CODEN [USA]: IAJPBB ISSN : 2349-7750

INDO AMERICAN JOURNAL OF
PHARMACEUTICAL SCIENCES
SJIF Impact Factor: 7.187

Avalable online at: http://www.iajps.com *Research Article*

FORMULATION AND *INVITRO* EVALUATION OF ETHOSOMAL GELS CONTAINING SAXAGLIPTIN AS THE MODEL DRUG

Sd. Riyaz Hussain*[1], Dr. Pawan kumar[2], Dr. Parwez Alam[3]
[1] Research Scholar, [2] Professor, [3] Professor
[1-3] Department of Pharmaceutical Sciences, Singhania University, Pacheri Bari, Jhunjhunu, Rajasthan – India.

Article Received: March 2021 **Accepted:** March 2021 **Published:** April 2021

Abstract:
The process Touitou et al (2000) was used with little change to prepare various ethosomal formulations with different levels of IPA (20% to 40%) and sonicity. It was simple and reproducible techniques. The prepared and discrete ethosomes have been developed. However, ethosomes are more uniform in size and small, necessary for skin penetration by the process of sonication. When the efficiency of the trap was compared, ethosomes containing 30 percent w/w IPA, which were generated by sonication, displayed the highest value with respect to all others, therefore, with 30 percent w/w IPA as the best formula for all other aspects, ethosomally provided by sonicity was completed. In all formulations, GF6 demonstrated full release of the drugs in 1440 min compared to other formulations. The in vitro release decreased with increased concentrations of polymer and copolymer. Centered on the research findings of the drug release process the drug discharge followed the non-fickian diffusion mechanism by formulations and followed the first order kinetics.

Key words: *Ethosomes, Sonication, Transdermal, Entrapment, Stability.*

Corresponding author:
Sd. Riyaz Hussain,
E mail: sdriyazhussain.phd@gmail.com
Contact number: 9014412525

QR code

Please cite this article in press Sd. Riyaz Hussain et al., ***Formulation and Invitro Evaluation of Ethosomal Gels Containing Saxagliptin As the Model Drug..,*** *Indo Am. J. P. Sci, 2021; 08(04).*

INTRODUCTION:

A managed drug supply system has been developed to track drug delivery speeds, maintain therapeutic duration and/or tissue delivery. drug delivery systems were developed.

A convenient four types of controlled drug delivery or modified drug deliveries are divided.

1) Delayed release.
2) Sustained release.
3) Site-specific targeting.
4) Receptor targeting.

Controlled delivery can be described more precisely as[1]:

(1) Sustained drug action by sustaining a comparatively stable, efficient body level of prescription drugs with concomitant reduction of undesirable side effects.

2) Spatial location of controlled discharge systems adjacent to or in the tissue of illness, localized drug activity.

3) Targeted action of drugs to supply the medication to a specific target cell type using carriers or chemical derivatives.

4) Have a prescription release mechanism based physiologically and therapically. In other words, physiological and clinical requirements of the body decide the volume and the rate of release of drugs"[2].

"Usually, a regulated method for the delivery of drugs is built in particular to supply the medication. Blood levels are kept stable and efficient for a time when the machine is distributing medication. Regulated drug entry typically leads to significantly constant active ingredient blood levels compared to uncontrolled fluctuations when several doses of rapid release are administered to patients for the traditional dosage types.

Currently, an oral route is the most common method of drug delivery. While the benefit of ease of administration is noteworthy, it also has significant disadvantages, namely poor bioavailability, due to metabolism in the first place and a propensity to generate high and low blood spikes, leading to a need for high or regular dosing, which can be both prohibitive as well as inconvenient"[2].

"The development of an emergent drug delivery system, enhancing the therapeutic effectiveness and drug protection, by making spatial and temporary placements within the body more reliable (i.e. precise site), reduces both size and number of doses, is required in order to solve these problems. New drug delivery schemes are also necessary in order to supply the site without any major immunosupply or biological inactivation with novel genetically engineered medicinal products (i.e. peptides, proteins). In addition to these benefits, pharmaceutical firms understand that the idea and technologies of a managed drug delivery system and the cost associated with getting new drug companies into the market are feasible for repatenting successable drugs. Transdermal delivery of medicinal substances through the skin for systemic effects was one of the most commonly used methods[2].

"The present study is equipped with two different polymer combinations: E RS100 and HPMC E 15, E RL 100 with HPMC E 15, to establish suitable matrix transdermal drug delivery systems for Ketorolac. The acrylic acid matrices E RL100 and E RS 100 are used to manufacture drug polymer matrix films, which are stated to be compatible with several drugs. Penetration changes that can help increase drug permeation partitioning[3]. Various D-Limonene[4] penetration enhancers, Oleic[5] and their effects on drug permeation, were used at various concentrations in the study" [4].

DRUG DELIVERY SYSTEMS OF TRANSDERMAL

"Transdermal drug delivery systems are topical medicines in the form of patches delivering medicines for a predetermined systemic impact and controlled pace. - And controlled rate.

A transdermal drug delivery system, 'which can be of an active or passive nature, is an alternate medication path. These devices can be administered through the skin barrier to pharmaceutical products. A medicine is administered to the skin of a patch at a reasonably high dose over a long period of time. The drug reaches the bloodstream directly through the skin through a dissemination process. The substance is still diffused in the blood" for a long period of time because the patch is high and the blood concentrations are low, and the drug persists in blood.

Advantages [4-8]

The transdermal route is an "interesting option," since it is a realistic and secure transdermal route. The beneficial characteristics of drug delivery through the skin are systemic consequences:

1. "Avoidance of first pass metabolism.
2. Avoidance of gastro intestinal incompatibility.
3. Predictable and extended duration of activity.
4. Minimizing undesirable side effects.

5. Provides utilization of drugs with short biological half-lives.
6. Improving physiological and pharmacological response.
7. Avoiding the fluctuation in drug levels.
8. Avoiding inter and intra patient variations.
9. Maintain plasma concentration of potent drugs.
10. Termination of therapy is easy at any point of time.
11. Greater patient compliance due to elimination of multiple dosing profile.
12. Ability to deliver drug more selectively to a specific site.
13. Provide suitability for self-administration and enhance" therapeutic efficacy.

Mechanism of penetration:

"While speculation still concerns the exact process of medicinal supply by ethosomes, a combination of processes probably contributes to the improvement of the effect. The multi-layer stratum corneum lipid is tightly packaged and highly conformational at physiological temperature. The high ethanol content makes ethosomes special, since ethanol is known to disrupt the organisation of the bilayer of the skin lipid. It thus enables the vesicles to penetrate a stratum corneum when integrated into a vesicle's membrane. The lipid membrane is packed less securely than traditional vesicles due to its high ethanol content, but has an equal strength that allows a structure that can be more malt-free and allows more mobility, squeezing through small squares, such as openings created to disrupt the lipid stratum corneum"[9].

"Ethanol interacts with lipid molecules in the hard group polar region, which decreases the stiffness of the corneal stratum lipids and increases their fluidity. The intercalation of ethanol into the environment of the polar head group will lead to increased membrane permeability. The ethosome itself can interact with the stratum corneal barrier as well as the effect of ethanol upon the layered corneal structure"[10].

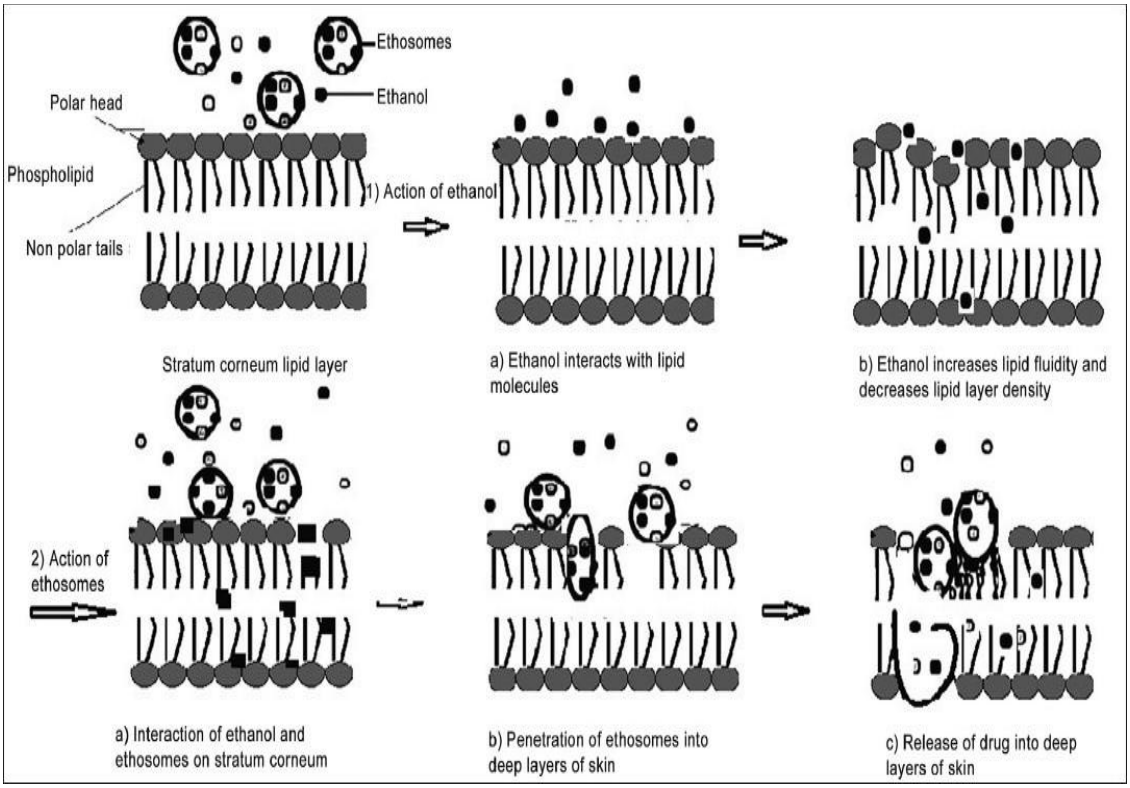

Figure 01: Proposed model of skin delivery ethosomal system.

The "interdigital and malleable vesicle of ethosomes will trace paths in the distorted stratum. The higher positive zeta-potential of the drugs can increase the skin attachment of the vesicles in the case of drug encapsulation ethosomes. The ethosomal system was shown to be highly efficient carriers for increased drug use in skin while encapsulated medication in classic liposomes remained primarily at the skin's surface. This method is a promising candidate for transdermal supply of the medication because of its efficient supply along with the long-term stability of the ethosomes[11]."

Planning Preparation

"The Touitou4 et al. reports that the ethosomal system can be produced with soybeanphosphotidyl choline 2 – 5 percent, 20 – 50 percent w/w, and medications and waters, at 100 percent W/w. "Ethosomal formulation and preparation is the result of Touitou *et al*. Ethanol dissolves 90 and medication for the preparation of ethosomesphospholipid. As a slender stream with a steady mix at 700 rpm, double distilled water was added slowly into a well settled bottle. For another 5 minutes, mixing has been continued. During the preparation, the machine was kept at 30 °C and then stored cold"[12].

Table 01: Different additives used in ethosomal formulation.

Class	Example	Uses
Phospholipid	"Soya phosphatidyl choline Egg phosphatidyl choline Dipalmityl phosphatidyl choline Distearyl phosphatidyl choline" [11]	Vesicles forming component
Polyglycol	Propylene glycol Transcutol RTM	As a skin penetration enhancer
Alcohol	Ethanol Isopropyl alcohol	"For providing the softness for vesicle membraneAs a penetration enhancer" [12]
Cholesterol	Cholesterol	For providing the stability to vesicle membrane
Dye	"Rhodamine-123 Rhodamine red Fluorescene Isothiocynate (FITC) 6- Carboxy fluorescence" [12]	For characterization study
Vehicle	Carbopol 934	As a gel former

SAXAGLIPTIN:

The new class of drugs that inhibit dipeptidyl peptidase-4 (DPP-4) orally active Saxagliptin, an anti-diabetic inhibitor for the treatment of type 2 diabetes is dipeptidyl peptidase-4 (DPP-4). The inhibitors of DPP-4 are a class of compounds that influence the effect of so-called incretins on natural hormones in the body. Incretins lower blood sugar by increasing the body's intake of sugar, primarily by increasing pancreas insulin output and reducing liver sugar production. DPP-4 is a membrane-associated peptidase present in many tissues, lymphocytes, and plasma. [Bristol-Myers Squibb Press Release]. The DPP-4 has two main pathways, an enzyme activity, and one where DPP-4 binds the adenosine deaminase, which, through dimerization, conveys intracellular signals. Saxagliptin forms the reversible, histidine-aided covalent bond of DPP-4 hydroxy oxygen between its nitrile group and S630. DPP-4 inhaled raises active blood glucagon levels like peptide 1 (GLP 1), which inhibits pancreatic alpha cell development of glucagon and improves pancreatic beta cell insulin production.

AIM AND OBJECTIVE:

To prepare and evaluate Saxagliptin ethosomes containing different concentration of ethanol and phospholipids by sonication for size reduction of vesicles.

The designated ethosomes of Saxagliptin are characterized by

- Size and shape.
- Entrapment effectiveness.
- Study of release.

The influence of sonication was also investigated on the characteristics of the ethosomes of Saxagliptin.

PLAN OF WORK:

- To formulate ethosomal gel.
- To characterize the prepared formulation using cold method.
- To carry out different criteria of assessment, including vesicular form and surface morphology, vesicular duration, drug quality, efficiency of trapping.
- To carry out *in vitro* drug diffusion study of ethosomal gel.

- **Preformulation studies:**
  - API Characterization.
  - Studies into solubility.
  - Compatibility reports of Drug Excipients.
- **Construction of Calibration curve.**
- **Formulation Development.**
- **Characterization of ethosomal gel.**
- Size and shape analysis
- Entrapment efficiency
- pH.
- Spreadability.
- Drug content and content uniformity.
- Drug release study.
- Invitro release kinetics.
- **Stability studies** (at 30ºC / 75 % RH and 40ºC / 75 % RH).

MATERIALS AND METHODS:

Glibenclamide was obtained as gift sample from Chandra labs pvt. Ltd, Hyderabad, Soya lecithin Mylan Chemicals, Propylene glycol Avantor chemicals, Methanol, Mark chemical Cholesterol, Virat lab (Mumbai), Carbopol-940 Srini Chemicals, Triethanol amine, Avantor chemicals, Ultrapure water Mark chemical reagents etc.

METHODOLOGY:

PRE-FORMULATION STUDIES:[13]

"Preformulation testing was an investigation of physical and chemical properties of a drug substance alone and when combined with excipients. It was the first step in the rational development of dosage forms.

PREPARATION OF SAXAGLIPTIN ETHOSOMES (BY COLD METHOD):

Preparation of saxagliptin ethosomes was followed by method suggested byTouitou *et al*.,with little modification.[7]

The "ethosomal system of Saxagliptin comprised of 2-6 % phospholipids, 20-40 % isopropyl alcohol, 10 % of propylene glycol,0.005g of cholesterol and aqueous phase to 100 % w/w. Saxagliptin 0.05g, was dissolved in IPA in a covered vessel at room temperature by vigorous stirring. Propylene glycol was added during stirring. This mixture was heated to 30^0 in a separate vessel and was added to the mixture drop wise in the center of the vessel, which was stirred for 5min at 700rpm in a covered vessel the vesicle size of ethosomal formulation can be decreased to desire extend using sonication or extrusion method. Finally, the formulation is stored under refrigeration[31].Ethosomes were formed spontaneously by the process"[14].

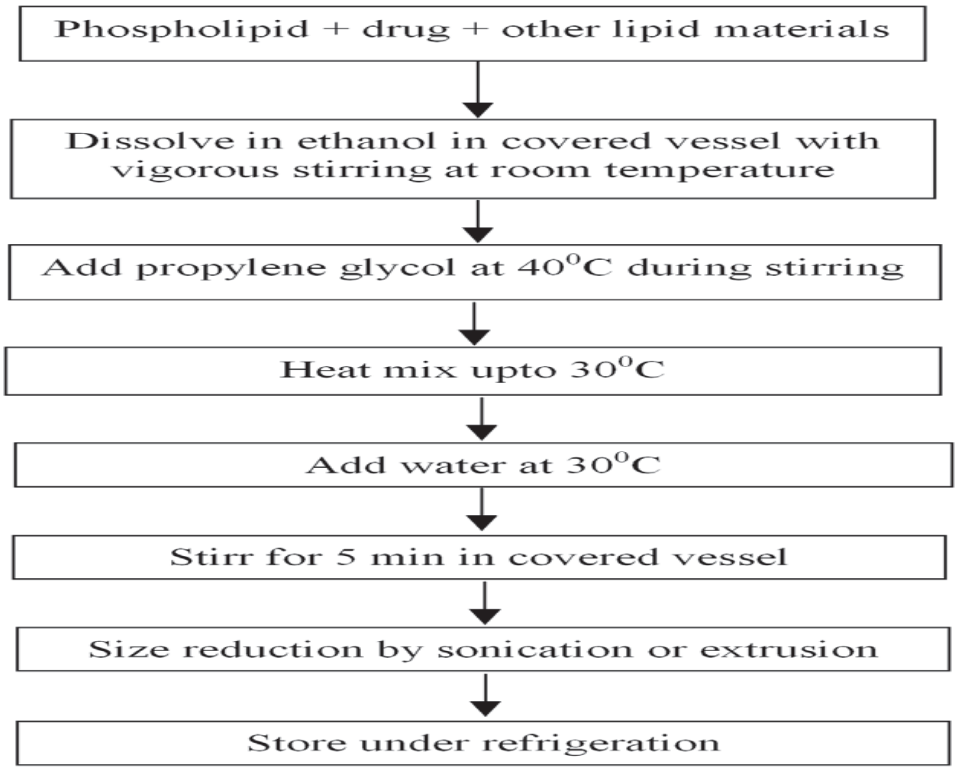

Figure 02: Cold method for the preparation of ethosomes.

Table 02: Composition of different ethosomal formulations.

Ethosomal formulation	Lecithin (Soya lecithin%)	IPA (%)	Propylene glycol (%)	Saxagliptin (g)	Cholesterol (g)	Water
GF_1	2ml	20ml	5ml	0.05g	0.005g	100ml
GF_2	4ml	20ml	5ml	0.05g	0.005g	100ml
GF_3	6ml	20ml	5ml	0.05g	0.005g	100ml
GF_4	2ml	30ml	5ml	0.05g	0.005g	100ml
GF_5	4ml	30ml	5ml	0.05g	0.005g	100ml
GF_6	6ml	30ml	5ml	0.05g	0.005g	100ml
GF_7	2ml	40ml	5ml	0.05g	0.005g	100ml
GF8	4ml	40ml	5ml	0.05g	0.005g	100ml
GF9	6ml	40ml	5ml	0.05g	0.005g	100ml

Preparation of Saxagliptin ethosomal gel:

The best achieved suspension of ethosomal vesicles was introduced into carbopol gel (1%, 1.5%, 2% w/w). The specified volume of carbopol 934 powder was slowly applied to ultrapure water and held for 20 minutes at 1000c. It was applied dropwise to triethanolamine. Sufficient quantity of formula (GF-2) comprising saxagliptin (1.5 percent w/w was then introduced into gel-base. Water q.s was added with other continuous stirring formulations before homogeneous formulation was obtained (G-1, G-2 and G-3). Free saxagliptin containing gel was prepared using 1.5 by similar process.

Table 03: Composition of different ethosomal gel formulation.

Gel formulation	Saxagliptinethosomal suspension(ml)	Carbopol 940(%)	Triethanolamine(ml)	Phosphate buffer (pH 6.8)
G-1	20ml	0.5	0.5	q.s
G-2	20ml	1	0.5	q.s
G-3	20ml	1.5	0.5	q.s

CHARACTERIZATION OF ETHOSOMES:
SIZE AND SHAPE ANALYSIS:
To determine the average size of the ethosomes, microscopic analysis was conducted. "In order to observe individual vesicles, a sample of ethosomes was sufficiently diluted with distilled water and a drop of diluted suspension was mounted on a glass slide covered with a cover slip and examined under a microscope (magnification 15 X 45 X). The diameters of 150 vesicles were randomly calculated using a calibrated stage micrometer eyepiece micrometer. The method used to measure the average diameter was"[15].

Average diameter = nd / n

n = number of vesicles
d = diameter of vesicles

The vesicle size was diminished by sonication. Although the vesicular dimension of these vesicles could not be measured with a magnification of 15 X 45 X using a microscopic process. Sonicated vesicles were then examined under a special microscope linked to software and photomicrographs were taken under magnifications of 400 and 800. For size analysis, more selected photomicrographs were analyzed using special "particle size analysis" software developed by BIOVIS. This particular programme functions on photomicrograph images of normal dimensional measurements.

SCANNING ELECTRON MICROSCOPY:
Determination of surface morphology (roundness, smoothness and formation of aggregates) of ethosomal gel with polymer was carried out by scanning electron microscopy (SEM).

***IN-VITRO* RELEASE STUDIES:**
DRUG RELEASE STUDY FROM DIALYSIS MEMBRANE OF SAXAGLIPTIN:
Saxagliptin skin permeation from ethosomal formulation was tested using open-ended diffusion cells primarily developed for literacy in our laboratory. The successful permeation area was 2.4 cm and 200 ml for the diffusion cell and receptor cell number, respectively. At 37 ± 0.5 °C.2 the temperature was preserved.

The "receptor compartment held 200 ml of pH 6.8 buffer and was continuously stirring at 100 rpm by means of a magnetic stirrer. Between the donor and the receptor compartments, ready dialysis was placed. The ethosomal formulation was added to the dialysis membrane and the material of the diffusion cell was placed under continuous stirring, during which 5 ml of samples were extracted at fixed time intervals from the receptor compartment of the diffusion cell and examined by spectrometric approach at 278 nm following sufficient dilution. The receptor process was instantly replenished with a fresh pH 6.8 buffer of equivalent length. For opioid release trials, triplicate experiments were performed"[16].

***In-vitro* release kinetics:(Harris shoaib *et al.*, 2006)[17]**
A series of "kinetic models were used to describe the in vitro release effects and analyse the way that they worked out.

The "results of the in vitro release profile obtained for all the formulations are defined in the following three data treatment modes: all release regulation, low and high release scenarios:

- A zero-order kinetic model of cumulative drug release percentage versus time.
- We plotted the course of first-order-Log cumulative percent for our therapy versus time.
- Cumulative percent drug release d versus square root of time, Hikuichi's model.
- The Korsmeyer Equation/Peppa Model- Log cumulative drug release percentage versus log time"[18].

Table 04: Developing a new diffusion model for cylindrical form.

S.No	Diffusion	Exponent (n) Overall solute Diffusion mechanism
1.	0.45	Fickian diffusion
2.	0.45<n<0.89	Anomalous (non-Fickian) diffusion
3.	0.89	Case-IItransport
4.	n>0.89	Super case-IItransport"[19]

STABILITY STUDIES:

The stability analysis for sexagliptin ethosomal preparation was performed at two separate temperatures, i.e. the cooling temperature (4 ± 2 °C) at room temperature (27 ± 2 °C) for 8 weeks (as per ICH guidelines). The formulation was subjected to a stability analysis and placed in a borosilicate bottle to prevent any contact between the ethosomal preparation and the container glass that could impact the findings.

- **In-vitro stability release study:**

Drug stability and vesicle stability are the key determinants of formulation stability, experiments have been carried out to determine the overall drug content at room temperature (27 ± 2 ° C) and cooling temperature (4 ± 2 ° C). Samples were collected every 2 weeks and absorption was seen at 229 nm in the U.V spectrometer.

RESULTS AND DISCUSSION:

PREFORMULATION STUDIES:

- **Description:**

These tests were performed as per the procedure and the results were illustrated in the following table:

Table 05: Table showing the description of Saxagliptin (API).

Test	Description
Colour	White crystalline powder

Result: The results were decided according to requirements**.**

- **Solubility:**

These studies were carried out carried out in conjunction with the protocol and the findings are shown in the following table.

Table 06: Table showing Saxagliptin Solubility (API) in different solvents.

SOLVENTS	SOLUBILITY
Water	Sparingly soluble
pH6.8 Phosphate buffer	Soluble
Methanol	Freely Soluble
Chloroform	Slightly Soluble
Acetonitrile	Freely Soluble

- **Melting Point:**

This test is performed as per procedure and the result was illustrated in the following table 07.

Table 07: Table showing the melting point of API's.

Material	Melting Point
Saxagliptin	103-107^{0}c

SAXAGLIPTIN LINEARIZATION ORDER IN PH 6.8 PHOSPHATE BUFFER:

Table 08: Calibration Curve Data of Saxagliptin.

CONCENTRATION (µg /ml)	ABSORBANCE
0	0
5	0.145
10	0.326
15	0.507
20	0.667
25	0.816

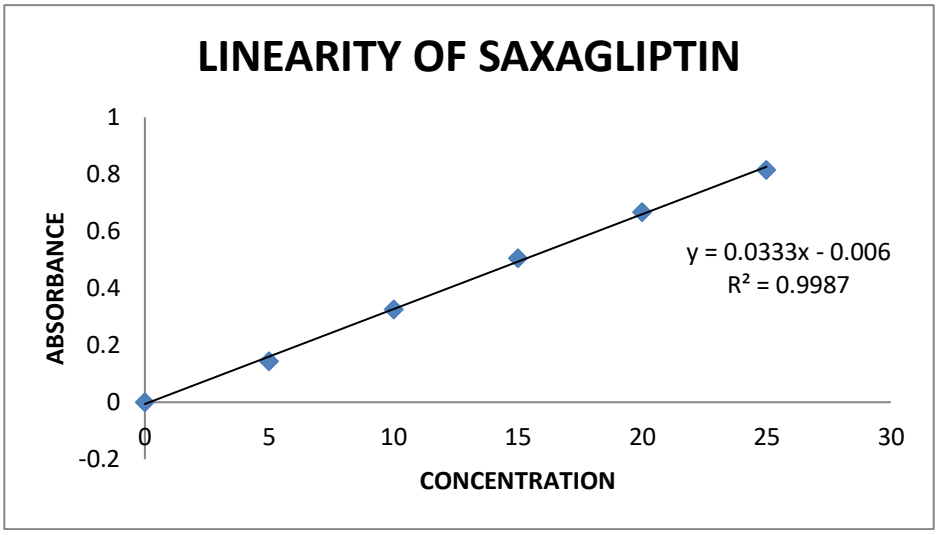

Figure 03 : Calibration curve plot of Saxagliptin.

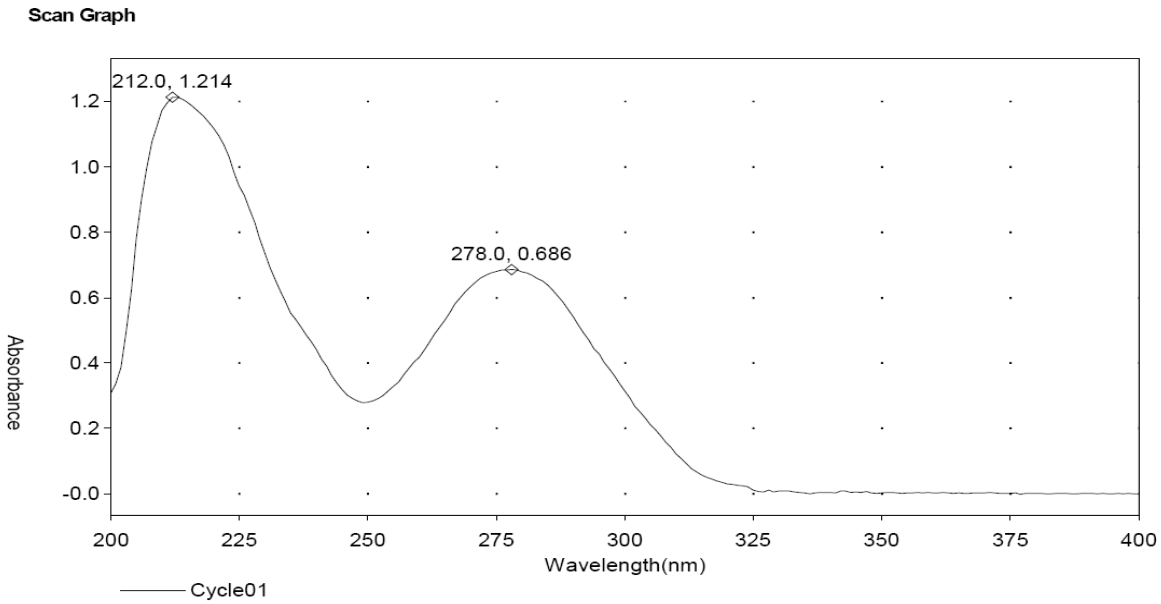

Figure 04: Spectrum of saxagliptin at 278nm by UV.

FT-IR STUDIES: In functional peaks and in functional classes, IRspectra were compared and checked for improvement. It is evident from the continuum that there is no correlation between the selected carriers, drugs and mixtures. Consequently, without any mutual interaction, the chosen carrier was found to be compatible with those selected carriers.

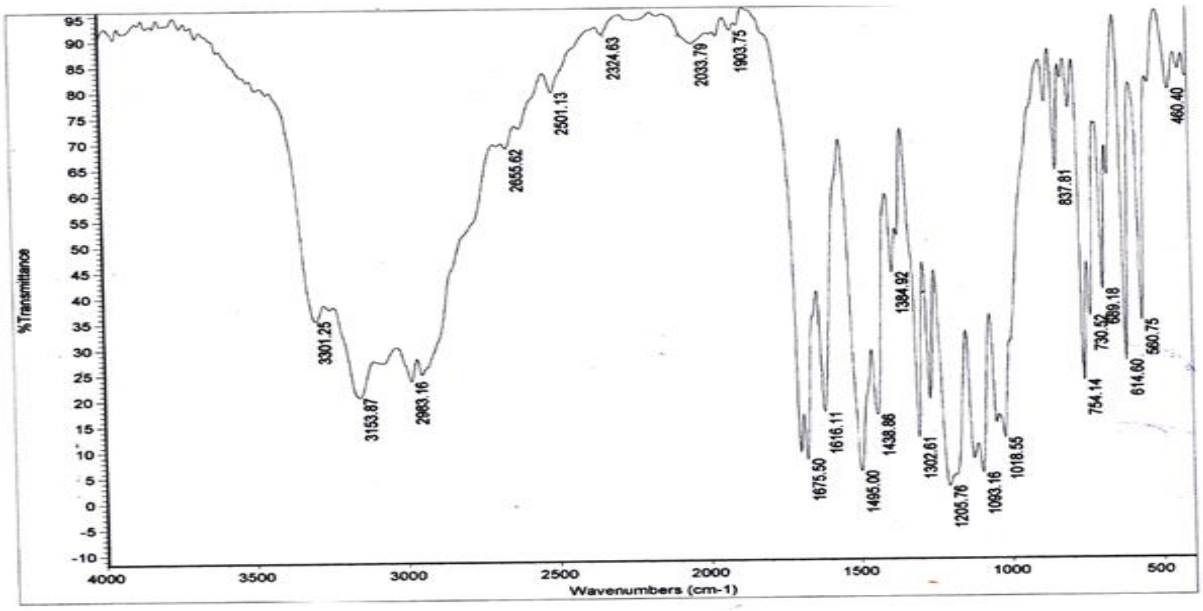

Figure 05: FTIR Spectra of SAXAGLIPTIN pure drug.

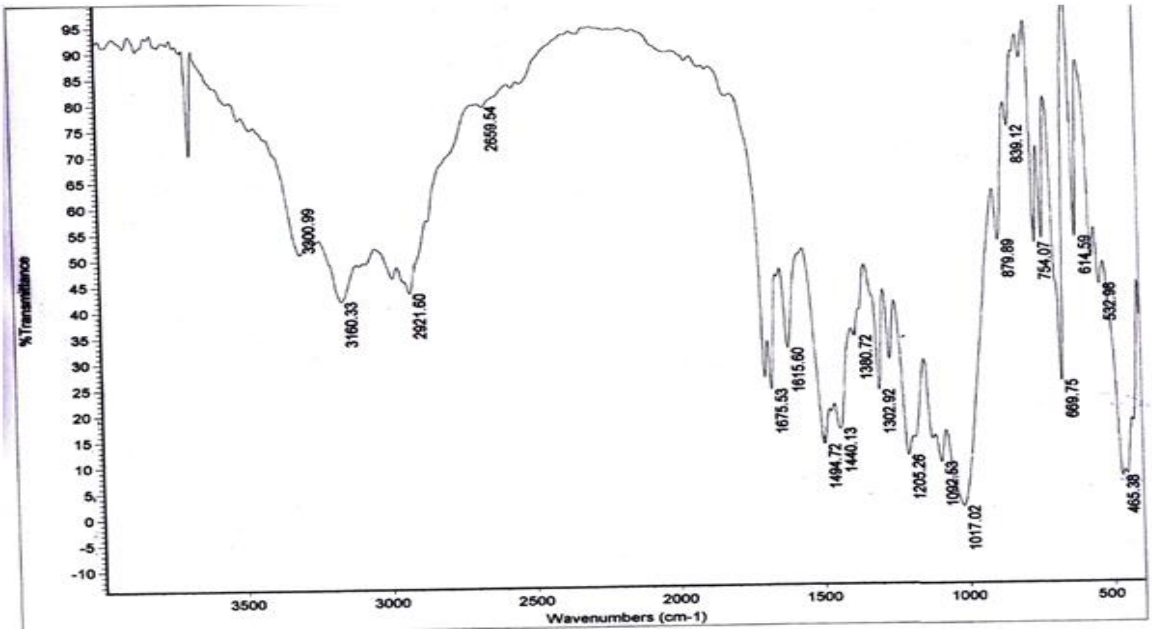

Figure 06: FTIR Spectra of optimized formulation.

FTIR experiments were performed to illustrate the safety of the drug with several excipients.

SIZE AND SHAPE ANALYSIS:
Microscopical inspection was conducted under distinct magnification to imagine the vesicular shape, lamellarity and the size of ethosomal preparations.

SCANNING ELECTRON MICROSCOPE (SEM):

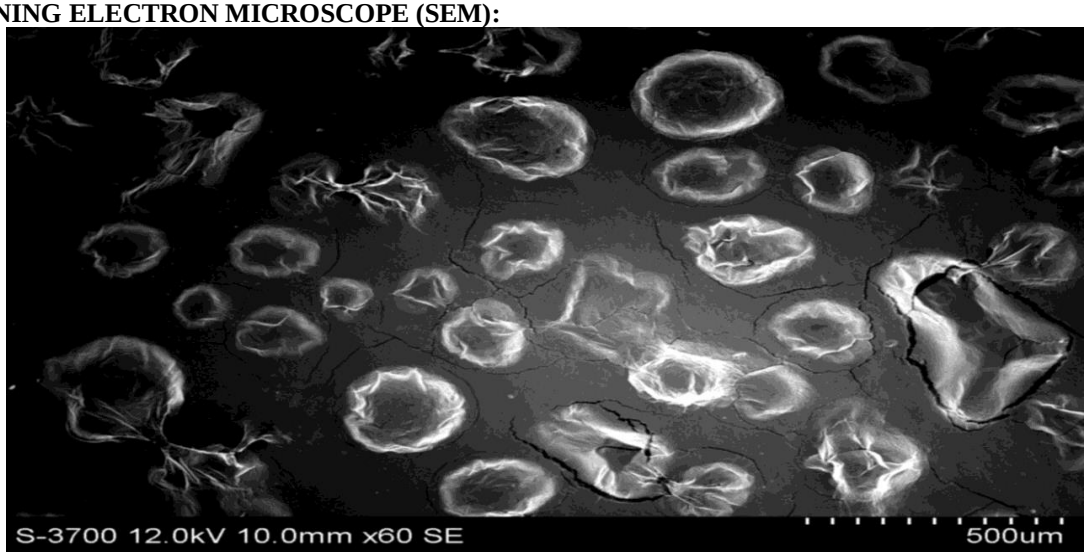

Figure 07: Scanning electron microscope image.

ETHOSOMAL GEL ENTRAPMENT EFFICIENCY:
Ultracentrifugation explored the potential of vesicles for drug trapping until the existence of bilayer vesicles was established in the ethosomal method. The method used to extract drug-containing vesicles and the untrapped or free drug used for the evaluation of trapping performance was ultra-centrifugation.

Table 09: Drug entrapment efficiency of Saxagliptin.

Formulation code	Entrapment efficiency (%)
GF1	71.8
GF2	77.6
GF3	78.8
GF4	77.9
GF5	81.27
GF6	85.42
GF7	82.46
GF8	82.17
GF9	86.67

As calculated by ultracentrifugation, the overall capture efficiency of ethosomal vesicles was 86.67 percent for saxagliptin formulations containing 40 percent IPA (GF9). As the IPA concentration increased from 20 percent to 40 percent w/w, the capture efficiency increased and the ethanol concentration increased more (> 40 percent w/w). Four percent phospholipid trapping results also indicate optimum trapping efficiency concentrations and, subsequently, higher or decreased phospholipid concentrations lower trapping efficiency. The findings further validate these findings of Jain NK *et al.*

The efficacy of ethosomal formulations in capture is substantially different and is stated in the given table. The potential decrease in vesicle size may be due to an improvement in entrapment performance. The negative effect on the vesicle, which is greater in size during ultra-centrifugation. The more uniform lamellae, smaller vesicles and uniform scale are given by Sonication which can also contribute to improved stabilization during ultracentrifuge and lower vesicular disruption.

EVALUATION OF ETHOSOMAL GEL:

Table 10: Organoleptic characteristics of ethosomal gel.

Organoleptic Characteristics:	Color: brownish-yellow Greasiness: Non greasy Grittiness: Free from grittiness Ease of application: Easily/smoothly applied Skin irritation: No skin irritation
Wash ability:	Easily washable without leaving any residue on the surface of the skin.

Table 11: Physical appearance:

Formulation code	Color	Homogeneity	Consistency	Phase separation
GF1	**Creamy white**	**Homogenous**	**Smooth**	**-**
GF2	**Creamy white**	**Homogenous**	**Smooth**	**-**
GF3	**Creamy white**	**Homogenous**	**Smooth**	**-**
GF4	**Creamy white**	**Homogenous**	**Smooth**	**-**
GF5	**Creamy white**	**Homogenous**	**Smooth**	**-**
GF6	**Creamy white**	**Homogenous**	**Smooth**	**-**
GF7	**Creamy white**	**Homogenous**	**Smooth**	**-**
GF8	**Creamy white**	**Homogenous**	**Smooth**	**-**
GF8	**Creamy white**	**Homogenous**	**Smooth**	**-**

Table 12: Spreadability studies:

S.no	Formulation code	Spreadability (g.cm/sec)
1	**GF1**	**14.28**
2	**GF2**	**16.42**
3	**GF3**	**13.74**
4	**GF4**	**15.26**
5	**GF5**	**16.67**
6	**GF6**	**17.81**
7	**GF7**	**18.54**
8	**GF8**	**15.49**
9	**GF9**	**18.76**

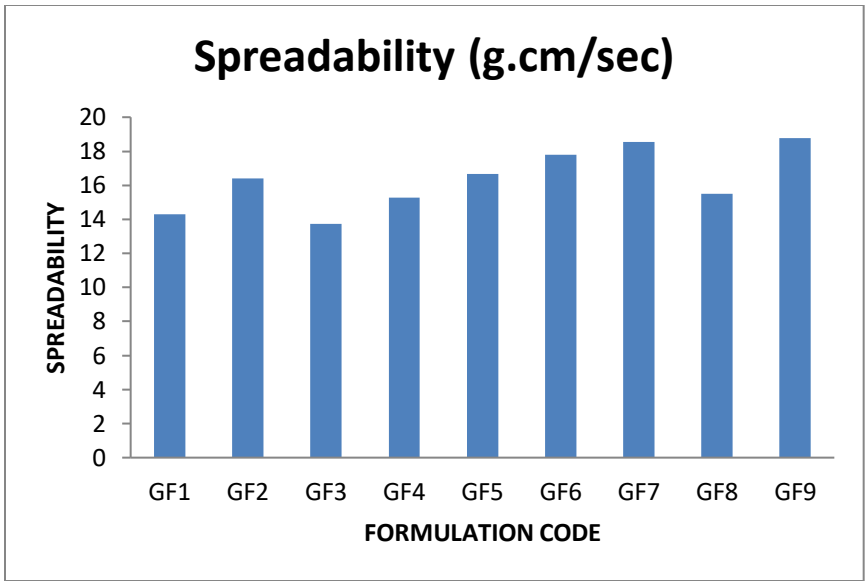

Figure 08: Spreadability graph for GF1-GF9.

Table 13: Rheological studies (for 10rpm spindle 6).

S.no	Formulation code	Viscosity (cps)
1	GF1	1815
2	GF2	1824
3	GF3	2046
4	GF4	2132
5	GF5	1674
6	GF6	1679
7	GF7	1670
8	GF8	1674
9	GF9	1672

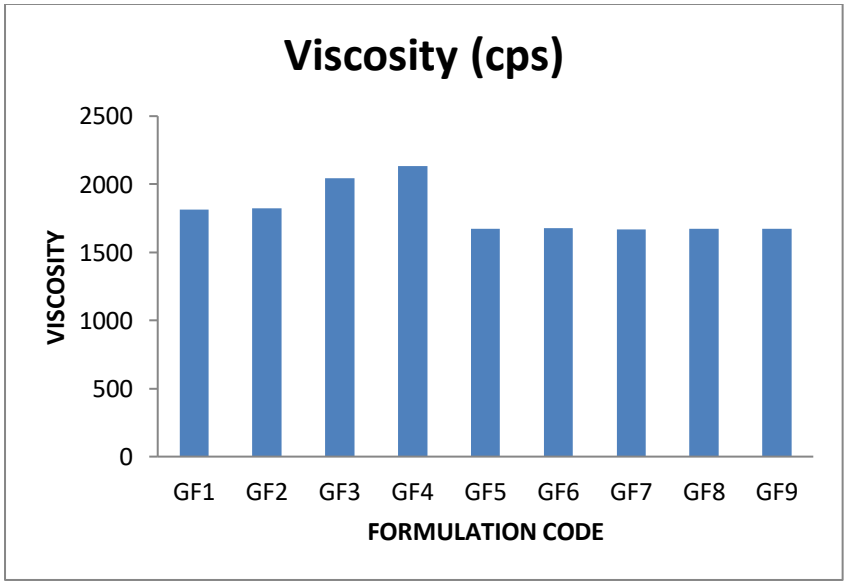

Figure 09: Viscosity Graph for GF1-GF9.

The pH of gels was measured by using electrode based digital pH meter.

P^H measurements of Ethosomal gel.

The pH values for all formulations were in the range of 5.9 to 6.6

Table 14: P^H measurements.

Formulation code	pH
GF1	5.9
GF2	6.3
GF3	6.8
GF4	5.9
GF5	6.4
GF6	6.6
GF7	6.5
GF8	6.6
GF9	6.8

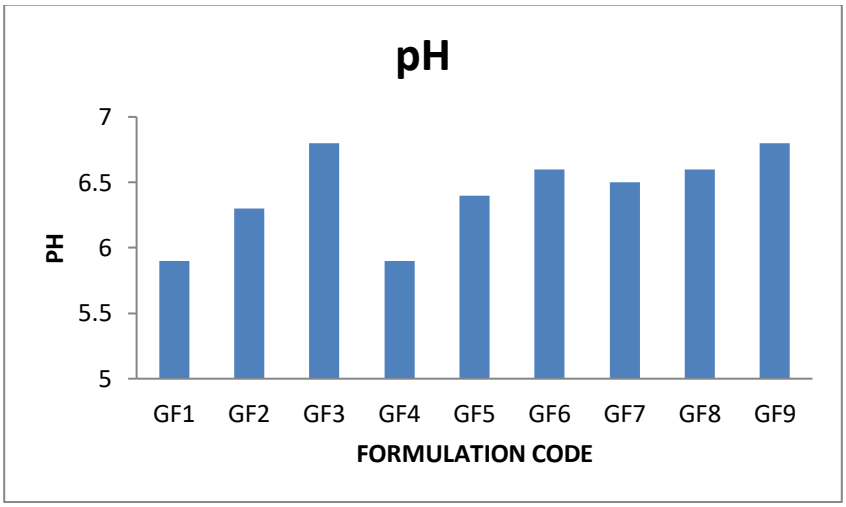

Figure 10: Surface PH graph for GF1-GF9.

Drug content and content uniformity.

Table 15: Drug content for Saxagliptin.

Formulation code	Drug content (%)
GF1	95.4
GF2	97.8
GF3	98.1
GF4	96.9
GF5	97.7
F6	98.6
GF7	94.7
GF8	96.4
GF9	98.2

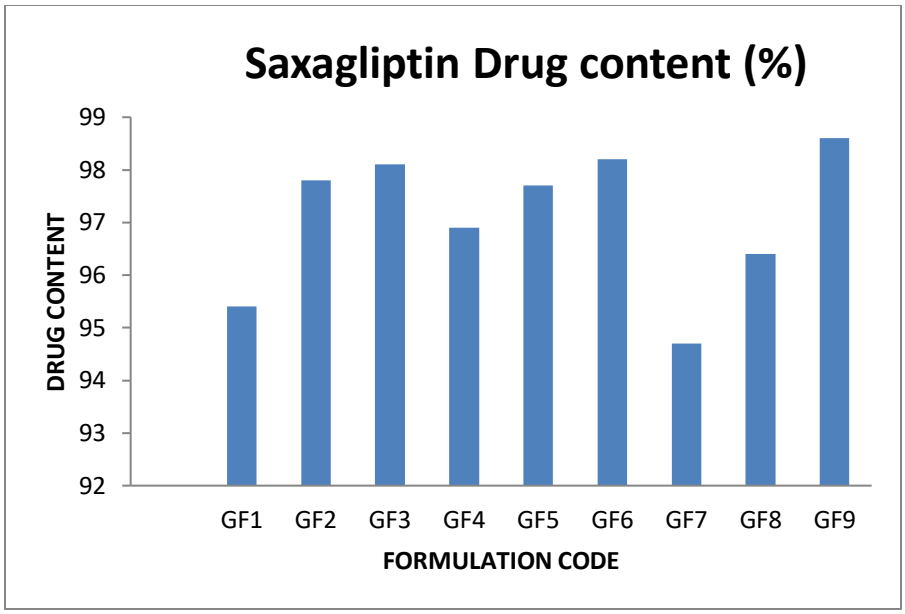

Figure 11: Drug Content Graph for GF1-GF9.

Vesicle size: Vesicle size of the ethosome was measured by zeta sizer. The size of the vesicle was found to be 430nm with a pdi of 0.168

Resultssize(d.nm):	**%Intensity**	**Width(d.nm)**		
Z- AVERAGE (D.NM): 430	**Peak1: 426.5**	**100**	**104.2**	
PDI: 0.168	**Peak2 :--**	**--**	**--**	
Intercept: 0.96	**Peak3 :--**	**--**	**--**	
Result quality: good				

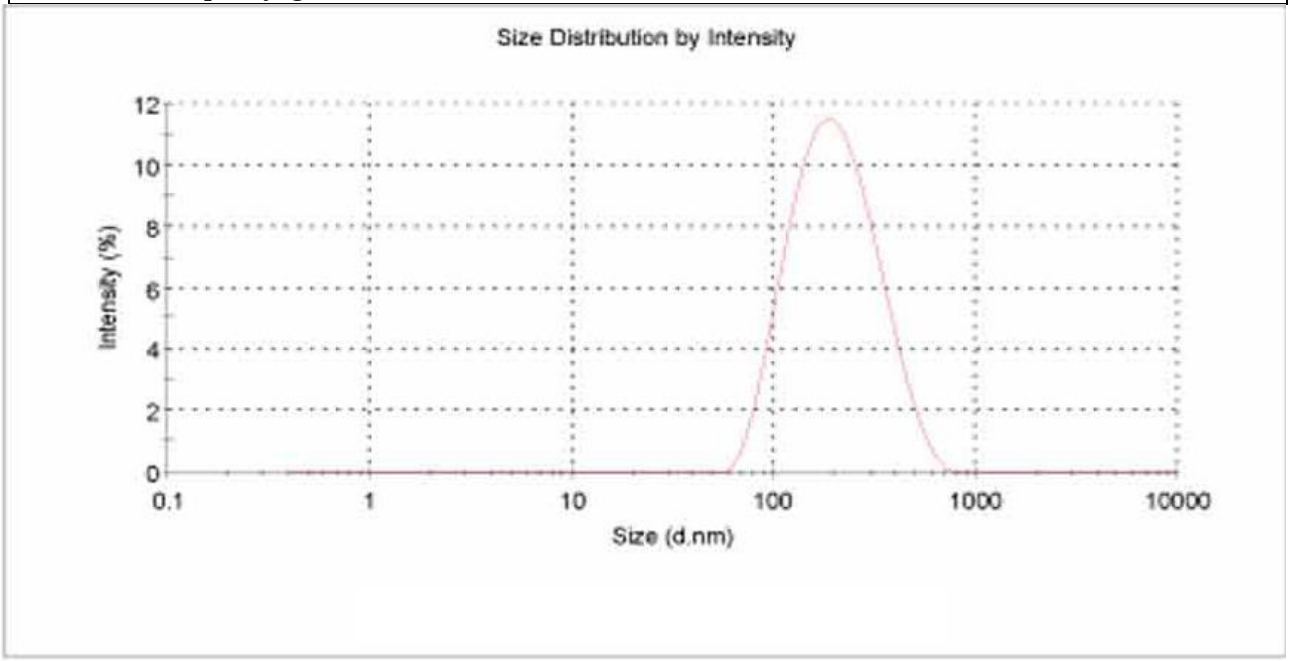

Table 16: Ethosomes *In-vitro* cumulative % drug release profile for Saxagliptin.

Time(hrs)	GF1	GF2	GF3	GF4	GF5	GF6	GF7	GF8	GF9
0.08	32.6	12.6	6.57	2.15	6.28	7.26	6.6	5.31	4.46
0.16	48.3	29.08	16.29	8.26	13.5	16.84	11.08	12.42	9.4
0.25	56.41	32.2	29.04	20.68	26.15	31.29	27.2	24.4	20.42
0.5	68.27	51.04	36.02	26.48	37.62	44.61	34.04	31.24	28.45
1	74.42	67.5	62.1	37.02	43.67	55.29	39.7	37.02	34.72
2	86.3	82.11	74.3	44.6	55.72	60.46	52.4	49.7	46.27
4	100.2	90.6	89.3	55.7	64.28	68.4	60.6	55.5	58.37
6		100.8	100.9	60.77	69.5	74.6	67.7	64.4	60.22
12				69.1	77.2	83.6	79.5	73.3	68.12
24				77.62	86.12	96.7	88.11	78.2	72.7

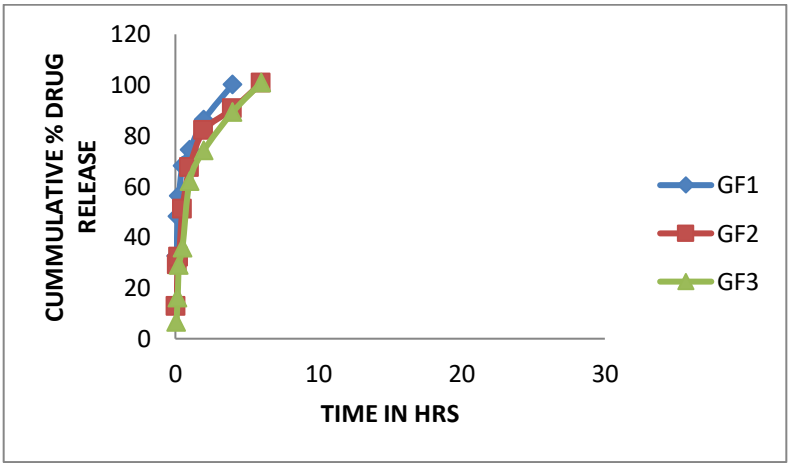

Figure 12: Ethosomes containing Saxagliptin dissolution profile for formulations GF1-GF3.

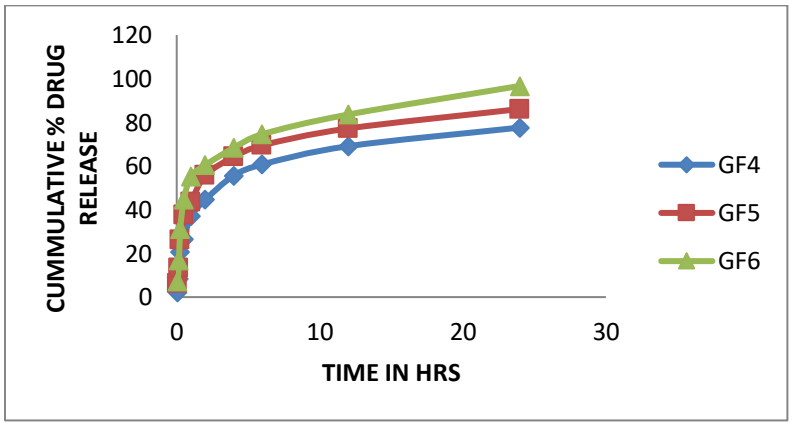

Figure 13: Ethosomes containing Saxagliptin dissolution profile for formulations GF4-GF6.

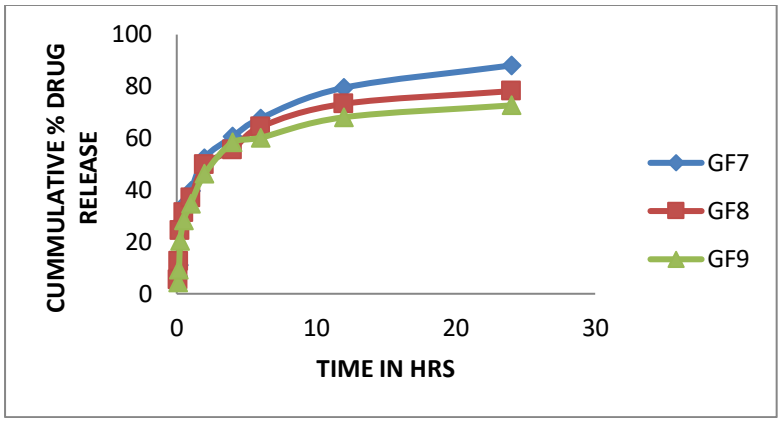

Figure 14: Ethosomes containing Saxagliptin dissolution profile for formulations GF7-GF9.

ETHOSOMAL GEL:

From the above tables, it was verified the ethosomal gel release theory of GF4, GF5, GF6, GF7, GF8, GF9 up to 24 hours in two products. And it was also confirmed from the table that the formulation (GF6) showed maximum drug release of up to 24 hours in both products.

PHARMACOKINETIC PROFILES FORGF6 ETHOSOMAL GEL:

Table 17: Release kinetics for optimized formulation.

	ZERO	FIRST	HIGUCHI	PEPPAS
	% CDR Vs T	Log % Remain Vs T	%CDR Vs √T	Log C Vs Log T
Slope	2.957925303	-0.05439445	17.25120161	0.38390172
Intercept	37.11954601	1.806980214	24.37709307	1.58555375
R 2	0.611771069	0.949144503	0.842450504	0.82013441

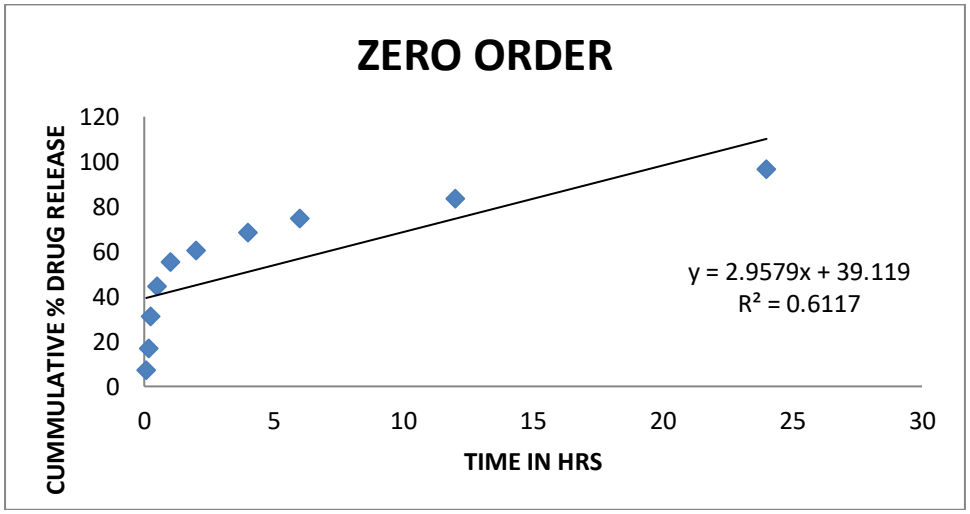

Figure 15: Zero Order Kinetics ForGF6Ethosomalgel.

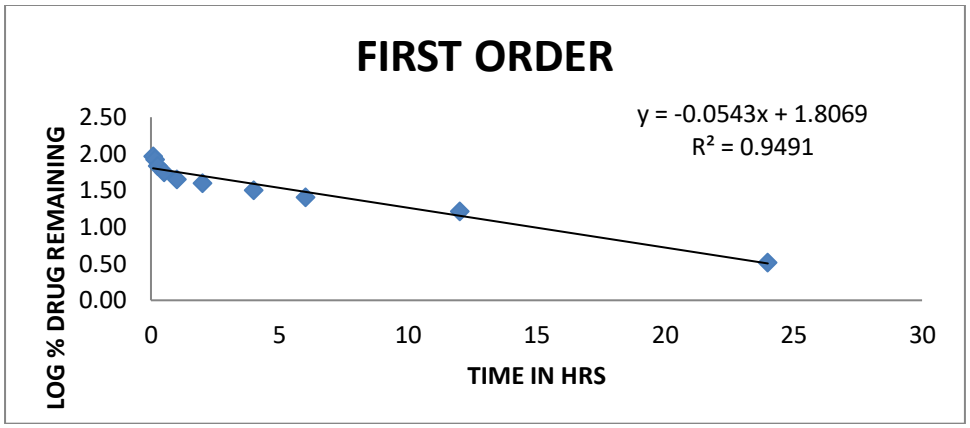

Figure 16: First Order Kinetics for GF6Ethosomalgel.

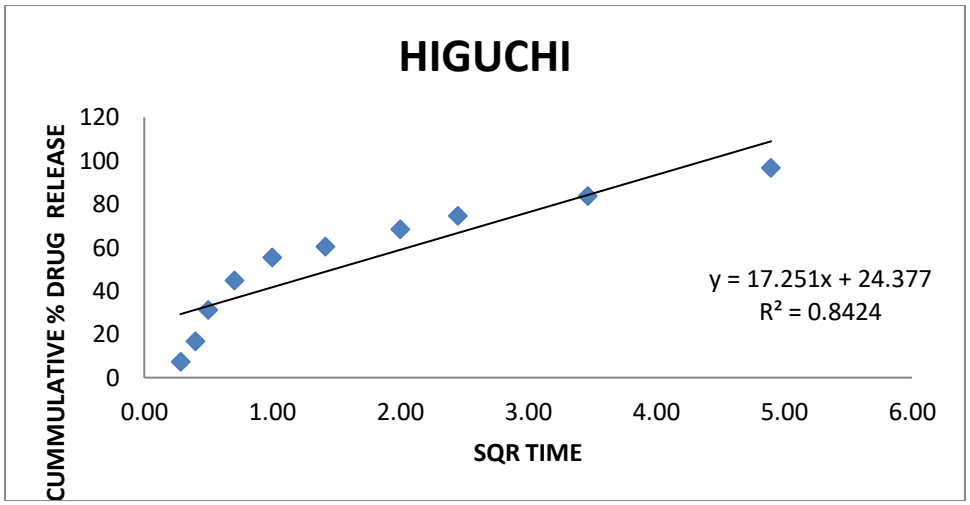

Figure 17: Higuchi's model For GF6Ethosomalgel.

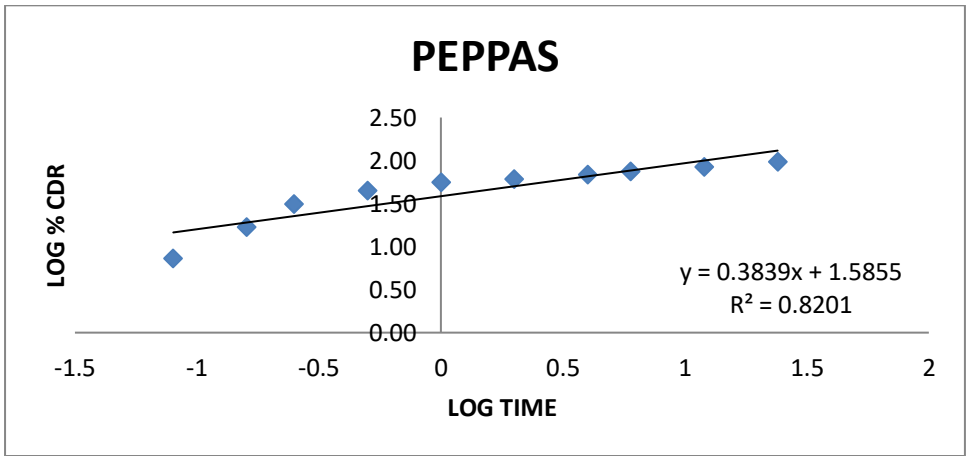

Figure 18: Peppas model For GF6Ethosomalgel.

Stability studies:
The stability experiments were performed in conjunction with the protocol mentioned in the section of the chapter. The findings are seen in the table below.

Table 18: % Entrapment efficiency and % Drug content after stability studies.

Number of Days	% Entrapment Efficiency at temperatures			% Drug Content at temperatures		
	4±2°C	25±2°C	37±2°C	4±2°C	25±2°C	37±2°C
15	84.8	84.8	84.8	98.6	98.47	98.60
30	84.79	84.48	83.49	98.12	98.92	98.71
45	84.26	83.28	83.27	97.11	98.42	98.50
90	83.47	83.22	82.78	97.42	98.34	98.02

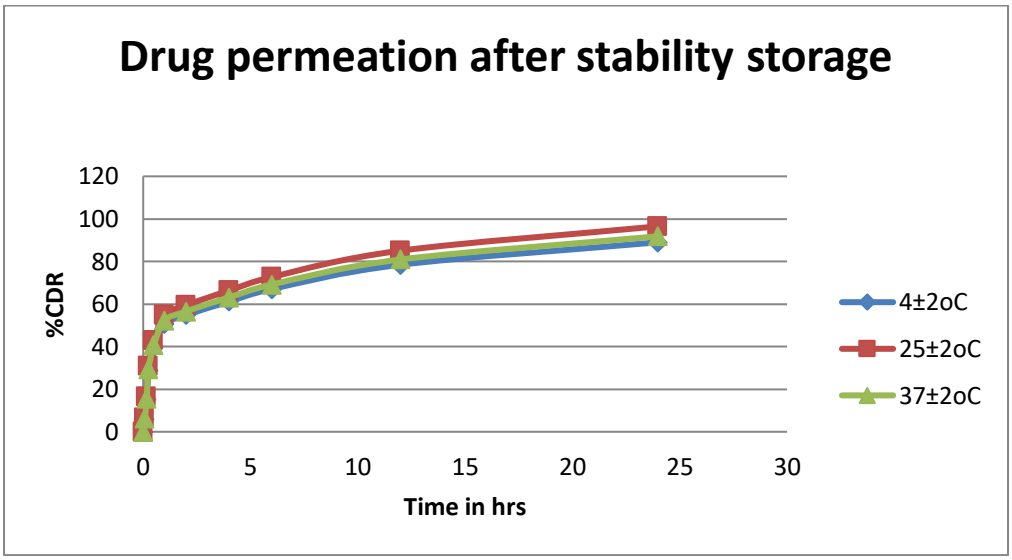

Figure 19: Graph showing dissolution profile for formulations GF6 after storage at different temperatures.

SUMMARY:

There are some possible benefits of the Transdermal path over traditional routes. These benefits include the prevention of first-pass effect, the predictable and sustained duration of action, the minimization of adverse side effects, the effectiveness of short half-life medications, the enhancement of physiological and pharmacological reactions, and, most notably, the avoidance of changes in blood levels. It offers comfort for patients. Yet poor penetration rates are one of the big issues with successful drug delivery.

The vesicular mechanism (liposomes and niosomes) is emerging as the next step in enhancing the topical delivery of drugs. Liposome technology has improved, so much so that they can now be called "Ehosomes", along with the "system" which has greater transdermal flux as it produces little skin inflammation and produces excellent skin depositing capability. With little change, the mixture of saxagliptin ethosomes was prepared using the method stated by Touitou etal. (2000). Ethosomes containing 20 percent, 30 percent, & 40 percent IPA with sonication were tested.

After verifying the presence of vesicles and their duration, ultra-centrifugation was tested for drugs trapped by the vesicular system. Sonicated ethosomes showed higher value i.e. 96.7 percent containing 40 percent w/w IPA. The dialysis membrane was used to conduct in-vitro liberation. The first order was found to be the order of drug release for optimized gel formulation. The ethosomes containing 30 percent

w/w IPAA have considered the percentage of opioid concentration in the skin to be maximum.

CONCLUSION:

In the present work, the mixture of ethosomal gels of saxagliptin has been formulated.

- The findings of this research suggest that the process of ion gelation can be successfully used to generate ethosomal gels of saxagliptin.
- The physical mixture's FT-IR spectrum showed that the drug is consistent with the polymers and copolymers used.
- Carbopol and IPA comprising ethosomal gels and phospholipids had a minimum size length of 613µm.
- Increased polymer concentration contributed to an improvement in the efficacy of drug entrapment, particle size, percentage.
- As the polymer and copolymer concentration increased, the in vitro drug release decreased.
- In comparison with other formulations, GF6 displays maximal drug release in 1440 min, across all formulations.
- Drug release mechanism review found that the drug release from the formulations followed the mechanism of non-fickian diffusion and followed first-order kinetics.
- Based on the results of evaluation tests, GF6 coded formulation was considered to be the best formulation.

REFERENCES:

1. Jain N, Talegonkar S, Jain NK. Modern methods of accessing the blood stream: Evolving transdermal drug delivery techniques. The Study in Pharma. Sep - October 2004;41-60.
2. Jain NK. Advances in the distribution of controlled and innovative medications. 1ed, Publishing of New Delhi:stCBS; 2001; 428 - 451.
3. Jain S, Bhandra D, Jain S, and Jain NK. Transfersomes, a novel transdermal drug delivery carrier. 1st Version Controlled and Novel Drug Distribution. CBS New Delhi Publishers and Distributors 1997: 426 - 451.
4. Vyas SP, Khar RK. Managed principles and developments in drug distribution. Fresh Delhi's Vallabhprakashan. The 2002 First Edition: 173 - 243.
5. Tuitou E, Godin B, and Weiss C. Improved transmission by Ethosomal carriers into and through the skin. Study on Substance Dev 2000; 50; 406 - 445.
6. Chien YW. Drug delivery systems for managed and modulated-release. In: Swarbrick J, Balyan JC, Pharmacy Science Encyclopaedia. New York: Dekker Marcel. From 1990;281-313.
7. Leon Shargel, Susanna Wu-Pong, Andrew BC Yu.Drug products with modified-release. In:Biopharmaceutics & Pharmacokinetics Applied, 5th ed. In 2004.
8. Meera C Singh, Ajinkya S Naik, Sawant SD. Method of delivery of transdermal medications with a big focus on transdermal patches. 2010 J Pharm Res; 3(10): 2537-2543.
9. Robinson JR, Lee VH. Controlled fundamentals and applications for drug distribution. Two-nd ed. New Delhi: Editors & Distributors of CBS; 2005; vol 29: 523-552.
10. KottaKranthi Kumar, Sasikanth K, Sabareesh M, Dorababu N. Diakerein cream formulation and assessment. Clinical Res of Asian J Pharm 2011; 4(2): 93-98.
11. Richard H Man, Jonathan Hadgraft. Transdermal drug delivery: problems of growth and effort for science. From Marcel Dekker. Vol 35: 1-16.
12. Kaplun-Frischoff Y and Touitou E. Skin permeation testosterone improvement through technique by eutectic drug development and association with skin lipids. 1997 J Pharm Sci; 86: 1394 - 9.
13. Bhavsar JD, Brahambhatt VG, Patel MR, Patel KR, Patel NM. Article of Review: Novel Semisolid Approaches. World Res 2011 Int J Pharm; 2(1): 1-22.
14. Khandare JN, Jiwandad BH and Uppal RR. Nimesulideniosomes for topical distribution planning and assessment. Int Drug 2001 April; 38(4): 197-202 respectively.
15. Facts on Medications and Reference. Facts and reference publication, edition of Missouri ® 1999: 25855.
16. Bhavana Vora, Khopade AJ and Jain NK. Transdermal device dependent on niosomes for distribution of contraceptives. Abstract of the 1996 IPC: AP59; 41.
17. Kaplun-Frischoff Y and Touitou E. Skin permeation testosterone improvement through technique by eutectic drug development and association with skin lipids. 1997 J Pharm Sci; 86: 1394 - 9.
18. Touitou E, Godin B, Dayan N, Weiss C, Piliponsky A and Levi-Schaffer f. Mediated intracellular and ethosomal carrier transmission. 2001;22: 3053-3059 biomaterials.