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Research Article

Fast Dissolving Sublingual Films Containing Sumatriptan Alone and Combined with Methoclopramide: Evaluation *in Vitro* Drug Release and Mucosal Permeation

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ABSTRACT

Background: Sumatriptan succinate is a 5-HT_1 receptor agonist which is used in the treatment of migraine. It shows low bioavailability (15%) due to high hepatic first pass metabolism. The present work intended to formulate mucoadhesive sublingual films of sumatriptan combined with metoclopramide and sumatriptan alone with the objective of improving the therapeutic efficacy, patient compliance, and bioavailability.

Methods: The sublingual films were formulated by solvent casting technique using mucoadhesive polymer of hydroxypropyl methylcellulose and propylene glycol as plasticizers. This study was also designed to evaluate the physicochemical and mucoadhesive characteristics of the films. The films were evaluated for their mechanical strength, folding endurance, drug content uniformity, swelling, *in vitro* residence time, *in vitro* release, *in vitro* bioadhesion, and *in vivo* mucoadhesion.

Results: They showed good appearance and elasticity. The best drugs of polymer ratio were S_3 (1:2) and SM_2 (2.7:1:8). The film of S_3 and SM_2 showed 10.6 and 11.01 mg weight, 2.2 and 22.5 µm thickness, 300 folding endurance, 55.9 and 100% content uniformity, respectively. The Differential Scanning Calorimetry (DSC) showed no stable sample of sumatriptan and metoclopramide in the drug loaded films and revealed amorphous form and transition of hydrate to anhydrous form for metoclopramide. The results showed that the films prepared were fast dissolving. The films (sumatriptan combined with metoclopramide and sumatriptan alone) exhibited very good mucoadhesive properties and shorter retention time (15-30 s).

Conclusion: The formulations were found to be suitable candidates for the development of sublingual films for therapeutic uses.

Introduction

Fast dissolving films have recently obtained great importance in the pharmaceutical industry due to their specific characteristics and special advantages over other forms such as no need of water for disintegration, accurate dosing, rapid onset of action, ease of transportability, ease of handling, pleasant taste, and enhanced patient compliance.[1] Fast dissolving film is a type of drug delivery system which upon placing in the oral cavity, promptly disintegrates and dissolves to release the drug for oromucosal and intragastric absorption. This dissolution occurs without chewing or intake of water.[2] This technology has developed over the past few years from the confection and oral care markets in the form of breath strips. Further, they can be utilized for local effect.[3,4] Holding a beneficial drug dosing method, this type of technology is suggested for special population groups such as pediatric, geriatric, bedridden patients, mentally ill patients, along with general population. The sublingual mucosa is comparatively permeable for having a thin membrane and big veins, which renders swift

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absorption and immediate bioavailability of drugs owing to high blood flow.[4,5] When the fast dissolving film is taken sublingually, it is rapidly absorbed. This lastly causes the rapid commencement of drug action, while the first pass metabolism of the drug is inhibited.

Migraine is recognized as one of the ten most disabling disorders worldwide, and in spite of recent developments in the management of migraine, its diagnosis and treatment is still open to research.[6] Epidemiological investigations on the migraine show that nausea has been experienced by a large majority of patients (>90%) during a migraine attack. Likely, most migraine-affected patients (almost 70%) have vomited at some time all along an attack, so they avoid excessive intake of liquid.[7] Moreover, the migraine-affected individuals have indicated a reduction in their physical abilities; so they would get benefit of effective treatment that would assist them in continuing their physical abilities as fast as possible. Of the recent generation of anti-migraine drugs, sumatriptan succinate (Sum) is known as a triptan derivative which is a serotonin agonist applied as a main drug in the migraine therapy. The main disadvantage of this drug is attributed to its low oral absolute bioavailability (only 15%) which might be due to its extensive first pass metabolism (nasal sumatriptan migraine).[8]

For a long period of time, metoclopramide hydrochloride (Met) has been used in the treatment of nausea linked to acute migraine. Besides its antiemetic properties, metoclopramide involves in relieving the gastric stasis and holds a competence in enhancing the absorption of other analgesics.[9]

Thin film drug delivery uses a dissolving film to administer drugs via absorption in the mouth (buccally or sublingually). The film is prepared using the hydrophilic polymers (e.g., Hydroxypropyl methylcellulose) which dissolves on the tongue or buccal cavity in a no while. And upon contacting with liquid, the drug is delivered to the systemic circulation through dissolution.[10]

Hydroxypropyl methylcellulose (HPMC) polymer is non-toxic, non-irritant and void of leachable impurities. It should have good wetting and spreadability characteristic. HPMC shows enough peel, shear and tensile strengths. Moreover, it is readily accessible and cheap. Accordingly, film strips should be tough adequately so that there would not be any damage while handling or transportation.[11]

Plasticizers (e.g., propylene glycol) help to improve the flexibility of the film and to decrease its brittleness. Plasticizers improve the film properties by reducing the glass transition temperature of the HPMC polymer. The mechanical characteristics of the films are also improved by the addition of plasticizers.[12] In this work, mucoadhesive sublingual films made of Sum and Met were developed using HPMC polymer.

Materials and Methods

Materials

Sumatriptan succinate (Sum), metoclopramide hydrochloride (Met), HPMC (E-15), ethanol, dichloromethane, buffer phosphate (pH 6.8), sodium chloride, potassium chloride, sodium sulfate, ammonium acetate, urea, lactic acid, and agar were obtained from Merck (Darmstadt, Germany). All solvents were of analytical grade.

Sum preparation method

Sublingual films of Sum were prepared by solvent casting technique using a film forming mucoadhesive polymer. HPMC was accurately weighed (200 mg) and dissolved in 2.5 ml of ethanol. The beaker holding polymer and ethanol was put aside for 5 min in order that the polymer could be swelled. Then, 2.5 ml of dichloromethane was added to the abovementioned polymer solution and the dispersion was stirred. Then, one drop of propylene glycol (0.030 g) was added to the polymer solution. At first, Sum drug was accurately weighed (50, 66.7, and 100 mg) and then dissolved in 1 ml of water in another beaker (Table 1). The drug solution was then added to the polymer solution and was mixed thoroughly with the aid of a magnetic stirrer.

Table 1. Sumatriptan succinate alone and combined with methoclopramide hydrochloride films prepared by solvent casting method with different drug and polymer ratio.

Formulation code	Drugs to polymer ratio	Sumatriptan succinate (mg)	Metoclopramide hydrochloride (mg)	Water (ml)	HPMC (mg)	Dichloromethane (ml)	Ethanol (ml)	Propylen glycol (g)
S_1	1:4	50	-	1	200	2.5	2.5	0.03
S2	1:3	66.7	-	1	200	2.5	2.5	0.03
S3	1:2	100	-	1	200	2.5	2.5	0.03
SM_1	2:1:8	50	25	2	200	2.5	2.5	0.03
SM_2	2.7:1:8	66.7	25	2	200	2.5	2.5	0.03
SM_3	4:1:8	100	25	2	200	2.5	2.5	0.03

The solution was wholly poured into the glass Petri dish placed over a flat surface. Afterwards, an inverted funnel was placed over the dish to prevent sudden evaporation. The mould containing the polymeric solution of drug was kept for 12 h at room temperature to dry. After drying, the films were observed and checked for probable imperfections after their removal from the moulds. They were then coated with wax paper and kept in desiccators until the evaluation tests were performed.

Method of preparation of films of SUM alone and combined with Met

Sublingual films of Sum and Met combination were prepared by solvent casting technique. At first, Sum drug was accurately weighed with a different drug to polymer ratio (50, 66.7 and 100 mg) and then dissolved in 2 ml of water in beaker. In second step, Met drug was weighed with constant amount (25 mg) and added to the Sum solution beaker (Table 1). The drugs' solution was added to the polymer solution and mixed thoroughly with the help of a magnetic stirrer. Sum and Met combined films were prepared under the same conditions (Sum film method).

Characterization of buccoadhesive films

Appearance of the films was appraised by observing the color, elegance, stickiness, and texture.

Weight uniformity of films

Six films of the size 1×1cm^2 for each formulation were individually weighed in a digital balance (Sartorius, Germany) and the weight variation was calculated.

Thickness uniformity of the films

Each film was measured for its thickness by using digital vernier calipers at five different points (center and four corners) of the film and the average was calculated (Mitutoyo, Japan).[13]

Folding endurance

The folding endurance of each film was determined by counting the number of times the film (size 1x1 cm^2) could be folded repeatedly (folded or broken up to 300 times), which was regarded reasonable to reveal good film properties.[14]

In vitro swelling studies

The swelling rate of films was evaluated by placing the film in a 2% (w/v) agar gel plate. Initial diameter of film (1x1 cm^2) was determined in agar gel plate and incubated at 37±1°C (D_1). Then at regular intervals (up to 1 h), swollen film diameter was re-measured (D_2) and the swelling index was calculated by the formula as follows:[4,14]

$$Swelling\ index = D_2 - D_1 / D_1 \quad \text{Eq.(1)}$$

Moisture content loss and moisture absorption

The films were accurately weighed and kept in desiccators containing: a) anhydrous calcium chloride and b) 100ml of saturated solution of aluminum chloride, which maintains 76% and 86% humidity, respectively (RH). After 3 days, the films were taken out to be weighed. The moisture content (%) was calculated through its moisture loss (%) according to the formula:[14]

$$Moisture\ content\ (\%) = Initial\ weight - Final\ weight / Initial\ weight \times 100 \quad \text{Eq.(2)}$$

Also, the moisture absorption was calculated using the formula:[14]

$$Moisture\ absorption\ (\%) = Final\ weight - initial\ weight / Initial\ weight \times 100 \quad \text{Eq.(3)}$$

Drug content, content uniformity and production yield

The films (five samples of each film) were analyzed for the content uniformity by dissolving 1×1cm^2 films in 10 ml phosphate buffer, pH 6.8, with simultaneous shaking for several hours. The absorbance of the solution (Sum and Met) was measured by UV spectrophotometer at 227.4 (Sum) nm and 272.4 (Met) nm. The production yield of the films was determined by calculating the last weight of the films obtained to the initial weight of the raw materials. All experiments were performed in triplicate.

Differential Scanning Colorimetry (DSC)

The physical state of drug in the microspheres was analyzed by Differential Scanning Calorimeter (Shimadzu, Japan). The thermograms were obtained at a scanning rate of 10 °C/min conducted over a temperature range of 25-300°C.

Ex vivo mucoadhesion time

The present study was conducted according to the Guide for the Care and Use of Laboratory Animals of Tabriz University of Medical Sciences, Tabriz-Iran (National Institutes of Health Publication No 85-23, revised 1985). E*x vivo* mucoadhesion test was performed on the selected film. The disintegration medium comprised 50 ml phosphate buffer, pH 6.8, maintained at 37°C. A segment of sheep buccal mucosa with a length of 3 cm was applied as glue to the surface of a glass slab and then was vertically attached to the disintegration apparatus (Erweka, Germany).[15] The mucoadhesive films were hydrated from one surface and then were contacted with the mucosal membrane. The glass slab was vertically fixed to the apparatus so that it could readily move up and down; thus, the

film could immerse in the buffer solution at the lowest point while it could be out at the highest point. The time required for either utter erosion or detachment of the films from the mucosal surface was noted down. The experiment was performed in triplicate.

Bioadhesion strength

The tensile strength asked for detaching the bioadhesive films from the surface of mucosa was claimed as a measure of the bioadhesive performance. The apparatus was locally gathered and constructed. The device principally held a two-arm balance (Figure 1). The mucoadhesive forces of films were determined by means of the mucoadhesive force-measuring device,[15] using the tissue cut from buccal mucosal area of sheep. The pieces of mucosa were stored frozen in phosphate buffer, pH 6.8, and thawed to room temperature before use. At the time of testing, a section of mucosa was secured to the upper glass vial (C) by a cyanoacrylate adhesive (E). The diameter of each exposed mucosal membrane was measured to be 1.5 cm. The vials were balanced and preserved at 37°C for 10 min. Next, one vial with a section of tissue (E) was attached to the balance (A) and the other vial was stabilized atop a pan, height-(F) and weight-adjustable (B). To expose the tissue on this vial, a constant amount of films (D) was applied. The height of vial was adjusted so that adherence to the mucosal tissues of both vials was made possible for the films (Figure 1).

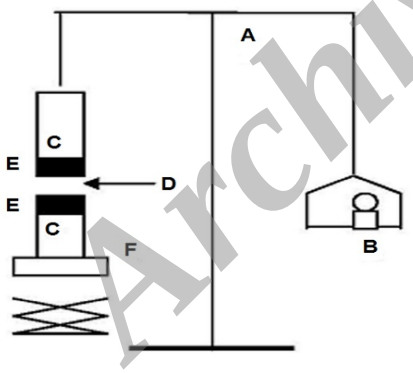

Figure1. Bioadhesive force measuring device: (A) modified balance; (B) weight; (C) glass vial; (D) discs/films; (E) tissue; (F) height-adjustable pan.

In no time, a constant force of 0.5 N was attended for 2 min to ascertain that a close contact could occur between the tissues and the samples. The vial was then moved upwards at a steady speed and associated with the balance. Next, weights were added at a regular rate to the pan on the other side of the modified balance of the utilized device till the time two vials were separated. During measurement, 150 µl of phosphate buffer (pH 6.8) was evenly spread onto the surface of the test membrane. The bioadhesive force, mentioned as the detachment stress in g/cm^2, was obtained from the minimal weights which detached the tissues from the surface of each formulation through the following equation:[15]

$$Detachment\ Stress\ \left(g/cm^2\right) = m / A \qquad \text{Eq.(4)}$$

Where m is the weight added to the balance in grams and A is the area of tissue exposed. Measurements were repeated three times for each of the films. All the above three experiments were conducted in triplicate.

Permeation studies

The *in vitro* permeation study of the Sum alone and the Sum and Met combined films through the buccal mucosal area of sheep was performed using Franz diffusion cell at 37 ± 0.2°C. Freshly obtained buccal mucosa was localized between the donor and receptor compartments so that the smooth mucosal surface faced the donor compartment. The films were positioned on the mucosa and the compartments were clenched together. The donor compartment was filled with 3 ml simulated saliva, pH 6.8 (sodium chloride 4.50 g, sodium sulfate 0.30 g, potassium chloride 0.30 g, urea 0.20 g, ammonium acetate 0.40 g, lactic acid 3 g, and distilled water up to 1,000 mL, adjusting pH of the solution to 6.8 by 1 M NaOH solution). The receptor compartment was filled with 22-25 ml phosphate buffer (pH 7.4) and stirred with a magnetic bead at 700 rpm.[16]

Three milliliters of samples were withdrawn at predetermined time intervals and analyzed for drugs at 228 (Sum) nm and 272.4 (Met) nm.

In vitro release studies

In vitro release studies were carried out using an incubator shaker at 37 ±0.5°C, at a stirring speed of 50 rpm. Films were fixed on glass slides and placed at the bottom of beaker. The studies were performed for all formulations (Sum and Sum and Met combination) in triplicate, using 50 ml (37°C, 50 rpm) of isotonic phosphate buffer (pH 6.8) as the dissolution medium. An aliquot of 3 ml sample was withdrawn at regular intervals and replaced immediately with an equal volume of fresh phosphate buffer (pH 6.8). Samples were then analyzed at 227.4 (Sum) nm and 272 (Met) nm with UV spectrophotometer.

Histopathological evaluation of mucosa

Histopathological evaluation of tissue incubated in phosphate buffer, pH 6.8, was compared with that treated with sublingual mucoadhesive films delivered from mucoadhesion time test. The tissue was fixed with 10% formalin, routinely processed, and embedded in paraffin. On glass slides, the sections were cut and stained with hematoxylin and eosin. A pathologist, blinded to the study, detected any damage to the tissue and examined the sections

on the light microscope.[16]

Results

In vitro characterization studies

Sublingual films containing Sum combined with Met and Sum alone were prepared by solvent casting technique. The physicochemical and mucoadhesive characteristics of all the formulations are shown in Table 2.

All of the formulations were smooth, flexible, colorless (transparent), non-sticky, and elegant in appearance, except for SM_3 film (dark color) (Figure 2). The cut films were 1x1 cm^2 in size. The weight and thickness of Sum alone and SM films were in the range of 4.4-10.6, 9.5-14.02 mg and 62.6-83.2, 110.3-243 µm, respectively (Table 2).

The flexibility of the Sum alone and SM films which was required for their easy handling was given by their folding endurance ranged from 89 to 300 times . All the S_1 to S_3 and SM_1, SM_2 films resisted breakage upon folding them for more than 300 times at the same place (Table 2). Hence it was taken as the end point. The values were observed to be optimum to reveal good film properties. The content of Sum alone films were in the range of 0.939-1.811 and of Sum combined with Met films were in the range of 0.23-1.03 mg/cm^2 (Sum) and 0.57-0.59 mg/cm^2 (Met), respectively.

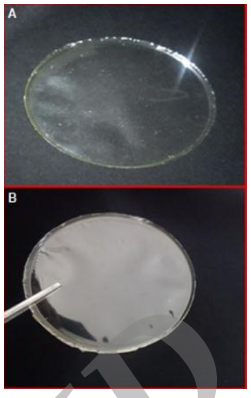

Figure2. Optical microscopic photograph of mucoadhesive films of (A) sumatriptan succinate alone (B) sumatriptane combined with metoclopramide.

Table2. Effect of drug to polymer ratio on physicochemical characteristics and mucoadhesivity sumatriptan alone and combined with methoclopramide films.

Variables	Formulation code					
	S_1	S_2	S_3	SM_1	SM_2	SM_3
Drugs : Polymer ratio	1: 4	1:3	1:42	2:1:8	2.7:1:8	4:1:8
Weight variation (mg ± SD)	4.4±1.2	6.8 ± 1.0	10.6 ± 1.4	9.50 ± 2.50	11.01 ± 0.80	14.02 ± 1.20
thickness (µm± SD)	62.6±1.0	73.0±1.0	83.2±0.9	110.3 ± 0.03	123 ± 0.007	243 ± 0.05
Folding endurance (n±SD)	>300	>300	>300	>300	>300	89 ± 12
Drug content (mg/cm^2±SD)	0.939 ± 0.22	1.115 ± 0.11	1.811 ± 0.4	**Sum:** 1.03 ± 0.33	1.15 ± 0.60	0.23 ± 0.02
				Met: 0.59 ± 0.50	0.60 ± 0.80	0.57 ± 0.74
Content uniformity (%±SD)	78±2.36	85±2.11	90±5.91	**Sum** 100 ± 0.02	98.59 ± 0.02	96.63 ± 0.02
				Met: 100 ± 0.01	99.9 ± 0.01	95.78 ± 0.06
Production Yield (%±SD)	85.5±29.30	98.02±11.80	100±8.30	99.10 ± 9.1	100 ± 6.60	99.40 ± 2.30
Absorbed moisture (% ±SD)	7.32±0.65	5.44±0.21	4.96±0.38	0.79 ± 0.44	1.06 ± 0.95	4.52 ± 1.79
Loss moisture (% ± SD)	1.89±0.27	5.38±0.01	6.20±0.07	0.79 ± 0.04	0.73 ± 0.08	2.85 ± 0.66
pH surface (±SD)	5.9±0.30	6.2±0.40	6.0±0.40	6.6 ± 0.2	6.8 ± 0.1	6.9 ± 0.1
Swelling index (%±SD)	25.74±0.05	18.88±0.05	18.38±0.05	19.85 ± 0.02	20 ± 0.01	14.18 ± 0.02
Mucoadhesive strength (g/cm^2±SD)	138.85±5.7	135.35±21.8	128.44±2.8	134.8 ± 13.1	127.61 ± 8.4	120.50 ± 6.6
Residence time (Sec±SD)	15±0.02	15±0.01	15±0.00	25 ± 0.14	20 ± 0.81	30 ± 0.26

Though there is minor change in the loss of drug (Sum and Met) among the formulations, more uniformity was seen in films in Table 2, 78-90% (Sum alone film) and 96.63-100% (SM film, Sum

combination with Met), 95.78-100% (SM' film, Met combination with Sum).

The percentage of moisture absorption was shown to range between 4.96±0.38 and 7.32±0.65 for Sum alone films and 0.79±0.44 to 4.52±1.79% for Sum combined with Met films. The moisture losses, 1.89-6.20% (for Sum alone) and 0.79-2.85% (for SM_1 to SM_3 films), are shown in Table 2. Hence, the high moisture absorbing capacity was detected in S_1 (7.32%) and SM_3 (4.52%), and more moisture loss was observed in S_3 (6.20%) and SM_3 (2.85%).

All formulations were of pH 5.9-6.2 and 6.6-6.9 for Sum alone and Sum combined with Met films, respectively and it may be concluded that the films are safe and non-irritating to oral mucosa (Table 2).

Pure Sum exhibited a sharp melting exothermic and endothermic peak around 173.27°C and Met was melted at 184.59-208.75°C (Figure 3).

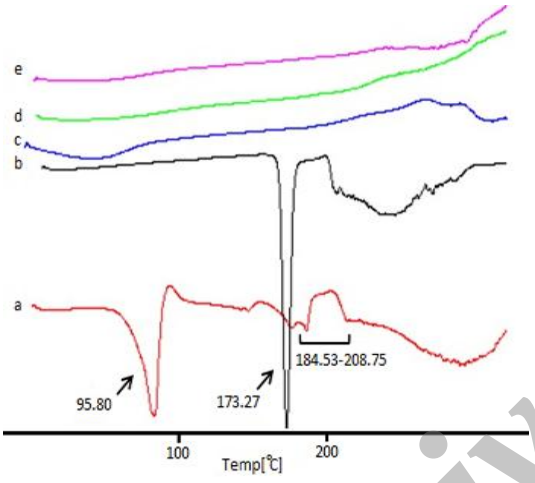

Figure 3. DSC thermogram of sumatriptan (a) metoclopramide (b)HPMC (c) S_3 (100 mg sumatriptan) (d) and (e) SM_2 (66.7 mg sumatriptan and 25 mg metoclopramide).

Sum drug fusion peak, however, for the SM film formulations was disappeared in comparison with pure drug (Sum).

Pure Met monohydrate has a very high melting point (184.59-208.75°C). The endothermic peak at around 95.80°C is probably due to the transition of drug to the anhydrous form via loss of one mole of water.

Ex-vivo mucoadhesive characterization studies

In vitro residence time determined the period of adhesion of the Sum alone (15 s) and Sum combined with Met films to the mucosa and ranged 25-30 s. All films showed low diameter swelling and the recorded swellings after 2 h were 18.38-25.74% (for Sum alone films) and 14.18-20% (for Sum combined with Met films).

The results of *in vitro* bioadhesive strength study are shown in Table 2. The bioadhesive properties were affected by the concentration of the bioadhesive polymer (HPMC). S_1 and SM_1 films 4:1 and 2:1:8 ratios (drug/s to polymer) indicated the highest mucoadhesivity (138.85±5.7 and 134.81±13.1 g/cm^2, respectively). S_3 and SM_3 Formulations containing 42:1 and 4:1:8 ratios (drug/s to polymer) showed the lowest mucoadhesivity (128.44±2.8 and 120.5±6.6 g/cm^2, respectively).

In vitro release studies

Accordingly, Figure 4 and Table 3 show that the initial Sum drug releases ($Rel_{0.25}$) for the SM_1 to SM_3 formulations were low (13.47%, 15.41% and 15.86%, respectively) and $Rel_{0.5}$ was 103.10%, 105.82% and 112.57%, respectively. Moreover, Met drug release of SM' (SM) films shows that high burst effects for SM'$_1$ to SM'$_3$ formulations were high (109.87%, 116.63% and 108.54%, respectively) and $Rel_{0.5}$ was high, too (106.67%, 110.82% and 110.8%, respectively) (Figure 4B).

Table 3. Amount of drug release and comparison of various release characteristics of sumatriptan alone and combined with methoclopramide from different film formulations.

Formulation code		[a]$Rel_{0.25}$ (%±SD)	[b]$Rel_{0.5}$ (%±SD)	[c]Rel_8 (%±SD)	[d]DE (%±SD)	[e]$t_{50\%}$ (min±SD)	[f]f1
S1		19.41±1.01	103.66±14.30	105.14±3.36	101.53±10.45	35.20±4.23	15.46
S2		18.47±0.86	104.65±5.91	104.92±10.66	101.73±8.79	36±8.12	2.29
S3		20.99±0.56	102.21±9.22	104.39±2.89	101.93±7.42	36.40±5.63	4.40
SM1	**S1**	15.86±3.27	103.10±2.90	111.59±15.30	100.26±12.32	49.08±5.47	0
	M1	109.87±3.71	106.67±3.04	110.75±4.71	93.85±10.24	138.16±14.52	0
SM2	**S2**	15.41±2.72	105.82±3.73	106.45±4.08	104.61±9.87	49.35±4.12	4.94
	M2	116.63±1.72	110.82±1.54	117.80±5.06	106.37±12.14	143.71±14.78	14.80
SM3	**S3**	13.47±1.34	112.57±4.78	123.86±5.44	100.32±13.12	54.22±4.78	2.46
	M3	108.54±7.03	110.80±1.59	118.23±4.42	116.60±14.39	71.23±6.25	23.94

a Rel0.25 = amount of drug release after 15 min; b Rel0.5 = amount of drug release after 30 min; c Rel8 = amount of drug release after 8h ;dDE = dissolution efficiency; et 50% = dissolution time for 50% fractions; f f1 = Differential factor (0<f1<15), SM1(1:2:8 ratio) is selected as reference formulation.

The release of Met drug from SM films was faster than the release of Sum drug from SM films

(p<0.05). During dissolution, HPMC containing films swelled forming a gel layer on the exposed mucous surfaces.

The release of drug from Sum combined with Met films with an increase in Sum concentration, and the interaction between the polymer and drug increased with the formation of a closer network, showed an increase in the diffusion of drug from the films (Figure 4A & 4B). The reason for the burst release ($Rel_{0.25}$) could be due to the presence of some pores and channels of polymer near to the surface of films. When water-soluble drugs (Sum and Met) did not show a tendency to migrate or remove air bubbles, therefore drug concentration in the films is increased and burst effect is induced.[12] The pores present in HPMC polymer act as channels for the entrance of liquid medium through the film surface and cause it to swell. Hydrogen bond between the hydroxyl groups of HPMC moiety and mucous surface decreases its porosity and permeability. Thus, if the ratio of drug to polymer is varied, the rate of release of drug could be controlled. Permeation of dissolution medium into the films is facilitated due to the high swelling action of polymer which leads to the transport of the available drug via more medium.

Permeation studies

Figure 5 represents the comparison of permeation of Sum alone films and Sum combined with Met films through the buccal mucosa for formulations containing a different drug-to-polymer ratio.

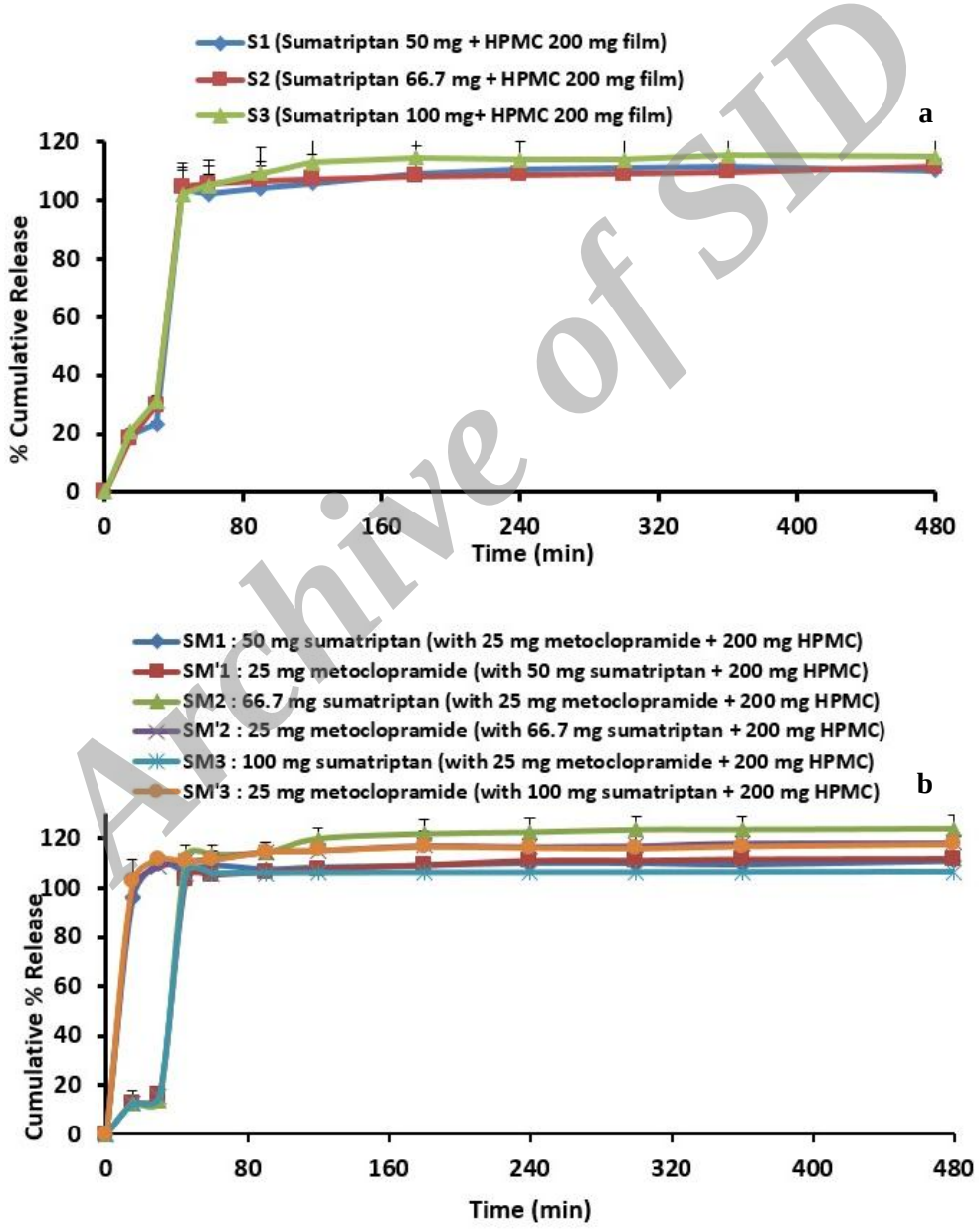

Figure 4. (a) Cumulative percent release of sumatriptan alone (b) and combined with metoclopramide from films prepared with different drug/s to polymer ratios.

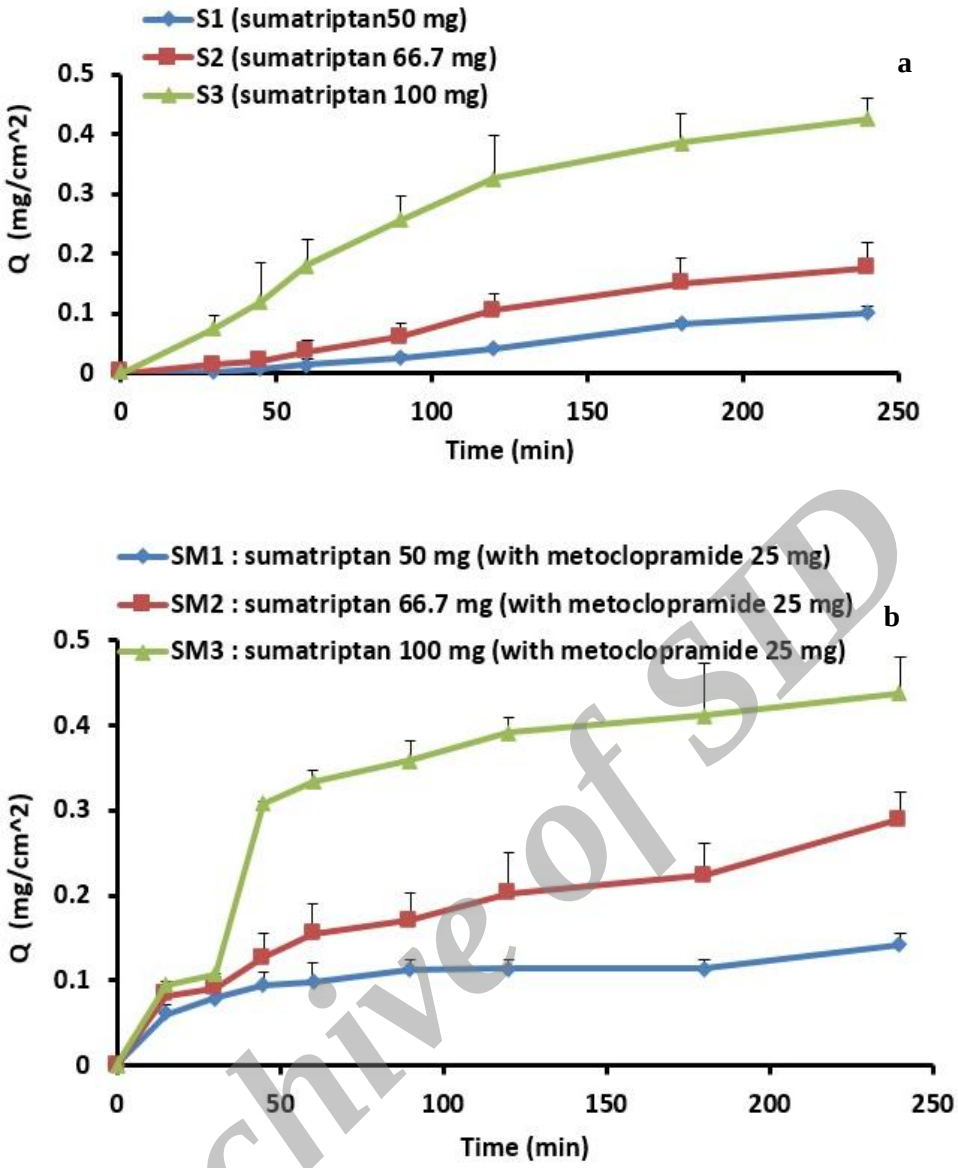

Figure 5. (a) Amount of sumatriptane alone release per unit surface area after 4 h (b) Amount of sumatriptan combined with metoclopramide release per unit surface area after 4 h through buccal mucosal area of sheep.

Slopes of the linear portion of the release profiles were calculated. These slopes depicted the rate of release or flux of Sum alone and Sum combined with Met films from different formulations (Table 4). The highest fluxes and regression coefficients for S_3 and SM_3 formulations were 0.0017 mg/cm^2.min, 0.9134 (Sum alone) and 0.0008 mg/cm^2.min, 0.9314 (Sum combined), 0.0016 mg/cm^2.min, and 0.8839 (Met combined), respectively.

Discussion

The required quantity of mucoadhesive polymer, in this technique, was treated with required quantity of solvent system and in a vortex state to allow the polymer to swell.

After swelling, mixture was treated with plasticizer and vortexed. Finally drug was dissolved in small volume of solvent and poured to the polymer solution and mixed very well.[17]

The variation in weight and thickness among the formulations may be the effect of difference in the concentration of drugs (Sum and Met) used in the films (Table 2).

Mucoadhesive polymer (HPMC) possesses numerous hydrophilic functional groups, including hydroxyl and carboxyl for example. These groups authorize hydrogen to bond with the substrate (mucus) and swell in aqueous media. Further, they permit maximal exposure of potential anchor sites. Besides these, swollen HPMC exhibits the maximum distance among their chains resulting in the increased chain flexibility and effective penetration of substrate. HPMC (with low molecular weights) would form loose gels or dissolve quickly. Chain flexibility is indispensible

for the interpretation and entanglement of mucoadhesive HPMC polymer. As water-soluble polymer becomes cross-linked, the mobility of distinct polymer chains decreases and accordingly the influencing length of the chain, penetrating into the mucus layer, decreases. It in turn lowers the bioadhesive strength.[18, 19]

Table 4. Flux or amount of drug release per unit surface area after 4 h, intercept and regression coefficient for different formulation and comparison of various release characteristics of sumatriptan alone and combined with methoclopramide from different film formulations.

Formulation code		Flux (mg/cm^2min)	Intercept (mg/cm^2)	r^2
S_1		0.0005	-0.0101	0.975
S_2		0.0008	-0.011	0.977
S_3		0.0017	0.0699	0.913
SM_1	S_1	0.0002	0.0812	0.872
	M_1	0.0004	0.0773	0.940
SM_2	S_2	0.0006	0.1018	0.970
	M_2	0.0006	0.1674	0.959
SM_3	S_3	0.0008	0.2964	0.931
	M_3	0.0016	0.1729	0.884

As the drug was uniformly dispersed in the matrix of the polymer, a significantly convenient volume of drug was loaded in all the formulations. The loss of drug could be related to its aqueous insolubility. Sum and Met are water-soluble and does not commence settling down from medicated solutions when dispersed for removal of air bubbles. Hence the solutions were casted as films containing complete amount of drugs.

Percentage of moisture absorption is correlated with the capacity of excipients to absorb water in vapor form. The HPMC polymer used is a hydrophilic polymer. It is hypothesized that the initial moisture content acts as a determinant factor in the moisture absorption.

Other films show initially high moisture content as is evinced by percentage of moisture lost. There is an inverse relationship between these two parameters; the higher the percentage of moisture lost, the lower the moisture absorbed and vice versa.[13]

The acidic or alkaline pH may render irritation to sublingual mucosa and may affect the release of drug and degree of polymer hydration. Therefore the surface pH of sublingual film was determined to optimize both drug release and mucoadhesion. The surface pH of all formulations was within ±0.5 units of the buccal pH (6.6-6.9) and hence no mucosal irritations were expected and ultimately patient compliance was achieved.[20]

It is clearly observed from the thermogram of the SM_1, SM_2, and SM_3 films (Figure3) that the drugs (Sum and Met) peak has been disappeared. However in the thermogram of SM_1, SM_2, and SM_3, the endothermic peak corresponding to the drugs' melting point was absent, suggesting the amorphous state of the drugs (Sum and Met).

The DSC analysis of films revealed a significant change in the melting point of Sum and Met drugs, indicating the modification or interaction between the drug and polymer (Figure 3). The integrity of Sum alone and Sum combined with Met films was early lost following the rapid uptake. Sum and Met drugs, possessing water-soluble molecules and permitting more water influx, result in quicker dissolution and erosion from mucosal surface. HPMC is a hydrophilic polymer and may have more affinity towards mucin which comprises 95% water. This may be the reason for longer residence time (integrity of the films is shorter). Moreover, as reported by some previous studies, the enhanced erosion rate was observed with the non ionic polymers such as HPMC. The swelling behavior was assessed through measuring the diameter swelling. For SM films attended for sublingual (local) therapy, the contact area was determined to be as large as possible, a demand that is indispensible to be balanced with patient compliance. Extra increase in film diameter may result in the discomfort and/or dislodgment of the swollen film (lower than 20% swelling for Sum combined with Met films).

Increase in concentration of drug (Sum) decreases the bioadhesive strength of formulations. The bioadhesive force depends on the molecular weight and swelling behavior of the polymers, and contact time with the mucus. The bioadhesion characteristics were affected by the type and ratio of the bioadhesive polymers. The highest detachment force was observed with the formulation S1 and SM1. The high bioadhesive force may be due to the formation of secondary bioadhesion bonds with mucin and interpenetration of polymeric chains in the interfacial region.

The release profiles for all films are illustrated in Figure 4. Films with high content uniformity or high drug entrapment showed a faster dissolution rate. As more drugs are released from the films, more channels and pores are probably produced, contributing to the faster drug release rates. Figure 4 shows that the initial Sum drug releases ($Rel_{0.25}$) for the S_1 to S_3 formulations were relatively low (19.41%, 20.99%, and 18.47%, respectively) and $Rel_{0.5}$ were 103.66%, 102.21%, and 104.65%, respectively. Accordingly, Figure 4 shows that the initial Sum drug releases ($Rel_{0.25}$) for the SM_1 to SM_3 formulations were low (15.86%, 15.41% and 13.47%, respectively) and $Rel_{0.5}$ were 103.10%, 105.82%, and 112.57%, respectively. Met drug release of SM' films (the same SM film) shows that high burst effect for SM'_1 to SM'_3 formulations

were high (109.87%, 116.63%, and 108.54%, respectively) and accordingly $Rel_{0.5}$ were high (106.67%, 110.82%, and 110.8%, respectively). The release of Met drug from SM films was faster than the release of Sum drug from SM films ($p<0.05$). During the dissolution, the HPMC-containing films swelled, forming a gel layer on the exposed mucous surfaces.

Sumatriptan succinate with a hydrophilic nature and Log *P* value of 0.93 exhibits low permeability through the buccal mucosa. The microscopic observations indicated that none of the films had significant effect on the microscopic structure of mucosa. As shown in Figure 6, no cell necrosis was observed.

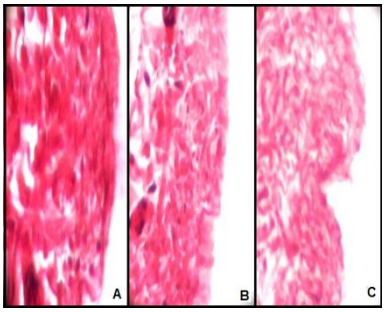

Figure 6. Histopathological evaluation of sections of buccal mucosal (A) un-treated (B) treated with film containing sumatriptan alone (C) treated with film containing sumatriptan combined with methoclopramide (magnitude X).

Conclusion

Fast dissolving thin Sum alone and Sum combined with Met films were successfully formulated using HPMC E15 formulations. The film had acceptable physical properties and drug content. This study clearly demonstrated that Sum alone and Sum combined with Met drugs can be successfully delivered through the sublingual route.

Acknowledgments

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Conflict of interests

The authors claim that there is no conflict of interest.

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