

Formulation, Development and Evaluation of Nasal Gel of Naratriptan

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Abstract

The prolonged residence of drug formulation in the nasal cavity is of most importance for intranasal drug delivery. The aim of the present study was to develop a mucoadhesive in situ gel with reduced nasal mucocilliary clearance in order to improve the bioavailability of the antimigraine drug, Naratriptan Hydrochloride. Hence it was planned to formulate in situ nasal gel of Naratriptan Hydrochloride for systemic delivery. Xanthan gum was used as a natural mucoadhesive polymer to formulate nasal in situ gel of Naratriptan Hydrochloride to sustain the release of drug, to reduce mucociliary clearance thereby increasing the contact of formulation with nasal mucosa and hence improving the absorption of drug. Carbopol 940 was used as a pH induced sol gel conversion of formulations. These formulations were evaluated for pH, drug content, viscosity, gel strength, mucoadhesive strength, *in vitro* drug release and *in vitro* permeation profile. A 3^2 full factorial design was applied to study effect of varying concentration of independent variables Carbopol 940 (X_1) and Xanthan gum (X_2) on dependent variables in vitro drug release, viscosity and mucoadhesive strength. In vitro drug release kinetics was studied using different kinetic models to know exact mechanism of drug release. It was found that formulation additives shows effect on drug release, viscosity and mucoadhesive strength, as the concentration of polymers increases mucoadhesive strength and viscosity increases, drug release was also increases.

Keywords: Nasal Gel, Carbopol 940, Naratriptan Hydrochloride, Xanthan gum, PEG400, Mucocilliary Clearance

1. Introduction

Oral route is taken into consideration to be the most convenient and cheap of drug administration but owing to its inefficiencies which includes poor bioavailability, first pass metabolism, drug solubility and absorption issues, there is an obvious need for different novel system for drug delivery. Parenteral routes of administration like subcutaneous route is another alternative but the dislike of injection make this dosage form less acceptable for the patient. Intranasal drug transport is currently recognized to be helpful and reliable different to oral and

parenteral routes. Intranasal (nasal cavity) route of drug offers some of one of a kind benefits; which includes, low proteolytic activity, prevention of harsh environmental conditions, hepatic first pass metabolism, and potential direct transport to the brain may want to serve the benefit of ease of access, improving bioavailability, excellent permeability in particular for lipophilic and low molecular weight drugs, via first pass metabolism, ease of self-administration, ease of handling with and may also be administered to sufferers in vomiting and unconscious state.^{1,2}

In recent years, research has established that the nasal route is a secure and acceptable alternative to the parenteral administration of drugs. The nasal route has additionally been discovered to be useful in targeting drugs to the central nervous system (CNS). The more permeability of nasal mucosa with large surface area provides a fast onset of therapeutic effect. The low metabolic environment of nose has potential to overcome the limitations of oral route and duplicate the benefit of intravenous administration.³

Naratriptan Hydrochloride is a selective 5-HT₁ agonist developed for the acute treatment of migraine which acts by stimulating constriction of dilated cranial arteries and by inhibiting the release of neurogenic inflammatory mediators. Naratriptan is available only in oral form with the recommended dose of 2.5 mg as its oral bioavailability of 50-60% and exhibits a six fold higher affinity for the human recombinant 5-HT_{1B} receptor than sumatriptan. Intranasal administration offers a practical noninvasive alternative mode for drug targeting to the brain. Therefore the present investigation was planned to develop Naratriptan Hydrochloride pH induced mucoadhesive in situ nasal gel which would enhance nasal residence time and to improve bioavailability of the drug as compared with oral route.⁴

In situ gelation can be achieved by using thermo sensitive smart polymers which by sensing nasal temperature forms gel on instillation. In order to formulate pH induced in situ gel for nasal administration, pH induced polymer must have sol gel in the nasal mucosal surface pH range (4.5-6.5). The use of mucoadhesive polymers can increase nasal absorption of drugs with several ways such as increasing the residence time of drug in the nasal cavity and hence improving the absorption of drug. Therefore the present investigation was planned to develop Naratriptan Hydrochloride pH induced mucoadhesive in situ nasal gel which would enhance nasal residence time and to improve bioavailability of the drug as compared with oral route.⁵

2. Materials

Naratriptan Hydrochloride was provided by USV Pharma, Daman. Xanthan gum was provided by Thermo Fisher Scientific India Ltd, Mumbai. Carbopol 940 (Research-Lab Fine Chem. Industry- Mumbai.) and polyethylene glycol 400, Methylparaben of analytical grade were used.

2.2 Methods ⁶

2.2.1 Determination of λ_{\max} of Naratriptan Hydrochloride

The UV spectrum of Naratriptan Hydrochloride was obtained using UV Agilent tech. Naratriptan Hydrochloride (10mg) was accurately weighed and transferred to 100 ml volumetric flask. It was then dissolved and diluted up to 100 ml with pH 6.4 phosphate buffer. The above made solution was further diluted to obtain concentration of 10 μ g/ml. The resulting solution was scanned from 200-400 nm and the spectrum was recorded to obtain the value of maximum wavelength. The λ_{\max} was found to be 281 nm.

2.2.2 Drug excipients compatibility study

Drug and polymer combination (1:1) were fitted in amber color vials. Vials were sealed and stored at room temperature for a period of 21 days. These samples were analyzed for any variation in IR spectrum.

3. PREPARATION OF MUCOADHESIVE NASAL IN SITU GELS: ⁷

In situ gels were prepared by cold technique. To the %w/v, solution of drug in distilled water, Carbopol 940 was added in the quantity of 0.15, 0.2, 0.25 %w/v. This solution was then stirred until Carbopol940 completely swells in it. After the complete swelling of Carbopol, Xanthan gum was added in the quantity 0.075, 0.1, 0.125 %w/v. After the complete hydration of both the polymers PEG 400(5%) and Methylparaben (0.0165%) were added to it. These resulting formulations were then kept at 4°C overnight until clear gel is obtained. Composition of all the formulations is shown in table 1.

3.1 Formulation optimization: ⁸

In situ gels were designed by 3² full factorial design to study the interaction variables of formulation on characterization of *In situ gels*. Amount of carbopol 940 and xanthan gum were selected as independent variable. Amount of Naratriptan Hydrochloride (%w/v) were kept constant.

Independent variables	Levels		
	Lower (-1)	Middle (0)	Upper (+1)
Carbopol 940 (X ₁)	0.15	0.2	0.25
Xanthan gum (X ₂)	0.075	0.1	0.125

3² full factorial design was applied to the formulation that showed the satisfactory results to see the effect of varying the concentration of independent variables carbopol 940 (X₁) and xanthan

gum (X₂) on dependent variables i.e.% cumulative drug release, viscosity, mucoadhesive strength. Composition of all the batches is shown in table 1.

Table 1: Composition of Formulation

Composition formulation code ↓	Naratriptan HCl (%w/v)	Carbopol 940(%w/v)	Xanthan gum (%w/v)	PEG 400 (%v/v)	Methyl paraben (%v/v)	Distilled water Up to (ml)
F1	2.5	0.15	0.075	5	0.0165	50
F2	2.5	0.2	0.075	5	0.0165	50
F3	2.5	0.25	0.075	5	0.0165	50
F4	2.5	0.15	0.1	5	0.0165	50
F5	2.5	0.2	0.1	5	0.0165	50
F6	2.5	0.25	0.1	5	0.0165	50
F7	2.5	0.15	0.125	5	0.0165	50
F8	2.5	0.2	0.125	5	0.0165	50
F9	2.5	0.25	0.125	5	0.0165	50

4. Evaluation of in situ nasal gel

4.1. pH ⁹

pH of each formulation was determined by using Digital pH meter (WPH-10, Wensar digital pH meter). The pH meter was calibrated using pH 4 and pH 7 buffer by using standard buffer tablet.

4.2. Viscosity ¹⁰

Viscosity (rheological properties) of prepared gel was determined with the help of Brookfield Viscometer; type DV-E using spindle 63. Viscosity of formulations were calculated at two different pH, formulation pH and at pH 7.4 with changing shear rate.

4.3. Measurement of gel strength ¹¹

A sample of 50g of the nasal gel was placed in a 50 ml graduated cylinder. A weight of 5 g was put onto the gel surface. The gel strength, which is a sign for the viscosity of the nasal gel at physiological temperature, was calculated by the time in seconds required by the weight to penetrate 5 cm deep into the gel.

4.4. Determination of Mucoadhesive strength: ¹²

The mucoadhesive strength of every formulation was determined by measuring the force needed to detach the formulation from goat nasal mucosal tissue by employing a changed chemical balance. A section of nasal membrane was cut from the goat's nasal cavity and mucosal aspect was instantly mounted into every glass bottle employing an elastic band. The vials with nasal membrane were kept at 37°C for five minutes. Then next bottle with a region of mucosa was connected to the balance in inverted position whereas initial bottle was placed on a height adjustable pan. Mounted quantities of sample of every formulation were placed onto the nasal membrane of first bottle. Then the height of second bottle was adjusted in order that tissue layer surfaces of each vials are available intimate contact. 2 minutes contact time was given to confirm intimate contact between tissues and therefore the sample. Then weight was increased within the pan till vials got detached.

The bioadhesive force, expressed as the detachment stress in dyne/cm², was determined from the minimal weights that detached the tissues from the surface for each formulation using the following equation.

$$\text{Detachment stress (dyne/cm}^2\text{)} = m \times g / A$$

Where, m =Weight required for detachment of two vials in gm

g = Acceleration due to gravity [980cm/s²]

A = Area of tissue exposed

The nasal mucosa was changed for each measurement. Measurements were repeated three times for each of the gel preparations.

4.5. Drug content ¹³

Drug content was determined by taking 1ml of formulation in 100 ml volumetric flask. It was dissolved in distilled water appropriately and final volume was made to 100 ml with distilled water. 1ml quantity from this solution was transferred into the 10ml volumetric flask and final volume was made to 10ml by using distilled water Finally the absorbance of prepared solution was measured at 281nm by using UV visible spectrophotometer. By using absorbance value % drug content in the formulation was calculated.

4.6. In Vitro Drug Release study ¹⁴

A) Preparation of simulated nasal solution: Weigh accurately 0.87% NaCl, 0.31%KCl and 0.088% CaCl₂·2H₂O and dissolve in 1000 ml of distilled water to produce simulated nasal solution; finally adjusted the pH with phosphoric acid to 6.4.

B] In vitro release study of the formulation was dispensed using laboratory designed diffusion cell through egg membrane. From the gel 1 ml was placed in donor compartment and freshly prepared simulated nasal solution (The simulated nasal fluid (SNF) contained in receptor compartment (100ml)). Egg membrane was mounted between donor and receptor compartment. Temperature of receiver section was maintained at $37\pm 2^{\circ}\text{C}$ during experiment and content of the receiver compartment was stirred via magnetic stirrer. The location of donor compartment was adjusted so that egg membrane just touches the diffusion fluid. An aliquot of 1 ml was withdrawn from receiver section after 1, 2, 3, 4, 5, 6, 7 and 8 hrs. And the same volume of recent medium was replaced. Aliquot so withdrawn were suitably diluted and analyzed using UV visible spectrophotometer at 281 nm. The concentration of drug was calculated from a previously constructed calibration curve.

$$(y = 0.019x + 0.003, R^2 = 0.999).$$

4.7. Drug release kinetics ¹⁵

It is generally understood that the release of the drug from gels can be measured as mass transport phenomenon involving diffusion of the molecules from a region of higher concentration to a region of lower concentration in the surrounding environment. The in vitro drug release information was fitted to different models, i.e. zero order, first order, Higuchi and Connor's and Korsmeyer's Peppas to study the drug release mechanism of the formulation.

4.8. In Vitro permeation study ¹⁶

Natural membranes are utilized to determine in vitro permeation study to mimic the in vivo permeation patterns. In this testing goat nasal mucosa was utilized because the respiratory area of goat is large and it is easy to get. Fresh mucosal tissue was detached from the nasal cavity of goat. The tissue was positioned on the diffusion cell with permeation area 0.75cm^2 . The acceptor chamber of the diffusion cell (laboratory designed) with a volume capacity 100ml was filled with simulated nasal fluid (SNF). From the gel formulation 1ml was placed in donor compartment. At predetermined time point of 1,2,3,4,5,6,7 and 8 hrs 1ml of sample was withdrawn from the acceptor compartment replacing the sample collected with simulated nasal fluid after each sampling for period of 8 hrs. Then samples were specifically diluted and absorbance was noted at 281nm. Permeability coefficient (p) was calculated by the following formula:

$$P = \frac{dQ}{dt} \cdot \frac{1}{C_0 \cdot A}$$

Where, dQ/dt is the flux or permeability rate (mg/h), C_0 is the initial concentration in the donor compartment, and A is the effective surface area of nasal mucosa.

4.9. Stability study¹⁷

Stability studies of the optimized formulations were carried out according to ICH guidelines $25^{\circ}\text{C} \pm 2^{\circ}\text{C}$, $60\% \pm 5\%$ RH to test the physical and chemical stability of the developed in situ nasal gel. At an interval of one month for 3 consecutive months. A sufficient amount of pH sensitive in situ gel, in screw capped vials was stored specified condition ($25^{\circ}\text{C} \pm 2^{\circ}\text{C}$, $60\% \pm 5\%$ RH). The results were compared with respect to pH, viscosity and drug content to indicate stability for optimized formulation.

5. RESULTS AND DISCUSSION

5.1. Compatibility study

Infrared Spectroscopy

The IR spectra of Naratriptan, polymers and physical mixture were generated. The IR absorption bands observed in the IR spectrum of drug and polymers resembles with that of found in the physical mixture proves compatibility of drug with polymers.

5.2. pH

The normal physiological pH of the nasal mucosa ranges from 4.5-6.5. But the nasal cavity has the capability to tolerate pH between 3-10. pH of all formulations was found to be between 5.2 to 5.9 that is within the range, which are presented in the Table 2.

5.3. Viscosity

Viscosities of all the formulations were noted at formulation pH and pH 7.4. It was observed that as the pH increases viscosity also increases. Mucoadhesive polymer Xanthan gum is also having synergistic effect with pH. All the formulations showed pseudoplastic flow. It was also observed that formulations shows decrease in viscosity as shear rate (rpm) was increased which indicate that gel has the pseudo plastic flow. Viscosities of all the formulations at 100rpm are shown in table 2 shows viscosity profile of all formulations.

5.4. Gel strength

Gel strength was recorded for all the formulations by using laboratory designed apparatus. It was observed that gel strength is showing synergistic effect with the viscosity, as the polymer concentration and pH increases gel strength also increases. Gel strength for the formulations is noted in Table 2.

5.5. Drug content

The percentage drug content of all the prepared gels formulations were checked and found to be in the Range of 98-103%. Therefore uniformity of content was maintained in all formulation. Drug content of all the formulations is listed in Table 2.

5.6. Mucoadhesive strength

Mucoadhesive strength was determined by measuring the force required to detach the formulation from mucosal surface that is detachment stress. Results reveal that increase in Carbopol 940 and Xanthan gum concentration increases the mucoadhesive strength. This was due to interaction of polymeric chains with the mucin strands to form weak chemical bonds due to stronger mucoadhesive force. Mucoadhesive strength is listed in Table 2.

5.7. In vitro drug release study

Out of nine formulations maximum release after 8 hrs was found for F8 formulation. This indicates release of 96.56% drug available for anti-migraine activity of the drug. F8 formulation showed steady state release up to 8hrs which also indicates that this formulation would show better contact with biological membrane. (Figure 1)

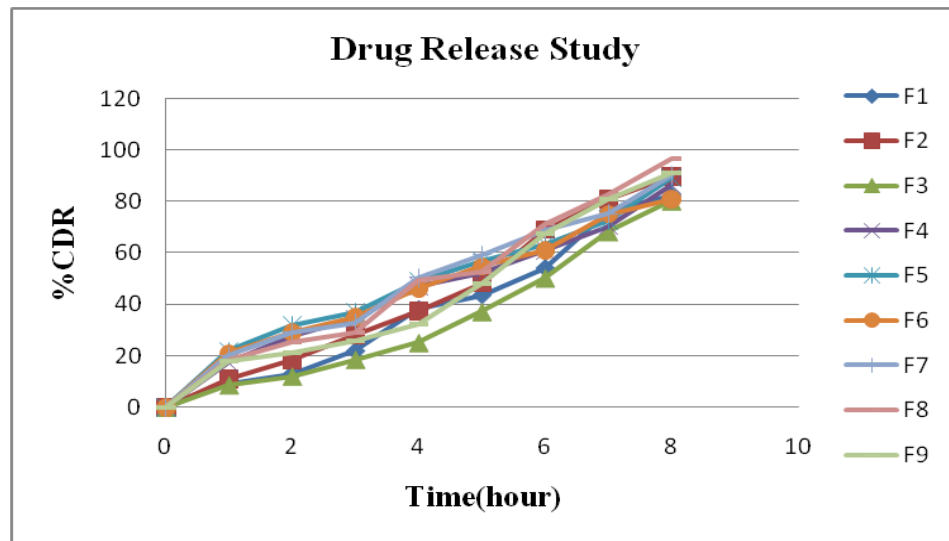


Figure 1: In-vitro drug release profile of Naratriptan from nasal gel formulation F1 to F9

Table 2: Evaluation parameters for all the formulations

Formulation code	pH	Gel strength [sec]	Viscosity [cps] at 100rpm	Drug content %	Mucoadhesive strength [gm]	In-vitro drug release study

F1	5.70±0.1	1.02±0.013	358	98.64±0.43	22.70±0.21	18.57 ± 0.15
F2	5.22±0.2	1.11±0.010	548	99.23±0.27	36.34±0.25	20.56± 0.21
F3	5.40±0.2	1.20±0.02	625	98.96±0.16	42.71±0.23	26.86± 0.15
F4	5.51±0.3	1.36±0.01	552	99.11±0.27	35.13±0.16	33.08± 0.15
F5	5.28±0.2	1.40±0.020	635	101.56±0.26	69.20±0.72	49.40 ± 0.23
F6	5.51±0.2	1.64±0.021	948	99.42±0.99	96.62±0.61	71.36± 0.17
F7	5.78±0.2	2.03±0.021	689	98.52±0.33	73.41±0.15	82.65± 0.22
F8	5.98±0.3	2.63±0.030	895	99.92±0.21	112.10±0.31	96.84± 0.16
F9	5.70±0.1	3.01±0.025	1023	103.64±0.43	146.25±0. 62	18.57 ± 0.15

5.8. Drug release kinetics

In vitro drug release kinetics was studied for all the formulations using different kinetic models. From the regression value it can be predicted that formulation follows Korsmeyer's Peppas because regression value was greater than 0.9 (concentration independent mechanism) Higuchi and Korsmeyer's Peppas release kinetics (r^2 value greater than 0.9), the n value of Korsmeyer's Peppas release kinetics was near to 0.5 from which we can conclude that formulation follows fickian release mechanism that is release by swellable polymeric matrix.

5.9. *In Vitro* permeation study

Table 3: *In Vitro* permeation study for optimized batch F8

TIME(hours)	% Cumulative drug permeation (±S.D.)
1	18.10 ± 0.12
2	34.05± 0.11
3	40.12± 0.12
4	52.16± 0.16
5	61.85± 0.10
6	69.36 ±0.08

7	73.65± 0.19
8	89.21± 0.15

In vitro drug release was observed for the optimized formulation by using goat nasal mucosa. Permeation of the drug from goat nasal mucosa was studied for 8 hrs. It was found to be 89.21% at 8th hr. Permeation of the drug shows synergistic mechanism with that of *in vitro* drug release.

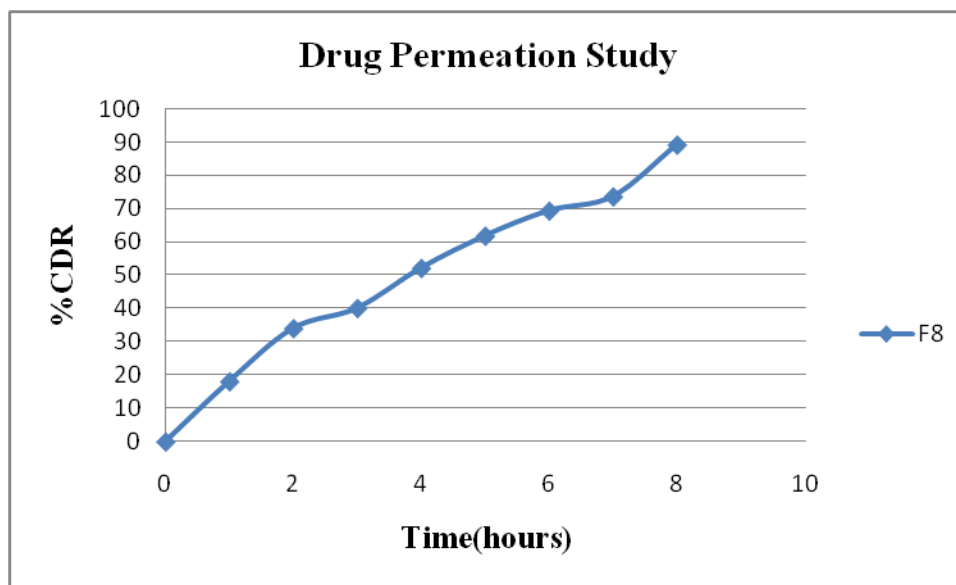


Figure 2: *In- vitro* permeation study for optimized batch F8

5.10. Stability study

Results of the stability studies showed that there is no change in the physical parameters of the formulation. Drug content of the formulation was also found to be same as that before stability testing.

Statistical analysis

A 3² full factorial design was selected and the 2 factors were evaluated at 3 levels, respectively. The percentage of Carbopol 940 (X_1) and Xanthan gum (X_2) were selected as independent variables and the dependent variable was % drug release, viscosity and Mucoadhesive strength. The data obtained were treated using Design expert version 7.0 software and analyzed statistically using analysis of variance (ANOVA). The data were also subjected to 3-D response surface methodology and contour plot to study the interaction of Carbopol940 (X_1) and Xanthan

gum(X_2) on dependent variable. ANOVA for the dependent variable % drug release. The values of X_1 and X_2 were found to be significant at $p < 0.05$, hence confirmed the significant effect of both the variables on the selected responses. From this data optimum concentration of Carbopol 940 0.2%w/v and Xanthan gum 0.125%w/v was found. Contour plot and 3-D response surface

Shown in figures.

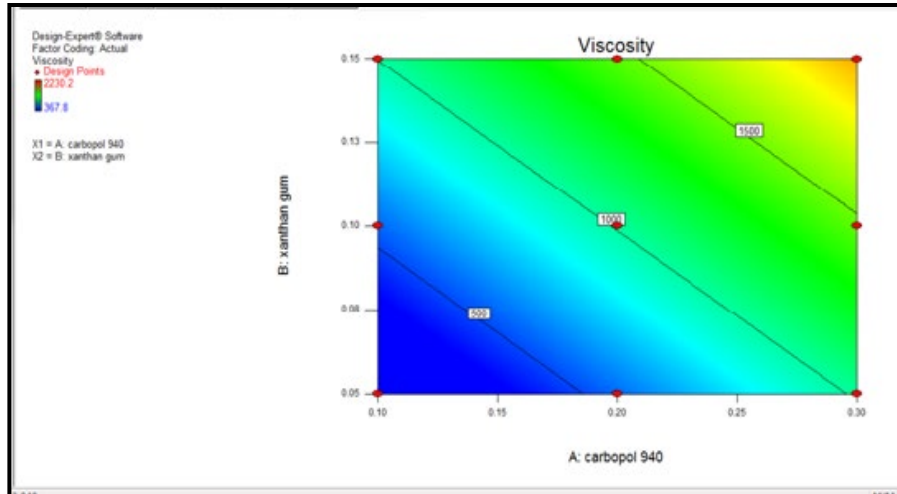


Figure 3: Contour plot showing effect of Carbopol 940 and xanthan gum on viscosity.

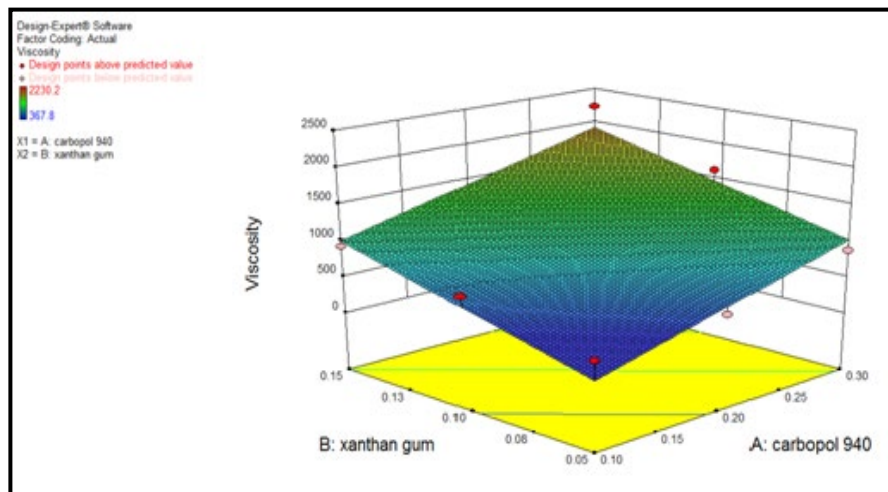


Figure 4: Surface response plot showing effect of carbopol 940 and xanthan gum on viscosity.

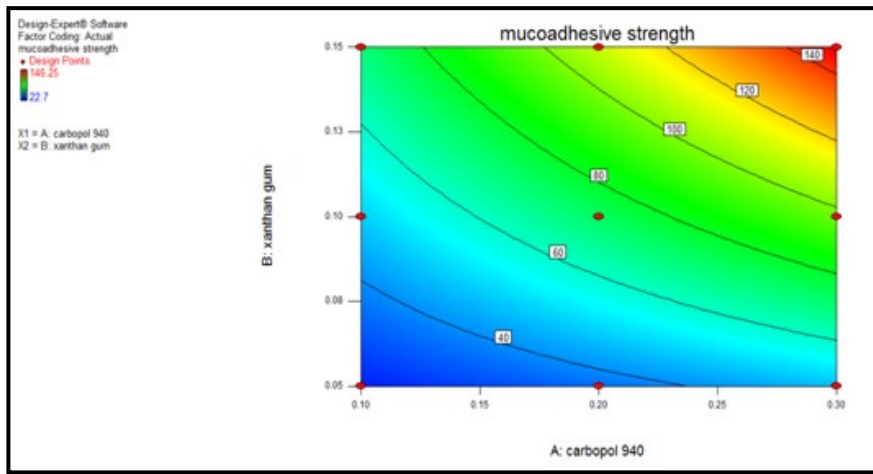


Figure 5: Contour plot showing effect of carbopol 940 and xanthan gum on mucoadhesive strength

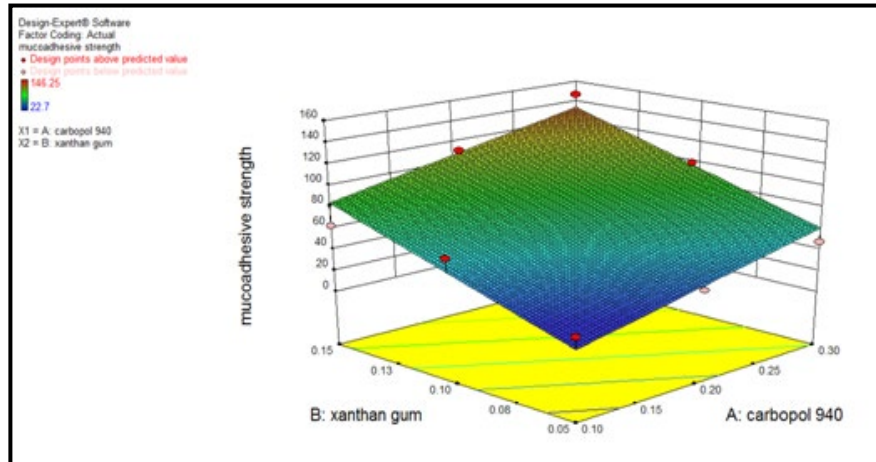


Figure 6: Surface response plot showing effect of carbopol 940 and xanthan gum on mucoadhesive strength

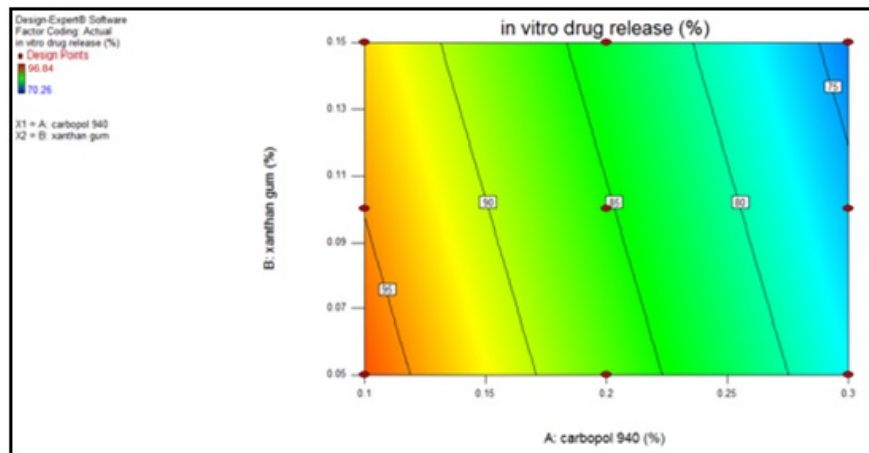


Figure 7: Contour plot showing effect of carbopol 940 and xanthan gum on drug release

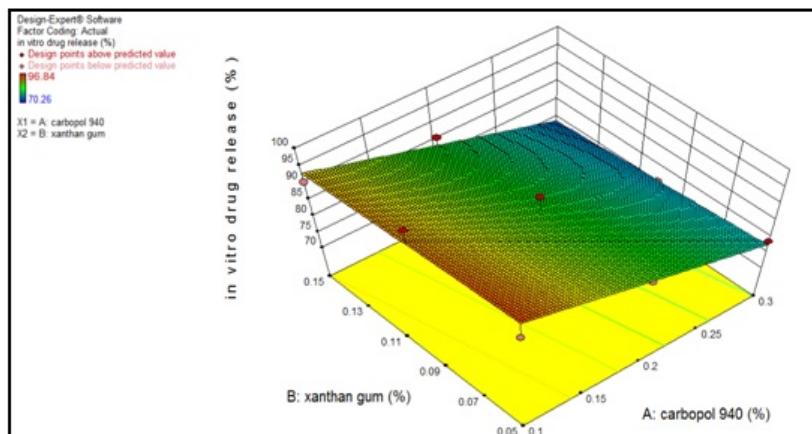


Figure 8: Surface response plot showing effect of carbopol 940 and xanthan gum on drug release

6. CONCLUSION

The formulation and development of *in situ* mucoadhesive gelling system for nasal administration for an anti migraine drug Naratriptan Hydrochloride by using Carbopol 940 and Xanthan gum achieves the systemic delivery of drug through the nasal route and thus avoiding its first pass metabolism. The *in situ* gels so prepared were considered for its viscosity, gel strength, mucoadhesion, drug content, drug release rate. Formulation (F8) was found to be optimized due to its desirable Drug release (96.56). The results of a 3^2 full factorial design

shown that the amount of Xanthan gum and Carbopol 940 significantly affect the dependent variables such as % cumulative drug release, mucoadhesive strength, viscosity. As a result, it can be concluded that, by adopting a systematic formulation approach, a best possible point can be reached in the shortest time with minimum efforts and improve drug bioavailability and patient compliance.

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