Thiomer Based Transmucosal Drug Delivery System for an Anti-Ulcer Drug

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Abstract

Objective: The present study was carried out to formulate bilayered patches of Esomeprazole for buccal delivery. **Methodology:** The patches were prepared using thiolated chitosan as mucoadhesive polymer and ethyl cellulose as the impermeable backing layer. The formulations were developed on the basis of Central Composite Design using Response Surface Methodology after preliminary trials. Concentration of Thiolated chitosan, PVP K30 and Glycerol were chosen as Independent variables and mucoadhesive time, mucoadhesive strength as dependent variables. **Results:** The patches showed good mucoadhesive strength in the range of 74 - 115 g. *In-vitro* drug release studies showed that formulations with high ratios of the polymers were able to sustain drug release up to 8 h. Optimized Formulation OPTI-TC containing the highest ratio of thiolated Chitosan was found to be the best formulation in terms of mucoadhesive strength, drug release and drug permeation. **Conclusion:** Thus a stable, safe and effective bilayered mucoadhesive delivery system of Esomeprazole in the form of patches was prepared to improve drug bioavailability, avoid degradation of drug and to prevent drug loss in saliva.

Key Words: *Promethazine HCl, Oro Dispersible Tablets, Super Disintegrant, Sublimation, Antiemetic*

1. INTRODUCTION

The buccal region of the oral cavity is an attractive target for administration of the drug of choice. Buccal delivery involves the administration of the desired drug through the buccal mucosal membrane lining of the oral cavity. Unlike oral drug delivery, which presents a hostile environment for drugs, especially proteins and polypeptides, due to acid hydrolysis and the hepatic first-pass effect, the mucosal lining of buccal tissues provides a much milder environment for drug absorption. They are a new generation of mucoadhesive polymers which are capable of forming covalent bonds with the mucus and the underlying cell layers, thus exhibiting improved mucoadhesiveness. The permeation enhancing and mucoadhesive properties of thiolated polymers have been investigated to be approximately 80-fold or140-fold higher in comparison to unmodified polymers.[1,2,3] The main aim of the present work was to develop a Transmucosal drug delivery system of Esomeprazole, in the form of bilayered buccal patches using a thiolated polymer for an effective clinical management of gastric ulcers.

The proposed research work includes the synthesis and characterization of a thiolated polymers and the study of effect of thiolated polymer on drug release from buccal patches.

2. MATERIALS AND METHODS

2.1 Chemicals and Reagents

Esomeprazole magnesium Tri- hydrate and PVP-K30 was procured from Yarrow Chem Products, Mumbai. Chitosan was procured from Marine chemicals, chitosan. All other chemicals and reagents used were of analytical grade unless otherwise indicated

2.2 Synthesis of Thiolated Chitosan

Chitosan (500 mg) was dissolved in 50 ml of 1% acetic acid. In order to facilitate reaction with thioglycolic acid, 100mg of ethyl-3-(3-dimethyl-aminopropyl) carbo-di-imide hydro-chloride (EDAC) was added to the Chitosan solution. After EDAC was dissolved, 30ml of thioglycolic acid was added and the pH was adjusted to 5.0 with 3N NaOH. The reaction

mixture was stirred and left for 3 h at room temperature. To eliminate the unbounded TGA and to isolate the polymer conjugates, the reaction mixture was dialyzed against 5 mM HCl five times (molecular weight cut-off 10 kDa) over a period of 3 days in the dark, then two times against 5 mM HCl containing 1.0% NaCl to reduce ionic interactions between the cationic polymer and the anionic sulfohydryl compound. The thiomer solution was kept in deep freezer for 24 hr. The frozen samples were then dried in a lyophillizer (Alpha 1-2 LD plus, Martin Christ, Germany) for a period of 24 hr at −75ºC condenser temperature maintaining a vacuum of 4×10−4 mbar pressure. The dried polymer conjugates were stored at 4ºC for further use.

2.3 Characterization of Thiolated Chitosan

2.3.1 Determination of Thiol Group Content by Ellman's Method

The degree of modification, i.e. the amount of thiol groups immobilized on chitosan, was determined spectrophotometrically with Ellman's reagent quantifying the thiol groups. First, 0.500 mg conjugate was hydrated in 250µl of demineralised water. Then, 500µl Ellman's reagent 3 mg (5, 5-dithiobis (2-nitrobenzoic acid) in 10 ml of 0.5M phosphate buffer (pH 8.0)) was added. The samples were incubated for 3h at room temperature. The supernatant was separated from the precipitated polymer by centrifugation (5000 rpm for 10 min). Thereafter, 300 µl of the supernatant was transferred to a micro cuvette and the absorbance was measured at a wavelength of 450/620 (nm) with a UV-VIS spectrophotometer. Thioglycolic acid standards were used to calculate the amount of thiol groups immobilized on the polymer.[4]

2.3.2 FTIR Studies

Fourier Transform Infrared Spectroscopy (FT-IR) was used to identify the chemical structure of chitosan and the thiolated chitosan. The FTIR spectra were measured with a (FTIR 8400 S, Shimadzu) in the region of 4000–400 cm^{-1}.

2.3.3 Thermogravimetric Analysis (TGA)

The thermal behaviour of thiolated chitosan was tested using a Thermal Analyzer (TGA 400, Perkin Elmer, USA). The operating conditions were as follows: Temperature range of 40-730 ºC, with a heating rate of 20ºC/min in nitrogen atmosphere.The thermal decomposition temperature of the sample was determined from the inflection points of the transition

2.3.4 X-ray Diffraction Analysis (XRD)

X-ray diffraction experiments were performed using AXS D8 Advance (Bruker, USA) diffractometer. The radiation used was nickel filtered CuKα, which was generated using acceleration voltage of 40 Kv and a cathode current of 35 mA. The samples were scanned over a 2-θ range of 3.0º-80º, with counting time of one second per 0.02º

2.3.5 Mass Spectroscopic Studies

Mass spectral analysis was used to identify the chemical structure of chitosan and thiolated chitosan (TGA-chitosan). The molecular weight of the thiolated chitosan was determined by Electron Spray Ionization- Mass spectroscopy (ESI-MS). The mass spectra were recorded on LCMS 2010 (Shimadzu, Japan).

2.4 Optimization Studies

The runs or formulations were developed based on Central Composite Design using Response Surface Methodology after carrying out preliminary trials. The responses were subjected to multiple regression analysis to find out the relationship between the factor used and the responses obtained. The responses subjected for the analysis were;

1. Mucoadhesive Time
2. Mucoadhesive Strength

The effect of formulation variables on the response variables were statistically evaluated by applying one way ANOVA at 0.05 level using Design Expert software (trial version 8.07, State Ease, USA). The design was evaluated by quadratic model.

The optimization of the buccal patch was carried out by taking into the consideration the concentration of Thiolated chitosan, PVP K30 and Glycerol as Independent variables and the mucoadhesive time (minutes) and mucoadhesive strength (g) as responses/ dependent variables. The software suggested 14 formulations (Table 1).

2.5 Preparation of bilayered buccal patches of Esomeprazole

2.5.1 Preparation of Backing Membrane

Backing membrane was prepared by dissolving 500mg of ethyl cellulose in 10ml of acetone by using 2% v/v Di-butyl phthalate as plasticizer. The solution was stirred continuously for 15 min and then poured in a Petri-plate (9 cm in diameter) and left for complete evaporation of solvent to form a backing layer.

2.5.2 Preparation of drug loaded patches

Fourteen formulations as suggested by DOE were prepared by solvent casting technique using ethyl cellulose as backing membrane (Table 2). The casting solutions were prepared by dissolving 20 mg of Esomeprazole and the appropriate polymers in the given range i.e. Thiolated chitosan (0.5-2.5%), PVP K30 (1-5%) in water.

The ingredients were mixed for 20 min using a magnetic stirrer to obtain a uniform dispersion. Glycerol was added at a concentration of 2-10 % w/w of polymers. The solution was then transferred quantitatively to glass petri plates of diameter 9 cm. Controlled solvent evaporation was achieved by placing an inverted funnel over the petridish.

Table 1. Design summary showing experimental runs as per DOE for bilayered buccal patches of Esomeprazole

Run	A: Thiolated Polymer (%)	B: Film Improver (%)	C: Plasticizer (%)
1	0.5	5	10
2	1.5	6.3	6
3	1.5	3	6
4	0.5	5	2
5	2.5	5	2
6	2.5	5	10
7	2.5	1	2
8	1.5	3	10
9	1.5	1	6
10	0.5	1	2
11	1.5	3	2
12	0.5	1	10
13	0.5	3	6
14	2.5	1	10

Table 2. Formulation table for Esomeprazole bilayered buccal patches

Formulation code	Esomeprazole magnesium trihydrate (mg)	Thiolated chitosan (mg)	PVP K30 (mg)	Glycerol (ml)	Purified water (q.s) in ml
R 1	282.6	125	1250	0.137	25
R 2	282.6	375	1590	0.117	25
R 3	282.6	375	750	0.067	25
R 4	282.6	125	1250	0.025	25
R 5	282.6	625	1250	0.037	25
R 6	282.6	625	1250	0.187	25
R 7	282.6	625	250	0.017	25
R 8	282.6	375	750	0.112	25
R 9	282.6	375	250	0.037	25
R 10	282.6	125	250	0.0075	25
R 11	282.6	375	750	0.022	25
R 12	282.6	125	250	0.026	25
R 13	282.6	125	750	0.052	25
R 14	282.6	625	250	0.057	25

2.6 Evaluation of Buccal Patches

2.6.1 Surface pH

The surface pH of the patches were determined in order to investigate the possibility of any side effects, *in-vivo*. An acidic or alkaline pH may cause irritation to the buccal mucosa. It was our attempt to keep the surface pH as close to neutral as possible. For the determination of surface pH, three patches (2× 2cm^2) from each formulation were kept in contact with 1ml of distilled water for 2hrs, in test tubes. Excess water from the tubes was drained and the pH was noted by bringing the electrode near to the surface of the film and allowing it to equilibrate for one min.

2.6.2 Swelling Studies

Three preweighed patches (2×2 cm^2) from each optimized formulations were placed in a beaker containing 20 ml of water. After an interval of 15 min the patches were removed, wiped with tissue paper and weighed again. The procedure was repeated every 15 min till 60 min.

$$S.I. = \frac{(W2 - W1)}{W1}\ x\ 100$$

Where, S.I is swelling index, W1 is weight of buccal patch before soaking in water and W2 is weight of buccal patch after swelling studies

2.6.3 Weight Uniformity

Three films of (2 × 2 cm^2) from each formulation were taken and weighed individually on a digital balance. The results were analyzed for mean and standard deviation

2.6.4 Scanning Electron Microscopy (SEM)

The shape and surface morphology of thiolated Chitosan patch was investigated using Scanning Electron Microscope (JEOL, JSM-6100). The samples were coated with gold and mounted on a sample holder. The electron micrographs were taken at an accelerating voltage of 5KV and 48.56 X magnification.

2.6.5 Drug Content

The distribution of active ingredient in the solution is important to achieve dose uniformity. Films of size (2 × 2 cm^2) were cut and randomly three films were selected and

dissolved in 25 ml of water. The resulting solution was filtered through Whatmann filter paper and diluted suitably with water. The absorbance of the resulting solution was measured spectrophotometrically at 305.5nm

2.6.6 Patch Thickness

Three films (2 × 2 cm^2) of each formulation were taken and the thicknesses were measured using screw gauge. The results were analyzed for mean and standard deviation.

2.6.7 Folding Endurance of the Patch

Folding endurance was determined by repeatedly folding a small strip of film at the same place till it brakes. The number of times, the films could be folded at the same place without breaking will give the value of folding endurance. The results were analyzed for mean and standard deviation

2.6.8 *In Vitro* Bio Adhesive Studies

A modified balance method was used for determination of the *in vitro* bioadhesive studies. Fresh pork buccal mucosa obtained from a local slaughter house was cut into pieces, washed with distilled water followed by phosphate buffer pH 6.8. A piece of buccal mucosa was fixed to the apparatus with instant adhesive. The patch was fixed on the pork buccal mucosa using few drops of phosphate buffer pH 6.8. The other side of the patch was attached to a pan through a pulley. The pan is used to keep the weights. A weight of 5 g was placed and checked if the patch was detaching. Weight was added slowly to the pan until the patch detached from the mucosal surface. This detachment force gave the mucoadhesive strength of the mucoadhesive patch in grams.

2.6.9 *In Vitro* Release Studies

For *in-vitro* release study, cellophane membrane was used as a barrier membrane with Phosphate buffer pH6.8 as a medium. The cellophane membrane was soaked for 24 hours in Phosphate buffer. The patches were evaluated for drug release using Keshary-Chein type diffusion cells. Cellophane membrane was mounted between the donor and receptors compartments. The patch was placed on the cellophane membrane. The diffusion cell was placed in a water bath maintained at 37±2ºC.The receptor compartment was filled with 15 ml phosphate buffer and hydrodynamics was maintained by stirring with a magnetic bead at 50 rpm. One ml sample was withdrawn at regular intervals and replaced with 1 ml fresh medium to maintain the sink condition. The samples were analyzed spectrophotometrically at 305.5 nm.

2.6.10 *Ex-Vivo* Permeation Studies

For *Ex-vivo* permeation study, porcine mucosa was used as a barrier membrane with Phosphate buffer (pH6.8) as a medium. The patches were evaluated for drug release using Keshary Chein type diffusion cells. Porcine mucosa was mounted between the donor and receptors compartments. The patch was placed on the porcine mucosa.

The diffusion cell was placed in a water bath maintained at 37±2ºC.The receptor compartment was filled with 15 ml phosphate buffer (pH6.8) and hydrodynamics was maintained by stirring with a magnetic bead at 50 rpm. One ml sample was withdrawn at regular intervals and replaced with 1 ml fresh medium to maintain the sink condition. The samples were analyzed spectrophotometrically at 305.5 nm.

3. RESULTS AND DISCUSSION

3.1 Synthesis of Thiolated Chitosan

Thiolated Chitosan was prepared by a method as described in section 2.2. Briefly, thioglycolic acid was introduced to chitosan via amide bond formation mediated by a carbodiimide. The properties of the resulting polymer were thereby altered in regard to water solubility, muco-adhesion, bio-

degradability and *in situ* gelling compared to the original polymer.

3.2 Characterization of Thiolated Chitosan

3.2.1 Determination of Thiol Group Content by Ellman's Method

The degree of modification, i.e. the amount of thiol groups immobilized on chitosan, was determined spectrophotometrically with Ellman's reagent quantifying the thiol groups. Thioglycolic acid standards were used to calculate the amount of thiol groups immobilized on the polymer. The concentration of thiol content in thiolated chitosan was found to be 1.75 µg/ml.

3.2.2 FTIR Studies

FTIR was performed for thiolated Chitosan. The IR spectrum confirms the presence of thiol group in thiolated chitosan (Figure 1)

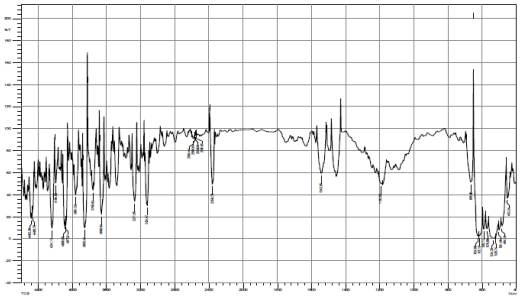

Figure 1: IR Spectrum of Thiolated chitosan

3.2.3 X-Ray Diffraction Studies

The XRD results were in good agreement with the thermal analysis data. The X- Ray diffraction spectra of pure drug Esomeprazole (Figure 2) revealed that the drug was in crystalline state as it showed sharp distinct peaks at 2θ diffraction angles (in order of intensities) of 4º, 10.5º, 13º, 16º, 20.5º, 23º, 26º. X-Ray diffraction spectra of thiolated chitosan (Figure 3) revealed that thiolated chitosan was in crystalline state as it showed sharp distinct peaks at 2θ diffraction angles (in order of intensities) of 29.5º, 32º, 46º, 54º, 57º, 66.5º, 75.5º. The spectra of drug and polymer in (Figure 4) showed sharp distinct peak at 2θ diffraction angles (in order of intensities) of 13º, 23º, 25.5º, 27.5º, 32º, 45.5º, 57º, 66.5º, 75.5º.The drug polymer combination had led to the appearance of a few new peaks, decrease in the intensities of existing peaks and shifting of certain peaks.

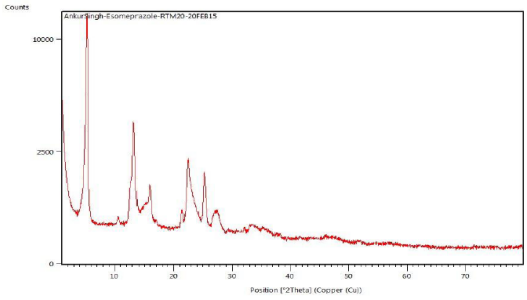

Figure 2: X-Ray Diffraction Spectra of Esomeprazole

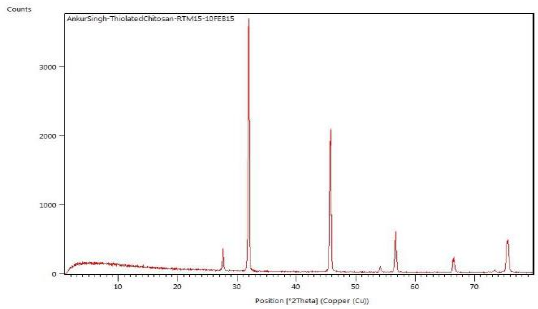

Figure 3: X-Ray Diffraction Spectra of Thiolated Chitosan

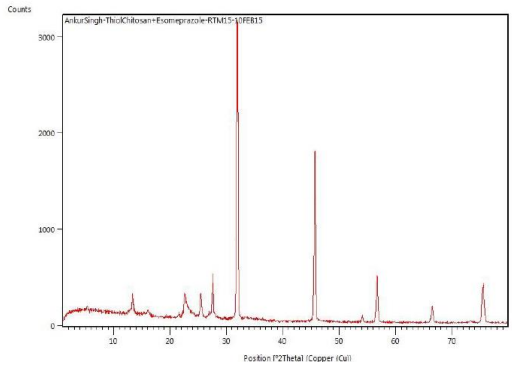

Figure 4: X-Ray Diffraction Spectra of Thiolated Chitosan with Esomeprazole

3.2.4 Thermogravimetric Analysis

The thermos-gravimetric analysis can characterize materials that exhibit weight loss or gain due to decomposition, oxidation or

dehydration. According to the TGA analysis, 3 stages of decomposition were observed in the samples (Figures 5 and 6).

In the first stage both thiolated chitosan and chitosan showed a weight loss between 25 to 125ºC which could be attributed to the loss of water molecules.

In the second stage thiolated chitosan showed decomposition at 240 to 250 ºC with 16.592% weight loss whereas Chitosan showed a decomposition at 300-310 ºC resulting in 56.9% weight loss. In the third stage thiolated chitosan showed decomposition at 300 ºC whereas chitosan decomposed at 350 ºC.

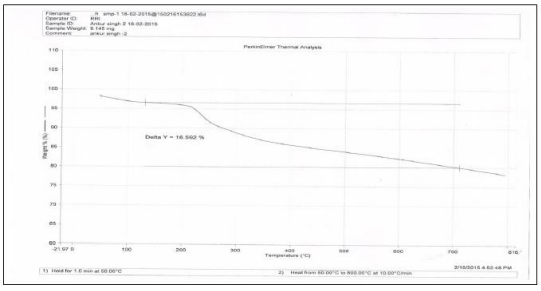

Figure 5: Thermo-Gravimetric Analysis Spectra of Chitosan

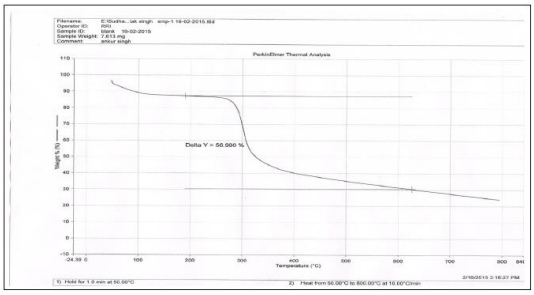

Figure 6: Thermo-Gravimetric Analysis Spectra of Thiolated Chitosan

3.2.5 Mass Spectroscopic Studies

Mass spectroscopy studies were carried out to find the molecular weight of thiolated chitosan. It was also used to identify the chemical structure of chitosan and thiolated chitosan. The base peak of thiolated chitosan was detected at 280 on m/z scale and base peak of chitosan was detected at 382 on m/z scale with high intensity (Figures 7 and 8). The results were in good agreement with the calculated molecular weight of the compound.

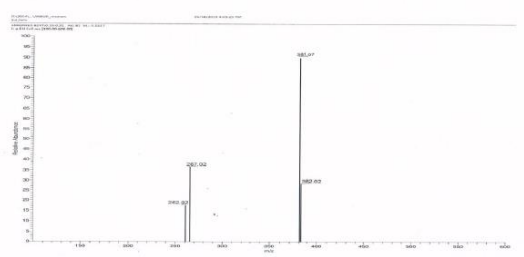

Figure 7: Mass Spectra of Chitosan

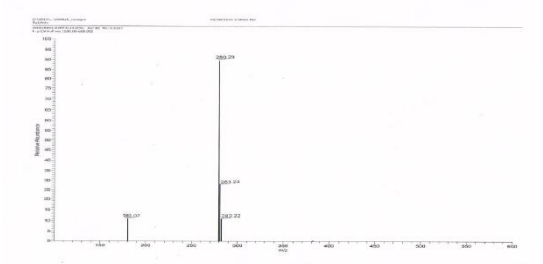

Figure 8: Mass Spectra of Thiolated Chitosan

3.3 Preparation of Bilayered Buccal Patches of Esomeprazole

The synthesized thiolated chitosan was further used for the preparation of bilayered buccal patches of Esomeprazole. Thiolation of chitosan had increased its water solubility which could improve mucoadhesivity in the oral cavity and faster release of drug. The backing membrane was prepared with the polymer ethyl cellulose and di butyl phthalate as the plasticizer. Drug loaded layer was prepared with different concentration of thiolated polymer using PVP K30 as the film improver and Glycerol as plasticizer

3.4 Optimization Studies

The runs or formulations were developed based on **Central Composite Design** using **Response Surface Methodology** after carrying out preliminary trials. Concentration of Thiolated chitosan, PVP K30 and Glycerin were selected as Independent variables and the mucoadhesive time (minutes) and mucoadhesive strength (g) as responses/dependent variables. The software suggested 14 formulations (Table 1).

3.5 Evaluation of Buccal Patches

3.5.1 Patch Thickness

The thickness of the patches including were measured at 3 different points with the help of screw gauge. The thickness ranged from 0.568 to 0.634 mm (Table 3). The thickness range was found to be satisfactory. Considering the fact that the films are of size 2 × 2 cm^2, It should not cause any inconvenience to the patient after application.

3.5.2 Weight Uniformity

Drug loaded patches of 2 × 2 cm^2 were tested for uniformity of weight and the data are given in Table 3. As the concentration of polymer increased the weight of the strip also increased.

3.5.3 Surface pH

The surface pH of all formulations were determined in order to investigate the possibility of any kind of side effects in the oral cavity as acidic or alkaline pH could cause irritation in the oral mucosa. The pH was found to be in the range of 6.8 to 7.2 (Table 3) which was well within the limit of pH of oral cavity.

3.5.4 Folding Endurance

The recorded folding endurance of all the formulation were between 311 to 343 fold (Table 3). The results suggested that the films were not brittle and had high flexibility.

3.5.5 Content Uniformity

Drug content uniformity of the formulation was determined according to the procedure described in 4.7. Drug content was analyzed by UV-Visible spectrophotometer at 305.5 nm. The percentage drug content was between 85 to 92 % as shown in Table 3, which proved uniform drug distribution within the patch

3.5.6 Swelling Studies

Swelling studies indicate the uptake of water into the buccal patch, producing an increase in weight. The swelling behavior of the polymer is reported to be crucial for its mucoadhesive parameters and the drug release. The swelling studies for all formulation were performed and it was observed that as the polymer concentration increased, there was a marked increase in the swelling index. Swelling index increased as the weight gain by the patch increased proportionally with the rate of hydration (Table 4).

3.5.7 Surface Morphology Determination by Scanning Electron Microscopy

The Scanning electron micrographs (Figure 9) of the formulation at 48.56 X magnification showed a showed smooth surface with uniform texture on the surface of the film.

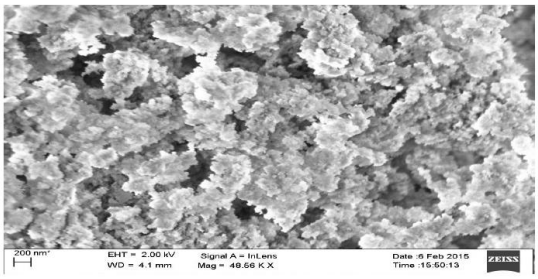

Figure 9: SEM of the Esomeprazole buccal patch at 48.56 X Magnification

3.5.8 *In-Vitro* Mucoadhesion Studies

Retention of a dosage form in the buccal cavity for extended period of time is desirable for therapeutic efficacy and to minimize frequent dosing. The mucoadhesive strength of the patches was found to be a function of nature and concentration of polymer as shown in Table 5. Formulations showed mucoadhesive strength in the range of 74 to 115 g. From the results it was very much clear that as the polymer concentration was increased the mucoadhesive strength also increased.

Formulations R5, R6, R7 and R14 showed increased mucoadhesive strength as they contained 2.5 % of thiolated Chitosan. The

force of adhesion for all formulations was found to be 0.72 to 1.127 N and mucoadhesive time was found to be 320 to 470 min. results shown in (Table 5).

Table 3. Evaluation studies on buccal patches prepared as per DOE

Formulation Code	Weight variation (mg)	Patch thickness (mm)	Folding endurance	Surface ph	Drug content (%)
R1	129	0.568	311	6.8	85
R2	140	0.596	323	6.8	89
R3	139	0.593	321	7.0	87
R4	131	0.571	325	6.8	84
R5	144	0.625	340	7.1	91
R6	145	0.611	336	7.2	90
R7	151	0.634	341	7.2	89
R8	137	0.592	336	6.9	86
R9	141	0.584	316	7.0	88
R10	127	0.566	330	7.0	85
R11	140	0.597	322	7.2	90
R12	130	0.572	317	7.2	90
R13	125	0.576	336	6.8	89
R14	147	0.631	343	7.2	92

Table 4. Swelling index of buccal patches prepared as per DOE

Formulation code	Swelling index (%)			
	15 (min)	30 (min)	45 (min)	60 (min)
R1	41.20	111.2	164.5	209
R2	51.28	99.6	166	221
R3	43.54	107.5	163	234
R4	26.04	105.4	171.5	230
R5	31.66	108	173.40	211
R6	43.31	114	171.02	217
R7	27.71	117	163.67	223
R8	26.63	102	174.49	219
R9	54.55	109	161.2	231
R10	50.52	114	165.66	214
R11	51.19	98.36	162.32	231
R12	50.00	106.32	172.91	227
R13	55.51	113.3	172.03	230
R14	50.52	115	174.56	232

Table 5. Data for the *In-vitro* bioadhesion studies of the Esomeprazole buccal patch

Formulation Code	Mucoadhesive Strength (g)	Force of Adhesion (N)	Mucoadhesive Time (min)
R1	77	0.75	350
R2	95	0.93	400
R3	91	0.89	390
R4	76	0.74	320
R5	115	1.127	470
R6	110	1.078	465
R7	105	1.029	370
R8	92	0.901	394
R9	91	0.89	390
R10	75	0.73	340
R11	92	0.90	395
R12	74	0.72	335
R13	76	0.74	345
R14	104	1.019	456

3.5.9 *In-Vitro* Drug Release Studies

As evident from the diverse nature of dissolution profiles, the influence of polymer seems to be vital in regulating the drug release. All the formulations showed a drug release of 54 to 87 % at the end of 8 hr study period (Table 6). Formulation R5 showed the highest drug release of 87 % at the end of 8 hrs.

3.5.10 *Ex-Vivo* Drug Permeation Studies

Ex-vivo drug permeation studies were carried out on all formulations according to the procedure mentioned in section 4.4.6. All the formulations showed a drug permeation of 76 to 91 % at the end of 8 hrs study period (Table 6) Formulation R6 showed the highest drug permeation of 91 % at the end of 8 hrs.

3.6 Optimization Studies

Optimization was carried out by using central composite design by taking concentration of thiolated Chitosan, PVP K30 and Glycerol as the independent variables and mucoadhesive strength, mucoadhesive time as the dependent variables. The central composite design was chosen since it is useful in analyzing the relationship between multiple variables with reduced number of experimental runs. Fourteen formulations were developed as per runs given in the optimization design. The results were fitted to quadratic and 2F1 model of regression as it showed the maximum values of R^2 and model sum of squares.

From the numerical optimization results, solution 1 was selected as the optimized formula since it had high desirability and coded as OPTI-TC for the preparation of optimized bilayered Esomeprazole buccal patch. The generated optimization study was conducted to study the constraint on the design space and vulnerability of the experimental model. This is important, since it suggests factor, responses and the goal for each variable with respect to the measured response. The results shown in the Table 7 confirmed the closeness of the observed result with that of the predicted results. It was observed that the response was almost similar to response predicted by the Design Expert Software.

Comparison of the Optimized Formulation with a Buccal Patch Containing Chitosan

In order to prove that the thiolated chitosan has superior mucoadhesive properties, the final optimized formulation was compared with a buccal patch containing the same concentration of chitosan as the mucoadhesive polymer. All

other ingredients were used at the same concentration as that of the optimized formulation. The patch was subjected to the same evaluation studies as that of designed runs and the results are reported in Table 8.

Table 6. Data for the *In-vitro* release and *ex-vivo* drug permeation studies

Formulation Code	Cumulative % Drug Release	Cumulative % Drug Permeation
R1	70	81
R2	75	84
R3	73	88
R4	70	82
R5	87	89
R6	82	91
R7	79	89
R8	74	87
R9	72	83
10	54	76
R11	73	84
R12	65	78
R13	67	79
R14	81	89

Table 7. Optimized Foramulation OPTI- TC (predicted value v/s actual value)

Number	Thiolated polymer (%)	Film improver (%)	Plasticizer (b b%)	Predicted value	
				Mucoadhesive strength (gm)	Mucoadhesive time (min)
Predicted value	2.5	5	10	109.3	472.8
Actual value	2.5	5	10	111.6	475

Table 8. Comparison between OPTI-TC and Chitosan

Parameters	OPTI-TC	Chitosan
Weight variation (mg)	150	103
Patch thickness (mm)	0.632	0.292
Folding endurance (fold)	346	268
Surface pH	7.2	7.2
Drug content (%)	94	91
Swelling index (%)	235	311
Mucoadhesive strength (g)	111.6	53
Force of Adhesion (N)	1.094	0.52
Mucoadhesive Time(min)	475	310
% Drug release	93	75

4. CONCLUSION

The present study was conceptualized to develop a transmucosal bilayered mucoadhesive drug delivery system for the proton pump inhibitor drug Esomeprazole. The main interest in such a dosage form was to provide direct entry of the drug into the systemic circulation to avoid extensive first pass metabolism and impart a prolonged effect, thereby providing increased therapeutic action. The bilayered mucoadhesive buccal patch of esomeprazole were prepared and evaluated for all the parameters and other evaluation studies specifically designed for mucoadhesive buccal patch formulations. The mucoadhesive bilayered buccal patches were successfully prepared by solvent casting method using

different concentrations of thiolated chitosan with ethyl cellulose as an impermeable backing layer. The results were found to be within the official limits. *In-vitro* drug release studies revealed that the formulations with OPTI-TC were able to sustain the drug release upto 8 h.

The results of *Ex-vivo* drug permeation studies showed that as the concentration of the polymer increased the drug permeation increased. Buccal delivery of Esomeprazole was found to be a promising route for increasing the overall effectiveness of the drug. This drug delivery can thus be used as an alternative to the conventional drug delivery systems containing esomeprazole which have the drawback of degradation of drug when it comes in contact with the acidic contents of the stomach. The mucoadhesive polymers chitosan and thiolated chitosan were found to be more suitable for mucoadhesion. Thiolated chitosan was found to be the best formulation in terms of mucoadhesive strength, swelling index, drug release and drug permeation. The impermeable backing layer of ethyl cellulose was successful in providing a unidirectional release of the drug decreasing the loss of the drug by wash out of saliva to a considerable extent.

5. REFERENCES

1. Pranshu Tangri NV, Satheesh Madhav, Shaffi Khurana. Mucoadhesive Drug Delivery: Mechanism and Method of Evaluation. *IJPBS* 2011; 2(1): 459-67
2. Rossi S, Sandri G, Caramella CM. Buccal Drug Delivery: A Challenge Already Won. *Drug Discovery Today: Technologies* 2005; 2(1): 51-65.
3. Schnurch AB, Steininger S. Synthesis and Characterization of Mucoadhesive Thiolated Polymer. *Int J Pharm* 2000; 194: 239–47
4. Kast CE, Frick W, Losert U, Schnurch AB. Chitosan-Thioglycolic Acid Conjugate: A New Scaffold Material for Tissue Engineering. *Int J Pharm* 2003; 256: 183–89.