.4.3 SEM

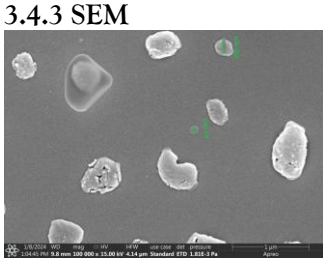
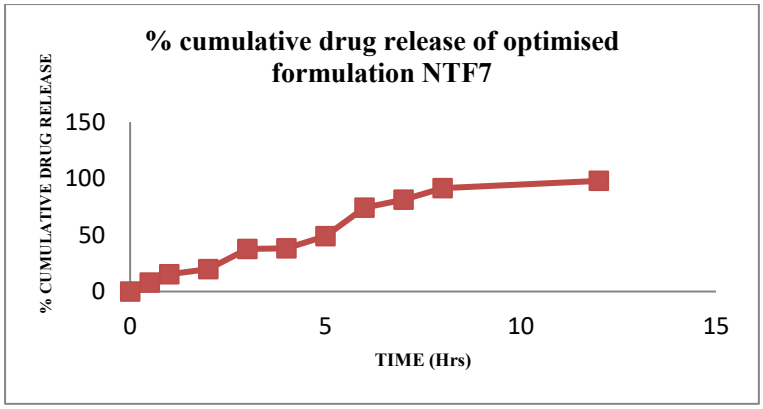


Figure 1: SEM image of the optimize transferosomes formulation NTF7

3.4.4 In vitro drug release of transferosomes

Table 53: represent the % cumulative drug release of the transferosomes loaded with drug

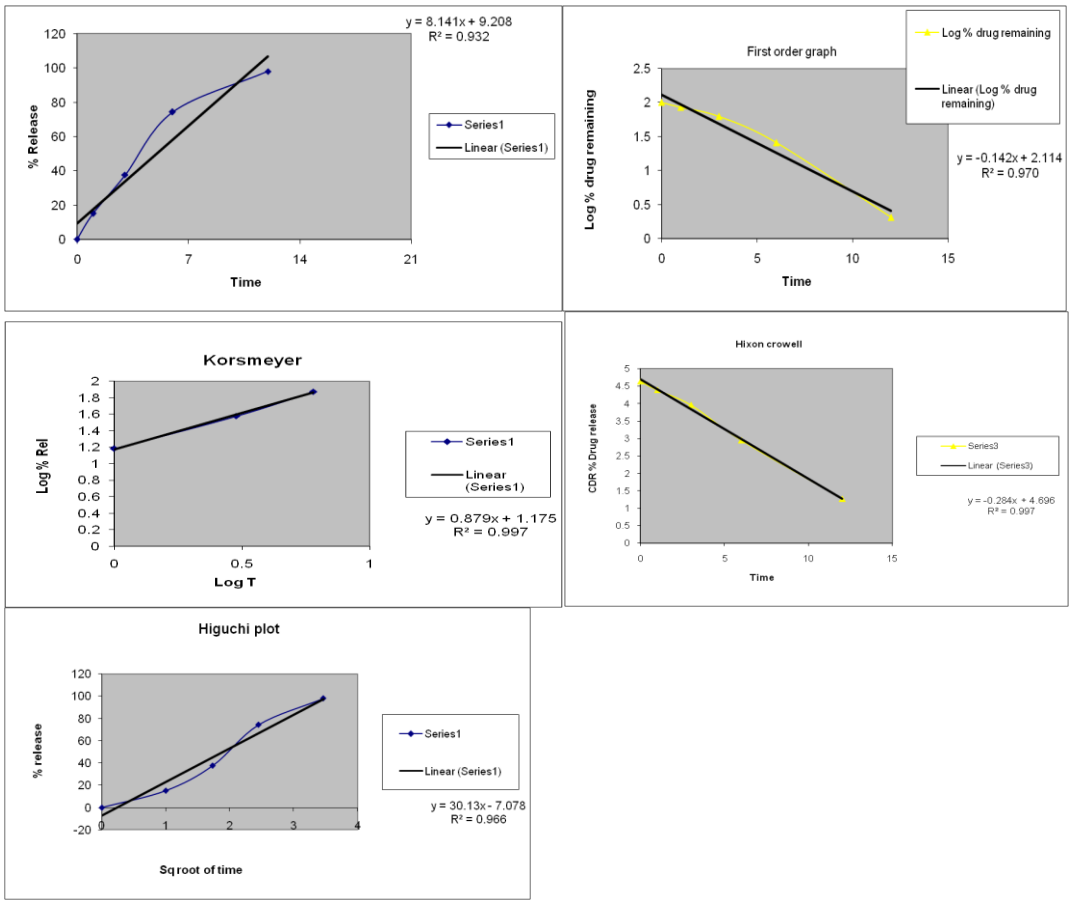
S.N.	Time (hrs)	NTF7
1	0	0
2	0.5	7.89 ± 0.45
3	1	15.22±0.32
4	2	19.86 ±0.26
5	3	37.68 ±0.44
6	4	38.22±0.68
7	5	49.11±0.72
8	6	74.31±0.19
9	7	81.27± 0.28
10	8	91.47±0.24
11	12	97.95±0.51



Graph 5: % cumulative drug release of NTF7

The % cumulative drug release of optimized formulation shows 94.95 % at 12 hrs.

3.4.5 Kinetic release model for the optimized formulation NTF7



Graph 6: zero order, First order, Korsmeyer, Hixon crowell, Higuchi kinetic release models for NTF7

Table 64: represent the kinetic release model regression values of the transferosomes NTF7 loaded drug

S.N.	Formulation code	Zero Order Release (R^2)	First order kinetic release (R^2)	Higuchi kinetic release (R^2)	Korsmeyer kinetic release (R^2)	Hixon Crowell Kinetic release (R^2)
1	NTF7	0.932	0.970	0.966	0.997	0.997

The study evaluated the drug release from the NTF7 transferosomal formulation, which showed a higher cumulative release owing to the vesicles' notable flexibility. The optimized formulation sustained drug release over 12 hours. Analysis of the in vitro release data indicated that the release followed first-order

kinetics and fit best to the Korsmeyer–Peppas model, suggesting a non-Fickian anomalous mechanism (R^2=0.997). Additionally, visual examination confirmed that all dermal gel formulations were smooth, translucent, non-greasy, and free from particulate matter

3.5 Characterisation of the optimized transferosomal gel

3.5.1 Visual characterisation

Table 15: represent the physical properties of the transferosomal gel loaded with drug

S.N.	Formulation code	Parameters	Result
1	NTFG7	Color	Off white in color
		Texture	Smooth
		Appearance	Pleasant
		Odour	Odourless
		Gritty	Non gritty

The homogeneity of the transferosomal gel of the optimised formulation were show that the formulation NTFG7 was properly homogenised mixture . This uniformity was required for the proper penetration of the drug.

3.5.2 Physical characterisation

Table 16: represent the homogeneity, pH, swelling index, Viscosity of the transferosomal gel loaded with drug

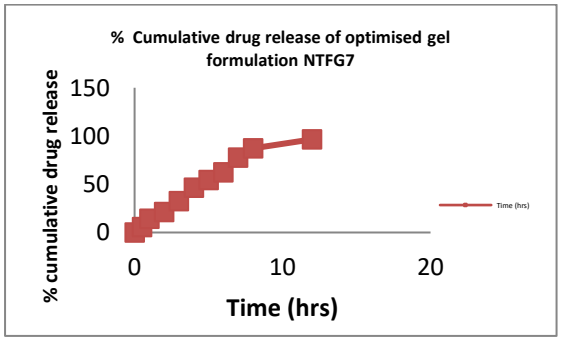
S.N.	Formulation code	Parameters	Result
1	NTFG7	**Homogeneity**	Uniform mixture
		Swelling index (%)	65.11±0.02
		pH.	7.7±0.05
		Viscosity (Viscosity (cps) at Room Temperature ± SD.)	11279±0.63

The pH of the optimised formulations from was found to be in the range of 6 to 7 ideally, the dermal gel should possess pH in the range of 6-7.The pH of the transferosomal gel was 7.7±0.05.

3.5.3 In -vitro drug release

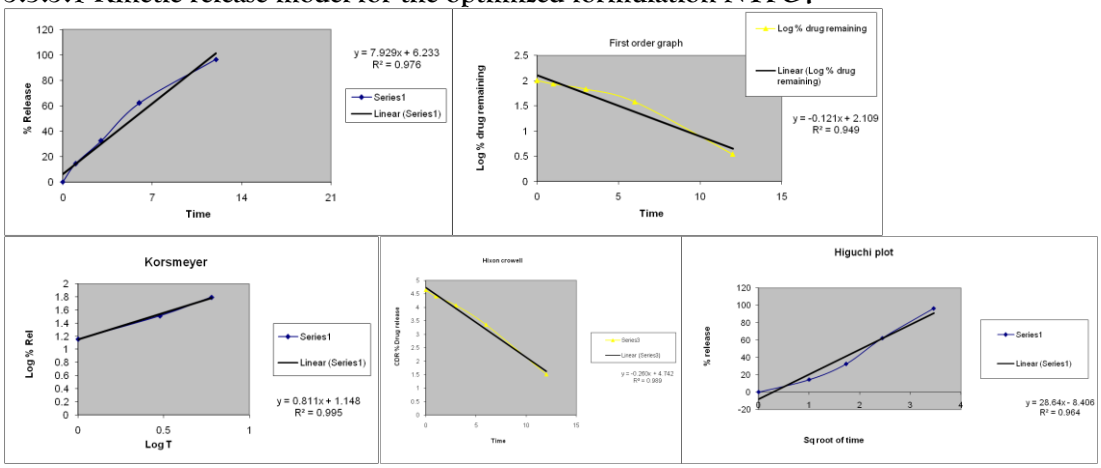
Table 17: represent the % cumulative drug release of the transferosomal gel loaded with drug

S.N.	Time (hrs)	% Cumulative drug release NTFG7
1	0	0
2	0.5	5.78±0.11
3	1	14.36±0.23
4	2	21.22±0.41
5	3	32.47±0.44
6	4	46.34±0.26
7	5	54.28±0.32
8	6	62.24±0.47
9	7	77.47±0.33
10	8	87.15±0.45
11	12	96.54±0.54



Graph 7: % cumulative drug release of NTFG7

3.5.3.1 Kinetic release model for the optimized formulation NTFG7



Graph 8: zero order, First order, Korsmeyer, Hixon crowell, Higuchi kinetic release model for NTFG7

Table 18: represent the kinetic release model regression values of the Transferosomal gel NTFG7

S.N.	Formulation code	Zero Order Release (R^2)	First order kinetic release (R^2)	Higuchi kinetic release (R^2)	Korsmeyer kinetic release (R^2)	Hixon Crowell Kinetic release (R^2)
1	NTFG7	0.976	0.949	0.964	0.995	0.989

The study analysed the release profiles of the NTFG7 formulation, revealing a higher cumulative drug release. Different kinetic models were applied to the in vitro release results, calculating regression coefficients (R2). The drug released from all investigated formulations followed a Korsmeyer kinetic release with a correlation coefficient greater than zero and first-order kinetics. The Korsmeyer-Peppas release exponent data showed a non-Fickian anomalous release with regressions R2= 0.999 for the optimized formulation.

3.6 Stability

Table 19: The stability study of the prepared transferosomal gel during storage at 4°C , 25°C,and 37°C over a period of 3 months

Parameter	Formula number	4°C	25°C	37°C
Texture	NTFG7	smooth	smooth	smooth
Appearance		No change	No change	No change
pH		7.7	7.6	7.69
Viscosity		13568±0.012	13547±0.052	13577±0.021

3.7 Characterization of transferosomal patches

Fig 2: showing the patches of the transferosomal gel containing drug

Table 20: composition of the transdermal patch

Formulation code	Gel quantity	Formulation code	Polymer	Polymer proportion	Solvent	Plasticizer (30% w/w)*
NTFG7 P(containing 5mg drug)	1 gm	NTFG7 P(containing 5mg drug)	PVA : PVP	1 : 1	Water	PG

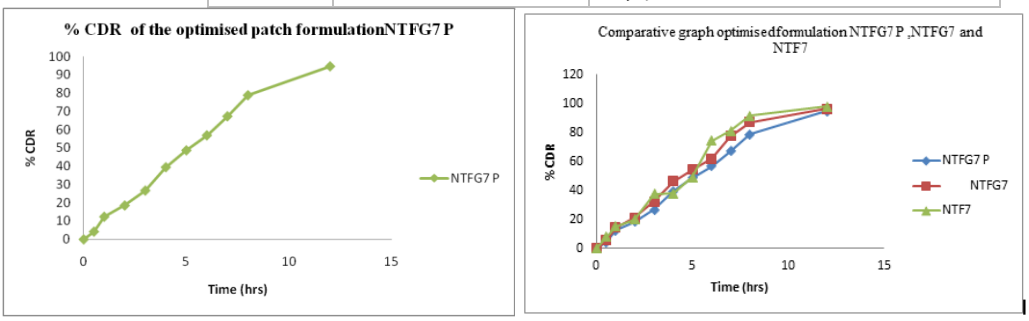
Table 21: appearance, folding endurance, thickness, Tensile strength and Percentage moisture content of the transdermal patch

Formulation code	Parameter	Observation

NTFG7 P(1 gm) (containing 5mg drug)	**Texture**	Smooth
	Color	White color gel incorporated in the patch Bottom layer white and covering layer aluminium foil
	Odor	Odorless
	Folding endurance	46±2.1
	Thickness (mm)	0.13±0.3
	Tensile strength(kg/cm2)	1.2±0.8
	Percentage moisture content	3.11 ± 0.019

Table 22: Skin permeation by Franz diffusion

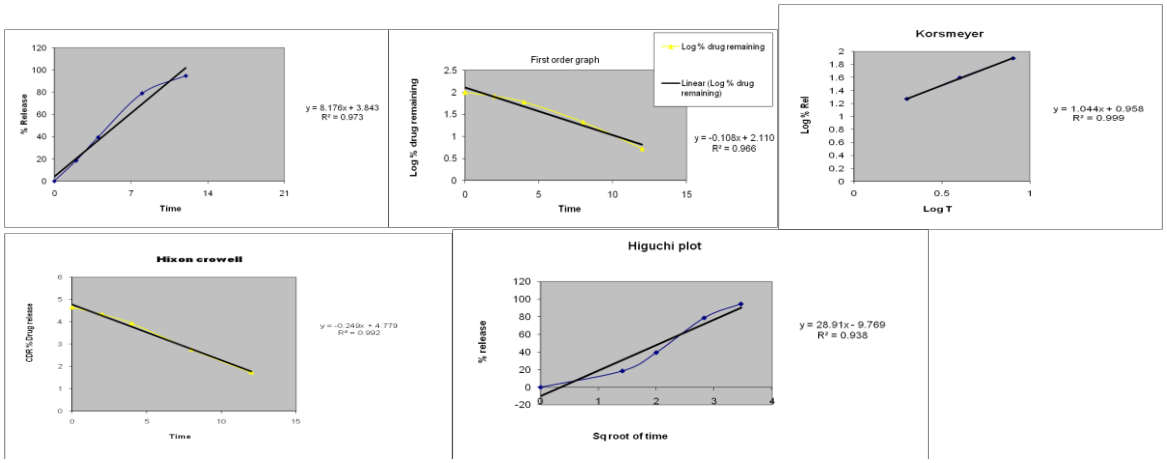
S.N.	Time (hrs)	% Cumulative drug release NTFG7 P
1	0	0
2	0.5	4.25±0.11
3	1	12.41 ±0.25
4	2	18.56±0.52
5	3	26.74±0.57
6	4	39.51±0.21
7	5	48.77±0.12
8	6	56.98±0.75
9	7	67.41±0.31
10	8	78.95±0.19
11	12	94.78±0.51



Graph 9: cumulative drug release of the NTFG7 P and comparative graph of the formulations

Table 23: represent the % cumulative drug release of the transferosomal gel loaded patch NTFG7 P

S.N.	Time (hrs)	NTFG7 P	% drug remaining	Log % drug remaining	Sqr root of time	log T	Log % Rel	CDR % drug remaining
1	0	0	100	2	0	0	0	4.641589
2	0.5	4.25±0.11	95.75	1.981139	0.707107	-0.30103	0.628389	4.574879
3	1	12.41 ±0.25	88	1.944483	1	0	1.079181	4.44796
4	2	18.56±0.52	81.44	1.910838	1.414214	0.30103	1.268578	4.334569
5	3	26.74±0.57	73.26	1.864867	1.732051	0.477121	1.427161	4.184295
6	4	39.51±0.21	60.49	1.781684	2	0.60206	1.596707	3.925496
7	5	48.77±0.12	51.23	1.709524	2.236068	0.69897	1.688153	3.713996
8	6	56.98±0.75	43.02	1.63367	2.44949	0.778151	1.755722	3.503941
9	7	67.41±0.31	32.59	1.513084	2.645751	0.845098	1.828724	3.194195
10	8	78.95±0.19	21.05	1.323252	2.828427	0.90309	1.897352	2.761112
11	12	94.78±0.51	5.22	0.717671	3.464102	1.079181	1.976717	1.734696



Graph 10: zero order, First order, Korsmeyer, Hixon crowell and Higuchi kinetic release model for NTFG7 P

Table 24: represent the kinetic release model regression values of the Transferosomal gel loaded patches NTFG7 P

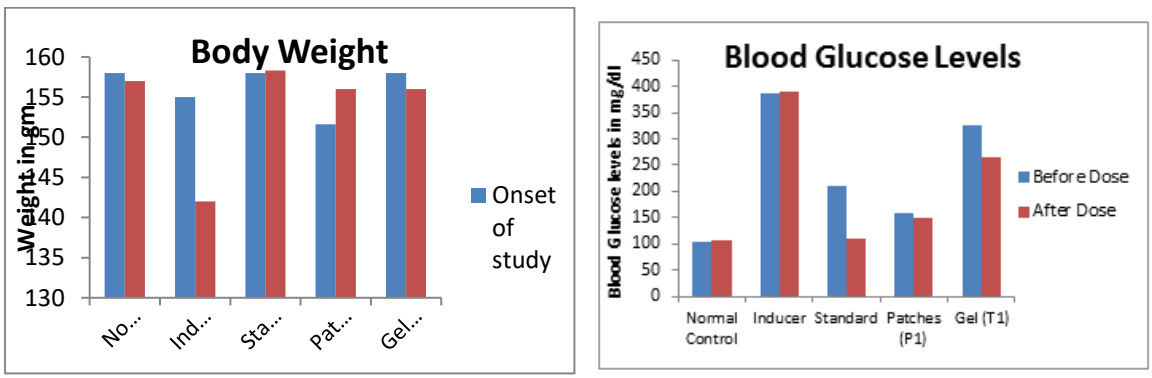
S.N.	Formulation code	Zero Order Release (R^2)	First order kinetic release (R^2)	Higuchi kinetic release (R^2)	Korsmeyer kinetic release (R^2)	Hixon Crowell Kinetic release (R^2)
1	NTFG7 P	0.973	0.966	0.938	0.999	0.992

3.7 In-vivo studies

Table.25 : Body Weight (g) and Antidiabetic Effect of Repaglinide on Blood Glucose Levels (mg/dl)

S. No.	Group	Body Weight at Start (g)	Body Weight at End (g)	Blood Glucose Before Dose (mg/dl)	Blood Glucose After Dose (mg/dl)
I	Normal Control	158 ± 6.08	157 ± 6.08	105.00 ± 4.00	107 ± 4.00
II	Inducer	155 ± 5.01	142 ± 7.21	387.00 ± 7.00	389.00 ± 6.90
III	Standard	158 ± 7.21	158.3 ± 10.40	209.50 ± 7.84*	109.3 ± 2.72
IV	Patches (P1)	151.6 ± 10.40	156 ± 8.71*	159.67 ± 5.03*	148.67 ± 9.14
V	Gel (T1)	158 ± 4.08	156 ± 4.72	327 ± 5.03	266.00 ± 7.84

***Significant compared to inducer group.**

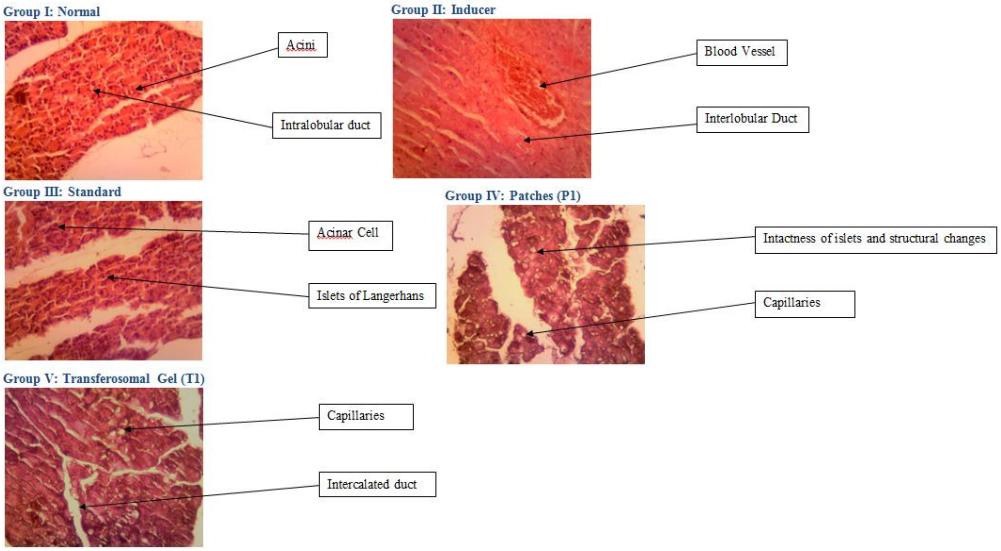


Graph 11and 12 represent the Body weight at starting and End of the study

3.7.2 Anti-diabetic Activity

Values are expressed as MEAN±SD at n=6, One-way ANOVA followed by Bonferroni test, *P<0.050

3.8 Histopathology



4. DISCUSSION

The study successfully developed and optimized a transferosomal gel formulation of repaglinide aimed at enhancing its transdermal delivery and antidiabetic activity. The initial phase involved the preparation of various transferosomal trial formulations using the reverse phase evaporation method. A factorial design approach was employed to identify the optimal levels of phospholipid concentration, surfactant ratio, and sonication time influencing critical parameters—particle size and entrapment efficiency. ANOVA results confirmed the significance of these factors and their interactions, particularly combinations like phospholipid–sonication time (AC) and surfactant–sonication time (BC), which significantly impacted particle size. The optimized formulation, coded as NTF7, demonstrated desirable nanoscale particle size (266.9 nm), low polydispersity index (PDI: 85.6%), and high zeta potential (+33.2 mV), indicating excellent colloidal stability. Notably, entrapment efficiency reached 97.8%, which is critical for ensuring adequate drug loading within the transferosomal vesicles. The sustained in vitro drug release over 12 hours (97.95%) followed Korsmeyer–Peppas and Hixson–Crowell kinetic models, indicating a combination of diffusion-controlled and erosion-based release mechanisms, further supporting its potential for prolonged therapeutic effect. Following gel incorporation (NTFG7) using Carbopol 934, the formulation retained its physicochemical integrity [13]. The gel exhibited a suitable pH (7.7), optimal viscosity (11279 cps), high swelling index (~65%), and excellent homogeneity. The in vitro release from the gel also demonstrated sustained release (96.54% at 12 hours), aligning with the vesicular release behavior and confirming the compatibility between the gel matrix and transferosomes. Additionally, the kinetic modeling for the gel confirmed a Korsmeyer–Peppas-based non-Fickian release, highlighting combined swelling-controlled and diffusion-mediated transport. Further formulation into transdermal patches (NTFG7 P) via solvent casting method showed satisfactory mechanical properties such as tensile strength, folding endurance, and minimal moisture content. Franz diffusion studies indicated effective skin permeation, achieving 94.78% cumulative drug release at 12 hours, again following Korsmeyer–Peppas kinetics. Skin irritation studies established the formulation's dermal safety with minimal erythema or edema, reflected in a low primary dermal irritation index (PDII: 0.33). In vivo, the transferosomal gel and patch significantly lowered blood glucose levels in STZ-HFD-induced diabetic rats. While the standard oral treatment group showed robust glycemic control, the NTFG7 P patch provided a comparable reduction in glucose levels, suggesting that transdermal administration is an effective alternative to conventional oral therapy [12] . Histopathological studies corroborated these findings, showing improvements in pancreatic morphology in treated groups. Overall, the optimized transferosomal gel and patch formulations offer a promising transdermal approach for repaglinide delivery, improving bioavailability, ensuring sustained release, and enhancing therapeutic outcomes in diabetes management.

5. CONCLUSION

The study developed and tested an optimized transferosomal gel of repaglinide for enhanced transdermal delivery and antidiabetic activity. The gel, NTF7, showed ideal physicochemical characteristics, including

a particle size of 266.9 nm, zeta potential of 33.2 mV, and high entrapment efficiency of 97.8%. In vitro drug release studies confirmed sustained release up to 12 hours, suggesting a non-Fickian release mechanism. The gel also displayed desirable physical properties, including homogeneity, pH compatibility, and viscosity. Stability studies showed minimal changes over three months at different storage conditions. The gel offers a promising alternative to oral repaglinide, potentially improving patient compliance and therapeutic outcomes in diabetes management.

REFERENCES

1. Hussain, M., Hafeez, A. & Kushwaha, S. P. Nanoformulation mediated transdermal delivery of anti-diabetic drugs: an updated review. Intell. Pharm. **1**, 192–200 (2023).
2. Kumar, A., Gangwar, R., Ahmad Zargar, A., Kumar, R. & Sharma, A. Prevalence of Diabetes in India: A Review of IDF Diabetes Atlas 10th Edition. Curr. Diabetes Rev. **20**, (2024).
3. Abdelwahab, A. F., Abdelmohymen, A. M., Mostafa, N. M., Magdy, G. & Mazyed, E. A. Formulation of Deformable Liponiosomal Hybrid of Repaglinide: In vitro Characterization and Evaluation of the Anti-Diabetic Effect. Int. J. Nanomedicine (2023) doi:10.2147/IJN.S434840.
4. Gupta, A., Aggarwal, G., Singla, S. & Arora, R. Transfersomes: A novel vesicular carrier for enhanced transdermal delivery of sertraline: Development, characterization, and performance evaluation. Sci. Pharm. (2012) doi:10.3797/scipharm.1208-02.
5. Prausnitz, M. R. & Langer, R. Transdermal drug delivery. Nat. Biotechnol. **26**, 1261–1268 (2008).
6. Cevc, G. & Blume, G. Lipid vesicles penetrate into intact skin owing to the transdermal osmotic gradients and hydration force. BBA - Biomembr. (1992) doi:10.1016/0005-2736(92)90154-E.
7. Camilo, C. J. et al. Lipid vesicles: Applications, principal components and methods used in their formulations. a review. Acta Biologica Colombiana at https://doi.org/10.15446/abc.v25n2.74830 (2020).
8. Parthiban, S., Pharm, G. S.-W. J. & 2016, undefined. Formulation of proliopsomal gel containing Repaglinide for effecctive transdermal drug delivery. wjpr.s3.ap-south-1.amazonaws.com (2016).
9. Abdallah, M. H., Abu Lila, A. S., El-Nahas, H. M. & Ibrahim, T. M. Optimization of Potential Nanoemulgels for Boosting Transdermal Glimepiride Delivery and Upgrading Its Anti-Diabetic Activity. Gels (2023) doi:10.3390/gels9060494.
10. Bnyan, R. et al. Formulation and optimisation of novel transfersomes for sustained release of local anaesthetic. J. Pharm. Pharmacol. **71**, 1508–1519 (2019).
11. Taymouri, S., Hajhashemi, V., Tabbakhian, M. & Torkashvand, M. Preparation and Evaluation of Imatinib Loaded Transfersomal gel for the Treatment of Rheumatoid Arthritis. Iran. J. Pharm. Res. (2021) doi:10.22037/ijpr.2021.115481.15394.
12. Khan, N. U. et al. Anti-diabetic Activity of Brucine in Streptozotocin-Induced Rats: In Silico, in Vitro, and in Vivo Studies. ACS Omega (2022) doi:10.1021/acsomega.2c04977.
13. Malakar, J., Sen, S. O., Nayak, A. K. & Sen, K. K. Formulation, optimization and evaluation of transferosomal gel for transdermal insulin delivery. Saudi Pharm. J. (2012) doi:10.1016/j.jsps.2012.02.001.