Research Article

Design and Evaluation of Buccal Patch Containing Combination of Hydrochlorothiazide and Lisinopril

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ABSTRACT

Purpose: The objective of the present study was to formulate and evaluate buccal patches containing combination of lisinopril (LP) and hydrochlorothiazide (HCZ). **Approach:** Films were fabricated by solvent casting method, using combination of mucoadhesive polymers such as hydroxypropylmethyl cellulose (HPMC), hydroxypropyl cellulose (HPC), polyvinyl alcohol (PVA) and polyvinyl pyrolidone (PVP) and ethyl cellulose (EC) as backing layer. The patches were evaluated for physicochemical characteristics such as weight, thickness, surface pH, folding endurance, bioadhesive strength, swelling index, drug content, tensile strength, elongation at break, mucoadhesion time, *in vitro* and *ex vivo* drug permeation. **Results:** The IR spectra showed no interaction between drug and polymer. Physicochemical characteristics of all the samples were found to be satisfactory. Swelling of the films increased with increasing content of HPMC or HPC and PVP. Bioadhesive force, tensile strength, percentage elongation and mucoadhesion time increased with higher proportions of HPMC, HPC and PVA. *In vitro* drug release studies demonstrated slower release of both drugs in formulations with higher amount of HPMC, HPC and PVA. The *in vitro* drug release data of most formulations best fitted first order model, except for the formulations FA3 and FC. *Ex vivo* drug permeation studies of formulations through porcine buccal mucosa showed similar results as *in vitro*. **Conclusion:** Buccal delivery of this combination can resolve the drawbacks like incomplete absorption in the gut thereby possible improvement in bioavailability, apart from controlled release of the drugs.

Key words: Buccal mucosa, Mucoadhesive, Hydrochlorothiazide (HCZ), Solvent casting method, Hydroxy propyl cellulose (HPMC), Lisinopril (LP).

INTRODUCTION

Oral drug delivery has been the most widely utilized route of administration among all the routes that have been explored for the systemic delivery of drugs for various pharmaceutical products of different dosage forms.[1] However, oral administration of drugs has disadvantages such as hepatic first pass metabolism and enzymatic degradation within the gastro intestinal tract (GIT), that prohibit administration of certain classes of drugs especially peptides and proteins. Transmucosal routes of drug delivery (i.e., the mucosal linings of the nasal, rectal, vaginal, ocular, and oral cavity) offer distinct advantages over oral administration for systemic drug delivery. These advantages include bypassing first pass effect, avoidance of presystemic elimination within the GIT, and depending on the particular drug, a better enzymatic flora for drug absorption. The most promising and challenging routes appear to be the nasal, sublingual and buccal.[2,3]

Buccal drug delivery has been proposed as an alternative to inefficient oral administration and inconvenient parenteral administration of drugs.[4] Buccal delivery for the transmucosal absorption of drugs into the systemic circulation provides a number of advantages such as ease of administration, sustained delivery and rapid onset of action, high blood levels, and avoidance of first-pass metabolism. The mucosa is relatively permeable, has a rich blood supply, is robust, and shows short recovery times after stress or damage. The attractive features of the

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oral mucosa include excellent accessibility and high patient acceptance and compliance**.** Moreover, since the oral mucosa is routinely exposed to a multitude of different foreign compounds; it is rather robust and less prone to irreversible irritation or damage by dosage form, or additives such as absorption promoters.[5,6] These factors make the oral mucosal cavity a very attractive and feasible site for systemic drug delivery.

Hydrochlorothiazide (HCZ) and Lisinopril (LP) were chosen as model drugs for the formulation into mucoadhesive buccal patches for controlled release. The rational for the combination of these two drugs for the said formulation is that they are available in the market as tablets and extensively prescribed by the physicians for the treatment of hypertension. However, considering the drawbacks associated with the individual drugs, it was thought to be worthwhile to formulate into buccal delivery formulation. HCZ has variable half-life of 5.6-14.8 h with the bioavailability of 65-72% as its absorption from GIT is found to be approximately 70% of the administered dose and is mainly eliminated by kidney.[7] On the other hand, LP is having onset of action 1-2 h and duration of action 24 h (once daily dosing). The peak plasma concentration is achieved after 7 h and the mean bioavailability is around 25% due to inter subject variability (6-60%), which is attributed to its slow and incomplete absorption from GIT.[8] Hence the present investigation is an attempt to improve the systemic bioavailability, reduce gastric intolerance and optimize therapeutic efficacy of the selected drugs by designing mucoadhesive buccal patches for controlled release.

MATERIALS AND METHODS

Materials

Hydrochlorothiazide and lisinopril were provided by Yarrow Chemicals Pvt. Ltd., Mumbai Hydroxypropyl cellulose, Hydroxypropylmethyl cellulose K4M, Polyvinyl pyrrolidone K-30 were provided by CDH Laboratory Reagent, New Delhi. Polyvinyl alcohol and propylene glycol were provided by Loba Chemie Pvt. Ltd., Mumbai. All other chemicals/reagents used were of analytical grade.

Methods

Preparation of buccal patches using PVP K-30 and HPMC/ HPC: Buccal patches were prepared by solvent casting method (Table 1). The weighed amount of drugs were dissolved in 2 ml of Dimethyl sulfoxide, followed by addition of PVP K-30. Stirred till the contents were dissolved and 10 ml of ethanol was added into the solution. The second polymer, HPMC/HPC was dissolved in it. Propylene glycol was added as plasticizer and the volume was made up to 20 ml using ethanol. The solution was then poured into glass moulds of diameter 9 cm containing backing layer and kept aside covered with funnel for controlled evaporation of solvent. The dried patches were cut into circular patches of 1.5 cm diameter, so that each patch contains about 7.5 mg of LP and 20 mg of HCZ.[7]

Preparation of buccal patches using PVA and PVP K-30

Buccal patches were prepared by solvent casting method (Table 1). Aqueous solution of polymers (PVP: PVA) in 20 ml hot water (80-100°) was prepared. Propylene glycol (5% w/v) was added as plasticizer. The drugs were dissolved in 2 ml of DMSO, incorporated into cooled gel solution of polymers and then made up to 30 ml volume. The gel was allowed to stand overnight at room temperature to remove all entrapped air bubbles. The solution was cast into the glass mould of 9 cm diameter containing backing layer. The dried patches were cut into circular patches of 1.5 cm diameter, so that each patch contains about 7.5 mg of LP and 20 mg of HCZ.[8]

Preparation of ethyl cellulose backing layer: Ethanol (5 ml) was added in a beaker containing 10 ml of acetone as solvent. Ethyl cellulose (1 g) was dissolved in the solvent with 0.35 ml of triethyl citrate as plasticizer. The polymer solution was kept for deaeration and then poured into 9 cm diameter petri dish with an aluminum foil spread over the surface. The solution was kept for controlled evaporation of the solvents at room temperature.

Preformulation study

Compatibility studies

FT-IR spectra matching approach was used for detection of any possible chemical interaction between the drug/s and polymers. The individual sample of drug/s and the three different drugs: polymers combination patches (powdered form) were mixed with suitable quantity of potassium bromide. About 50 mg of this mixture was compressed to form a transparent pellet using a hydraulic press at 15 tons pressure. It was scanned from 4000-600 cm^{-1} in a Bruker FTIR spectrophotometer. The IR spectrums of the formulations were compared with those of pure drugs and matching was done to detect appearance or disappearance of any peak.

Evaluation of Buccal Patches

Physical Characterization[8,9,10]

Uniformity of weight

The individual weight of 10 samples of each formulation was determined and the average weight was calculated.

Patch thickness

The thickness of 10 patches of each formulation was determined using micrometer screw gauge and average was determined.

Folding endurance

This test indicates the ability of the films to sustain mechanical handling as well as pliability during use in the oral cavity. The folding endurance was determined by repeatedly folding one patch at the same place till it broke or folded up to 300 times which is considered to be satisfactory for good film properties. The number of times the patch could be folded at the same place without breaking gives the value of the folding endurance.

Surface pH of the buccal patches

The surface pH of the patches was determined in order to investigate the possibility of any side effects due to change in pH *in vivo*, since an acidic or alkaline pH may cause irritation to the buccal mucosa. The patch to be tested was placed in petri dish and was moistened with 0.5 ml of distilled water and kept for 30 s. The pH was noted after bringing the electrode of pH meter in contact with the surface of the formulation and allowing equilibrating for 1 min. The average of 10 determinations for each of the formulation was taken.

Measurement of bioadhesive strength

Satisfactory bioadhesion is essential for successful application of bioadhesive drug delivery systems in order to increase the residence time at the site of application and hence prolonged absorption of the drug. The tensile strength required to detach the bioadhesive patch from the mucosal surface, is a measure of the bioadhesive performance. Several techniques have been reported in the literature for the measurement of bioadhesive strength. In the present work, a specially designed and fabricated assembly based on published literature was used. Porcine cheek pouch was used as a model surface for bioadhesion testing. After the cheek pouch was excised and trimmed evenly, it was washed in simulated salivary fluid, and then used immediately for the test.

Fabrication of the test assembly

The working of a double beam physical balance formed the basis of the bioadhesion test assembly. The right pan was removed and a stainless steel chain was hung. A Teflon block with 1.5 inches height and 1.5 inches diameter was hung with the stainless steel chain to balance the weight of the other pan. The height of total set up was adjusted to accommodate a glass container or beaker below it, leaving a headspace of about 0.5 cm in between. Another Teflon block of 2 inches height and 1.5 inches diameter was kept inside the glass container, which was then placed below the top hung Teflon block. Suitable weights were added (15.0 g) on the left pan to balance the beam of the balance.

Method

The porcine cheek membrane was attached with the mucosal side upward over the lower Teflon block which was then placed into the glass container, which was then filled with simulated salivary fluid, such that the salivary fluid just touches the surface of the mucosal membrane to keep it moist. This was then kept below the upper Teflon block. The patch under test was fixed to the surface of the upper block with glue. The 15.0 g weight on the right pan was removed and this lowered the upper Teflon block along with the patch, so that it is in contact with mucosal surface. A load of 20.0 g was placed as initial pressure on the upper block for 3 min and then slowly weights were added on the left pan starting from 100 mg till the patch separated from the mucosal surface. The excess weight on the pan (i.e., the total weights minus 15.0 g) required to separate the patch from the mucosa was noted

$$F = \frac{W_w \times g}{A}$$

Where F is the bioadhesion force (kg/m/s^2) Ww is the mass applied, g is the acceleration due to gravity (cm/s^2) and A is the surface area of the patch (cm^2).

Swelling studies

The patch sample of 1.5 cm diameter was weighed and placed in a pre-weighed stainless steel wire sieve of approximately 800 μm mesh. The mesh containing the sample was then submerged into 15 ml of simulated salivary fluid of pH 6.8 contained in a porcelain dish. At definite time intervals, the stainless steel mesh was removed; excess moisture was removed by carefully wiping with absorbent tissue and reweighed. Increase in weight of the film was determined at each time interval until a constant weight was observed. The degree of swelling was calculated using the formula,

$$Swelling\ index\ (S.I.) = \frac{W_t - W_0}{W_0}$$

Where, W_t is weight of the patch at time t and W_0 is weight of the patch at time zero.

Uniformity of drug content

Buccal patches of PVP K-30 and HPMC / HPC

Uniformity of drug content was determined by dissolving one patch of 1.5 cm diameter designed to contain 20 mg of HCZ and 7.5 mg of LP by homogenization in a mixture of 5 ml of ethyl alcohol and 2 ml of DMSO for 5 h with occasional shaking and diluted to

50 ml with distilled water. After filtration to remove insoluble residue, 1 ml of the filtrate was diluted to 10 ml with simulated saliva of pH 6.8. The absorbance was measured at 269.8 nm and 217 nm using UV spectrophotometer. The experiments were carried out in triplicate for all formulations.

Buccal patches of PVA and PVP K-30

One patch of 1.5 cm diameter was dissolved in 50 ml of distilled water by homogenization for 6 h with occasional shaking. After filtration to remove insoluble residue, 1 ml of the filtrate was diluted to 10 ml with simulated saliva of pH 6.8. The absorbance was measured at 269.8 nm and 217 nm using UV spectrophotometer. The experiments were carried out in triplicate for all formulations.

Measurement of tensile strength

This mechanical property was evaluated using Instron universal testing instrument (Model 1121, Instron Ltd., Japan, NITK, Surathkal, India) with a 5 kg load cell. Film strips in special dimension and free from air bubbles or physical imperfections were held between two clamps positioned at a distance of 3 cm. During measurement, the strips were pulled by the top clamp at a rate of 100 mm/min; the force and elongation were measured when the film broke. Results from film samples, which broke at and not between clamps, were not included in the calculations. Measurements were run in triplicate for each film. Two mechanical properties, namely, tensile strength and percentage elongation were computed for the evaluation of the film. Tensile strength is the maximum stress applied to a point at which the film specimen breaks and can be computed from the applied load at rupture as a mean of three measurements and cross sectional area of fractured film as described from the following equation.[11]

Tensile strength=force at break/initial cross section area of the sample (mm^2)

Percentage elongation at break=increase in length×100/Original length

Ex vivo mucoadhesion time

Ex vivo mucoadhesion was performed by application of the patch on freshly cut porcine buccal mucosa. The porcine tissues were fixed on the internal side of a beaker with cyanoacrylate glue. The patch was wetted with 50 µl of simulated salivary fluid and was attached to the porcine buccal tissue by applying light force with fingertip for 20 s. The beaker was filled with 200 ml of simulated salivary fluid and kept at 37°. After 2 min, stirring at 50 rpm was maintained to simulate the buccal cavity environment. The time taken for the patch to completely erode or detach from the mucosa was observed as the *ex vivo* mucoadhesion time.

Drug release studies

In vitro release studies

In vitro release studies were carried out by slight modification of the method suggested by Perioli L *et al* and Ilango *et al*.[12,13] A buccal patch was attached to the wall of the dissolution vessel such as a 250 ml beaker midway from the bottom with instant adhesive. After 2 min the vessel was filled with 200 ml of simulated saliva of pH 6.8 and placed on a magnetic stirrer. The temperature of the dissolution medium was maintained at 37° and stirred at 50 rpm. Samples of 3 ml were withdrawn at predetermined time intervals and replaced with fresh medium. The samples were diluted appropriately with simulated saliva and assayed spectrophotometrically at 269.8 nm and 217 nm by simultaneous estimation method. Three patches of each formulation were subjected to drug release studies in the same manner and the average cumulative percentage drug was determined.[14]

Kinetic analysis of in vitro release data [15,16,17]

In order to determine the release mechanism that provides the best description to the pattern of drug release, the *in vitro* release data were fitted to zero order, first order, and Higuchi model. The release data were also kinetically analysed using the Korsmeyer-Peppas model. The release exponent (n) describing the mechanism of drug release from the matrices was calculated by regression analysis using the following equation:

$$M_t/M_\infty = Kt^n$$

Where, M_t/M_∞ is the fraction of drug released (using values of M_t/M_∞ within the range 0.10-0.60) at time t and K is a constant incorporating the structural and geometric characteristics of the release device. A value of n=0.5 indicates case I (Fickian) diffusion, 0.5<n<1 indicates anomalous (non-Fickian) diffusion, and n= indicates case II transport (Zero order release), n>1 indicates Super case II transport.

Ex vivo permeation studies

Permeation of HCZ and LP from aqueous solution

Before the film formulations are actually subjected to *ex vivo* buccal permeation studies, it was considered necessary to determine whether HCZ and LP were able to penetrate the buccal mucosa and what would be the extent of permeation. For this study, the drug in the most available form, i.e., an aqueous solution (20 mg of HCZ and 7.5 mg of LP in 5 ml of simulated saliva) was placed in the donor compartment.

Permeation of HCZ and LP from buccal patches

Ex vivo permeation study was carried out by using modified Franz diffusion cell of internal diameter of 2.5 cm. It consists of a glass tube open at both end. Porcine buccal mucosa was chosen as the model membrane. The buccal pouch of freshly sacrificed pig was procured from the local slaughter house. The buccal mucosa was excised and trimmed evenly from the sides and then washed in isotonic phosphate buffer of pH 6.8 and used immediately. The membrane was stabilized before mounting in order to remove the soluble components. The mucosa was mounted between the donor and receptor compartments. The receptor compartment was filled with 200 ml of isotonic phosphate buffer of pH 7.4 which was maintained at 37° and the hydrodynamics was maintained by stirring with a magnetic bead at 50 rpm. The patch (1.5 cm diameter) was placed in intimate contact with the mucosal surface of the membrane that was previously moistened with a few drops of simulated saliva. At predetermined time intervals, 1 ml sample was withdrawn and analyzed using an UV spectrophotometer at 269.8 nm and 217 nm.[18,19]

Stability study

The stability studies of HCZ and LP buccal patches were conducted to evaluate physical appearance, surface pH, swelling index and *in vitro* drug release at the end of eight weeks when stored under conditions at 25°C ± 2°C/60% ± 5% RH and 40°C ± 2°C/75% ± 5% RH.[24]

RESULTS

Compatibility studies

IR studies were carried out for pure drugs and excipients like HPMC, HPC, PVA and PVP K-30, which were used in formulations to determine the interaction between drugs and the polymers. The IR spectra are given in the (Figure 1A-F). The spectral values for the drugs were compared with reference standard sample spectra.[20] The IR spectrum of the HCZ (Figure 1A) showed the characteristic peaks at 3362.04 cm^{-1} (NH stretching group), 750.33 cm^{-1} (-NH bending group), 1604.83 cm^{-1} (C-C stretching group), 1244.13 cm^{-1} (SO_2 stretching) and 2960 cm^{-1} (-CH_2 group). The IR spectrum of LP (Figure 1B) exhibited the principal peaks at 3557.85 cm^{-1} due to N-H stretching, O-H stretching around 3300 cm^{-1}, aromatic C-H stretching around 2900 cm^{-1}, C=O stretching around 1700 cm^{-1} and C-O stretching around 1045 cm^{-1}. The spectra of formulations (Figure 1C-F), showed presence of peaks in the region of characteristic peaks of drugs confirmed the absence of interaction between the drugs and excipients used in the formulation.

Physical characterization

The results of physical characterization of all the formulations are given in Table 2. All the formulated films were found to be smooth in texture. The PVA films were found to be transparent and all others were translucent. The percentage drug content of all formulation was found to be in the range of 97.891-99.035 %. The results of all formulations are given in Table 2. The thickness of the formulated patches was found to be in the range of 0.775 ± 0.0058-0.965 ± 0.0057 (Table 2). It was found that all the formulations showed good folding endurance of greater than 300. Surface pH of the buccal patches was found to be in the range of 6.15-6.66. The bioadhesive force for the different formulations was found to be 73.59-128.95 $kg/m/s.^2$ Swelling index of the formulations was found to be in the range of 1.5-2.8, with least for FA1 and maximum for FC3. The results of tensile strength and percentage elongation of all the formulations are given in the Table 3. The residence time for the formulation i.e., the time taken for the patch to detach or erode completely from the mucosa was found to be between 3.1-9.0 h.

In vitro drug release studies

The percentage amount of drug released is plotted against time to obtain the release profiles as shown in Figure 2(A-C). The release pattern of the drugs from formulations followed FA1>FA2>FA3, FB1>FB2>FB3 and FC3<FC2<FC1.

Ex vivo permeation studies

The percentage amount of drug permeated is plotted against time to obtain the release profiles as shown in Figure 3(A-C). The results of *ex vivo* drug permeation studies showed permeation of drug slower than that of *in vitro* drug release studies by an hour.

Stability study

The results of stability studies of buccal patches showed no significant change with respect to physical appearance, surface pH, swelling index and *in vitro* drug release at the end of eight weeks (Table 4). PVA patches of HCZ and LP showed a mild shrinkage at the end of eight weeks. This was probably due to loss of moisture and plasticizer from the patches when stored at this temperature. Aging did not alter the drug release profiles of any of the films significantly at the end of the storage period.[21,24]

Table 1: Formulations of films loaded with Hydrochlorothiazide and lisinopril

Ingredients	Formulation Code								
	FA1	FA2	FA3	FB1	FB2	FB3	FC1	FC2	FC3
HCZ (g)	0.72	0.72	0.72	0.72	0.72	0.72	0.72	0.72	0.72
LP (g)	0.45	0.45	0.45	0.45	0.45	0.45	0.45	0.45	0.45
HPMC K-15 (g)	0.468	0.561	0.623	-	-	-	-	-	-
HPC (g)		-	-	0.468	0.561	0.623	-	-	-
PVP (g)	0.468	0.374	0.312	0.468	0.374	0.312	1.116	0.992	0.868
PVA (g)	-	-	-	-	-	-	0.124	0.248	0.372
DMSO (ml)	2	2	-	2	2	2	2	2	2
Propylene glycol (ml)	1.25	1.25	1.25	1.25	1.25	1.25	1.25	1.25	1.25
Ethanol q.s (ml)	30	30	30	30	30	30		-	-
Distilled water q.s (ml)	-	-		-	30	30	30	-	-

HCZ-Hydrochlorothiazide, LP-Lisinopril, Hydroxypropylmethyl cellulose (HPMC), Hydroxy propyl cellulose (HPC), polyvinyl alcohol (PVA) and polyvinyl pyrolidine (PVP), Di methyl sulfoxide (DMSO).

Table 2: Results of physical characterization

Formulation Code	Weight (mg)	Thickness (mg)	Surface pH	Folding endurance	Bioadhesive force ($Kg/m/s^2$)
FA1	35.0 ± 0.8165	0.775 ± 0.0058	6.33 ± 0.009	>300	86.38 ± 1.2109
FA2	35.25 ± 0.9574	0.7575 ± 0.0126	6.29 ± 0.008	>300	68.90 ± 0.8937
FA3	36.5 ± 0.5774	0.7875 ± 0.0096	6.46 ± 0.019	>300	73.59 ± 0.6223
FB1	32.5 ± 1.291	0.6975 ± 0.0126	6.15 ± 0.017	>300	74.25 ± 1.0879
FB2	33 ± 0.8165	0.6875 ± 0.0096	6.29 ± 0.01	>300	79.13 ± 0.6223
FB3	34.75 ± 0.9574	0.71 ± 0.0082	6.25 ± 0.015	>300	86.38 ± 1.2109
FC1	38.5 ± 1.291	0.98 ± 0.0081	6.66 ± 0.022	>300	128.95 ± 1.232
FC2	40 ± 2.1602	0.9675 ± 0.0095	6.59 ± 0.013	>300	123.55 ± 1.0622
FC3	38.5 ± 1.7078	0.965 ± 0.0057	6.57 ± 0.022	>300	118.29 ± 0.9681

Data are represented as mean ± SD (*n=3*)

Table 3: Results of tensile strength and percentage elongation of all formulations

Formulation code	Tensile Strength	% Elongation
FA1	0.095 ± 0.0103	11.34 ± 0.0023
FA2	0.117 ± 0.0150	16.16 ± 0.0015
FA3	0.132 ± 0.0026	21.34 ± 0.0026
FB1	0.065 ± 0.0093	97.38 ± 0.0128
FB2	0.090 ± 0.0015	115.96 ± 0.0349
FB3	0.127 ± 0.0023	121.24 ± 0.1281
FC1	0.825 ± 0.0062	153.34 ± 0.0681
FC2	0.732 ± 0.0053	124.96 ± 0.0046
FC3	0.630 ± 0.0201	101.76 ± 0.0352

Data are mean ± SD

Table 4: Results of stability study

Formulation Code	Parameters	Before storage	At the end of 8 weeks	
			25°/60% RH	40°/75% RH
FA1	Physical appearance	Translucent & smooth	-	-
	Surface pH	6.33	6.31	6.33
	Maximum Swelling index	1.023	1.022	1.021
	Maximum % drug release			
	HCZ	96.05	96.07	96.09
	LP	97.43	97.41	97.44
FA2	Physical appearance	Translucent & smooth	-	-
	Surface pH	6.29	6.26	6.28
	Maximum Swelling index	1.35	1.33	1.34
	Maximum % drug release			
	HCZ	98.04	98.02	98.03
	LP	97.45	97.42	97.41
FA3	Physical appearance	Translucent & smooth	-	-
	Surface pH	6.44	6.46	6.45
	Maximum Swelling index	1.023	1.022	1.021
	Maximum % drug release			
	HCZ	90.48	90.45	90.40
	LP	99.48	99.45	99.40
FB1	Physical appearance	Translucent & smooth	-	-
	Surface pH	6.15	6.11	6.13
	Maximum Swelling index	1.023	1.022	1.021
	Maximum % drug release			
	HCZ	96.05	96.07	96.09
	LP	97.43	97.41	97.44
FB2	Physical appearance	Translucent & smooth	-	-
	Surface pH	6.33	6.31	6.33
	Maximum Swelling index	1.250	1.23	1.24
	Maximum % drug release			
	HCZ	97.76	97.02	97.20
	LP	99.13	99.09	99.05
FB3	Physical appearance	Translucent & smooth	-	-
	Surface pH	6.25	6.23	6.24
	Maximum Swelling index	2.17	2.15	2.13
	Maximum % drug release			
	HCZ	97.28	97.25	97.20
	LP	99.43	99.40	99.35
FC1	Physical appearance	Translucent & smooth	-	-
	Surface pH	6.66	6.55	6.64
	Maximum Swelling index	2.12	2.11	2.10
	Maximum % drug release			
	HCZ	93.52	93.50	93.45
	LP	98.50	98.45	98.40
FC2	Physical appearance	Translucent & smooth	-	-
	Surface pH	6.59	6.52	6.55
	Maximum Swelling index	2.50	2.47	2.45
	Maximum % drug release			
	HCZ	94.55	94.50	95.44
	LP	99.8	99.80	99.72
FC3	Physical appearance	Translucent & smooth	-	-
	Surface pH	6.57	6.53	6.55
	Maximum Swelling index	2.76	2.75	2.72
	Maximum % drug release			
	HCZ	96.49	96.40	96.34
	LP	97.16	97.12	97.07

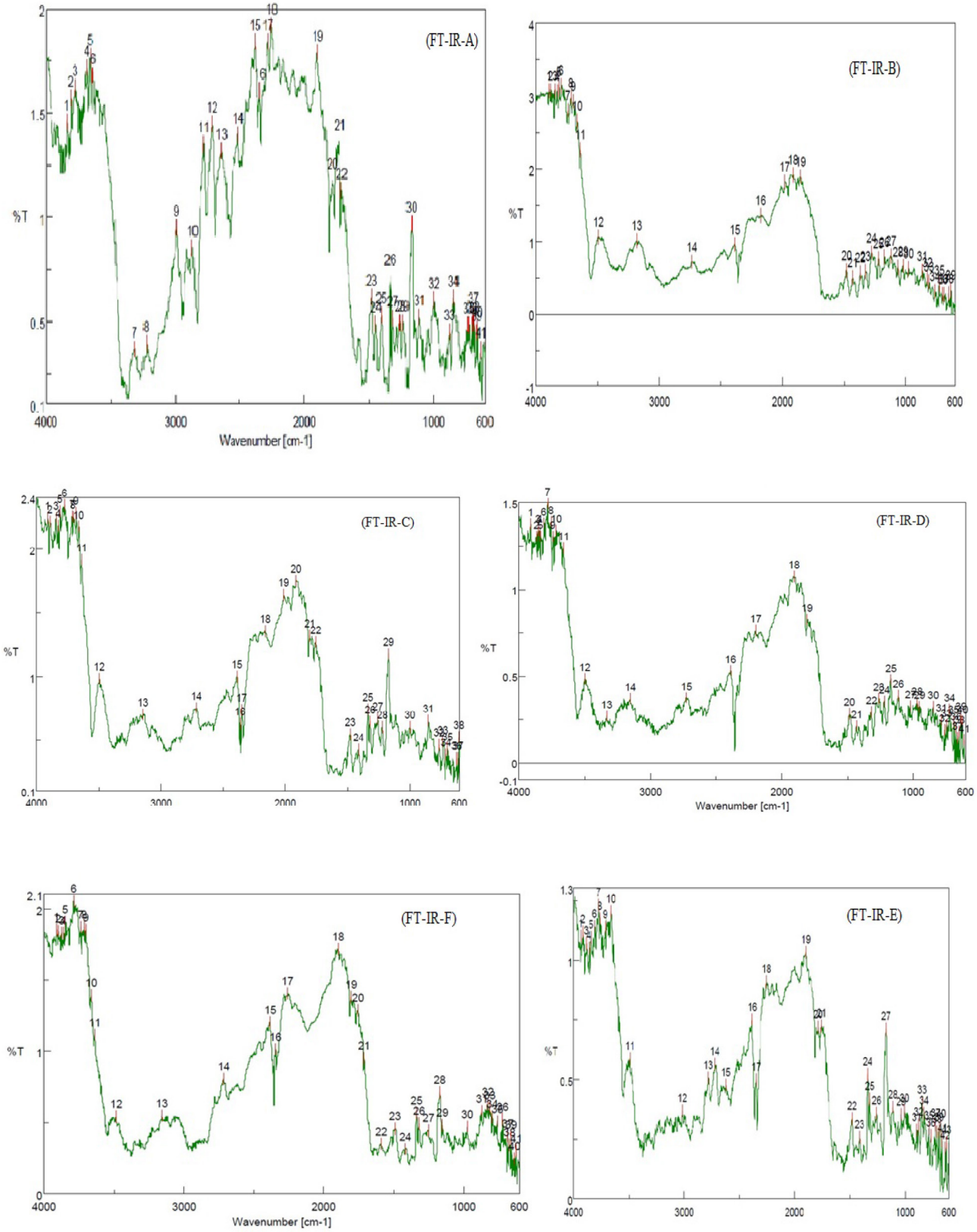

Figure 1: FTIR Spectra. (A) HCZ (B) LP (C) HCZ + LP (D) FA1 (E) FB1 (F) FC1

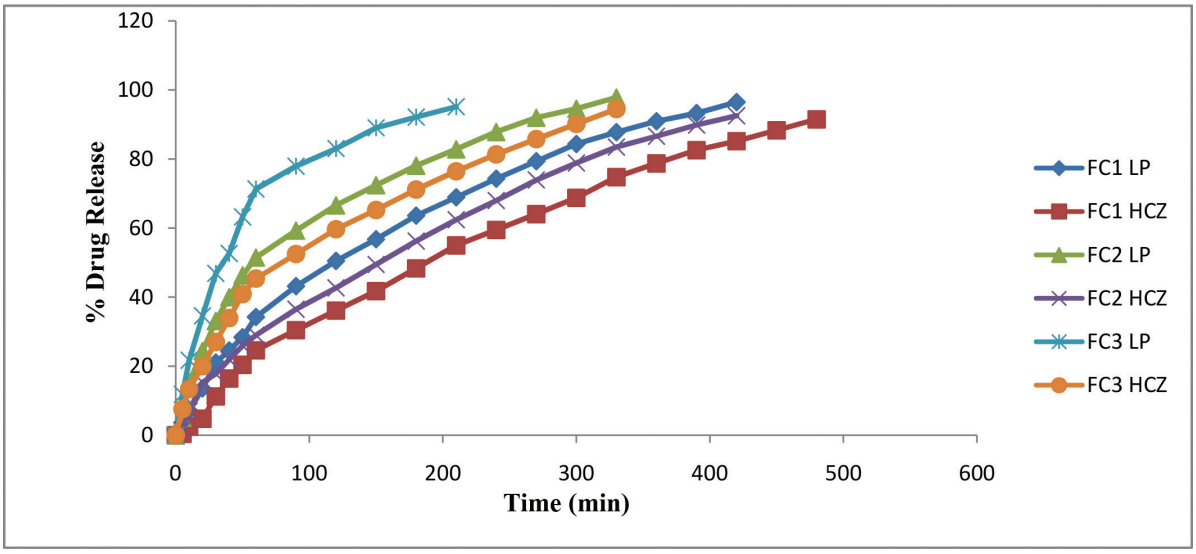

Figure 2 (A): *In vitro* release profile of HCZ and LP from HPMC & PVP formulations

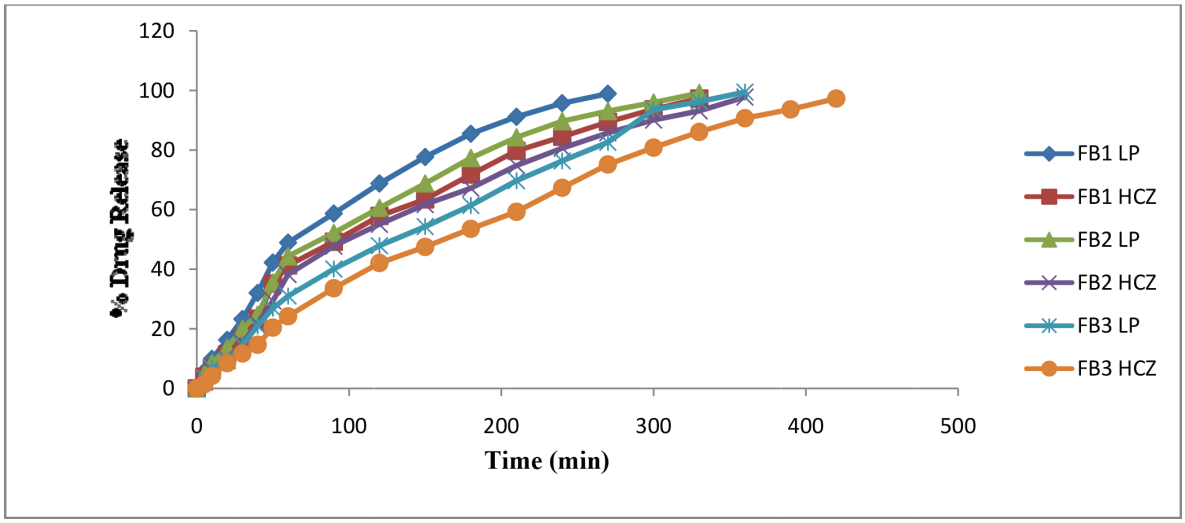

Figure 2(B): *In vitro* release profile of HCZ and LP from HPC & PVP formulations

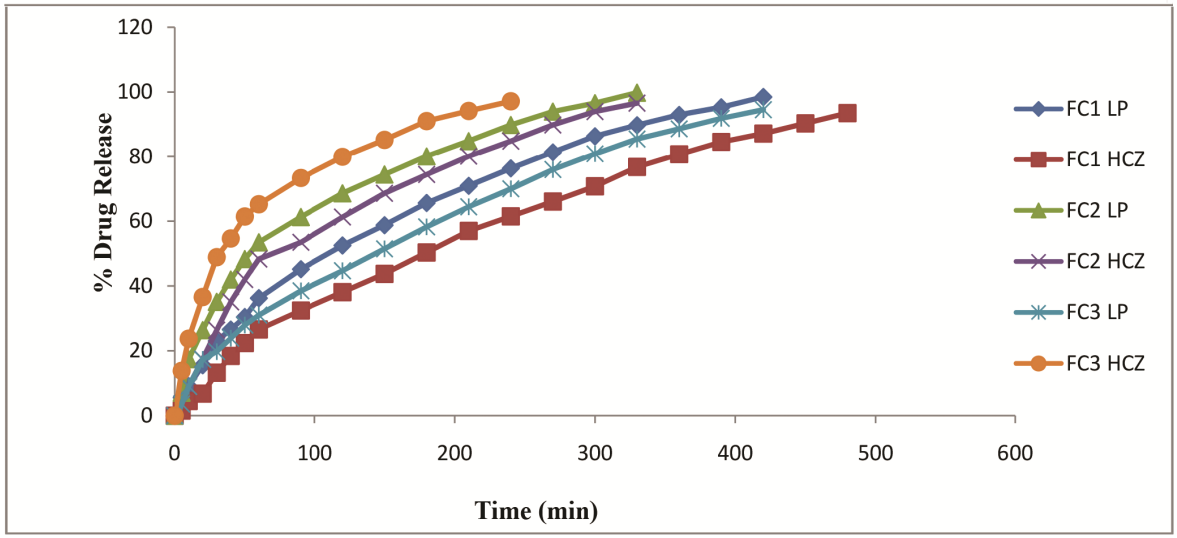

Figure 2(C): *In vitro* release profile of HCZ and LP from PVA & PVP formulations

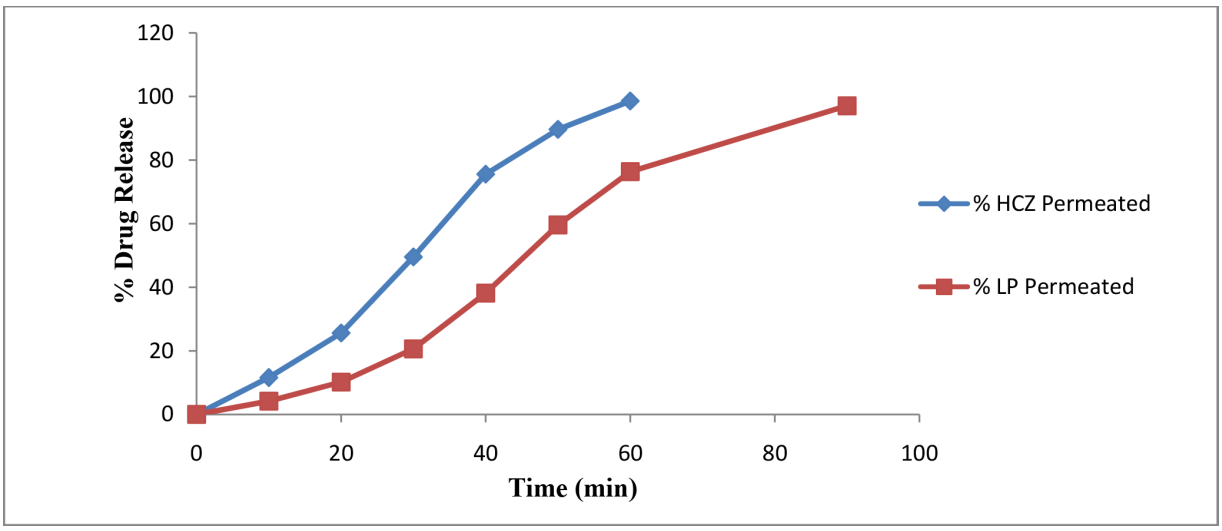

Figure 3(A): *Ex vivo* release profile of aqueous solution of HCZ and LP

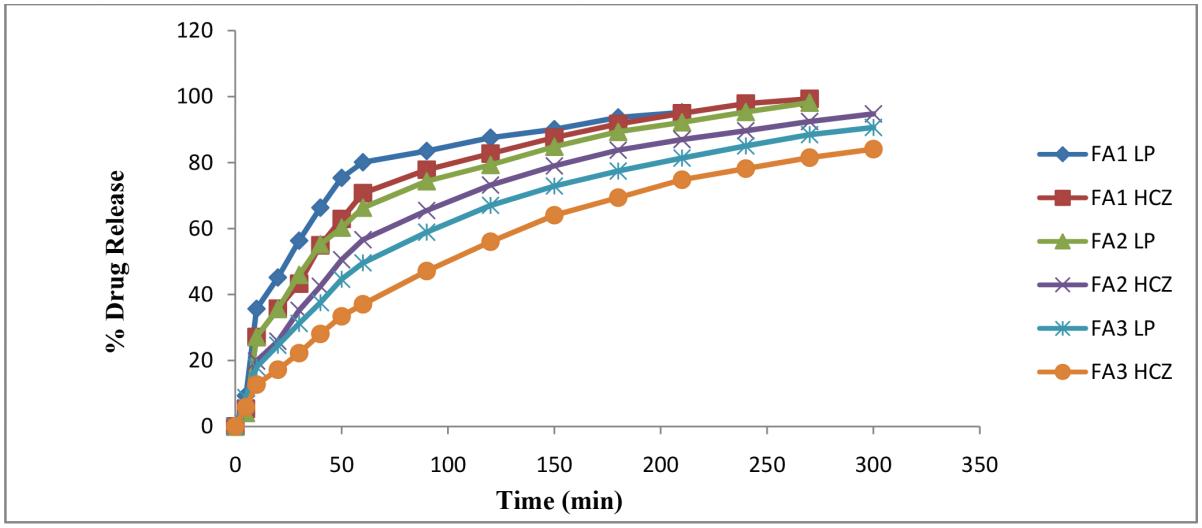

Figure 3(B): *Ex vivo* release profile of HCZ and LP from HPMC & PVP formulations

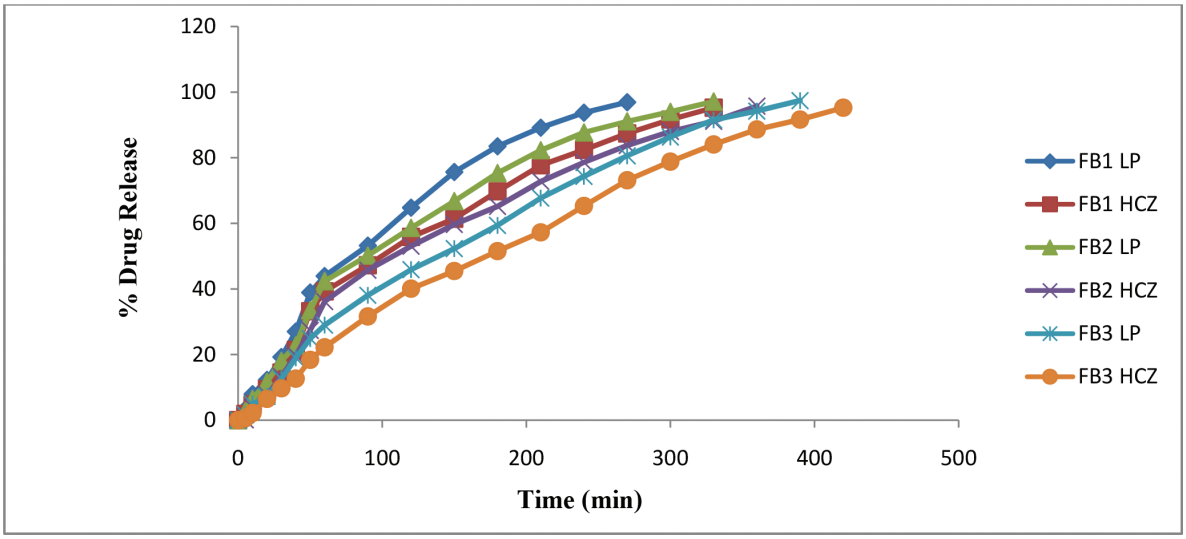

Figure 3(C): *Ex vivo* release profile of HCZ and LP from HPC & PVP formulations

DISCUSSION

Films with good physical properties were obtained by the solvent casting method. The concentration of the polymers plays a significant role in the preparation of the film. As all the films were formulated from swellable hydrophilic polymers, concentrations exceeding those indicated in the formula could not be used, because the resulting solutions were found to be too viscous to handle and pour. Casting of film was done after complete deaeration as it causes imperfection in the films. Films with higher percentage of PVP could not be prepared as it releases the drug at faster rate probably due to the solubilization effect. All the formulated films were found to be smooth in texture. The PVA films were found to be transparent and all others were translucent.

Compatibility studies

From FT-IR spectra it is observed that there is no significant change in the original peak of the drug and the polymers when compared with spectra of formulated patches and this indicated that there was no interaction between drug and polymer.

Physical characterization

It was observed that weight of the entire film sample in each formulation was uniform. Between formulations, the weight increased with increased content of polymers used. The thicknesses of the all film samples were found to be uniform in each formulation. The films with increased polymer content showed a marginal increase in thickness. It was found that all the formulations showed good folding endurance greater than 300. The surface pH of each film was found to be in the range of 6.15 to 6.66 and hence it can be concluded as less potential to irritate the buccal mucosa, thereby they can be comfortable to the patient when used in the buccal cavity. It was found that among the formulations FA & FB, those with more amount of HPMC or HPC (FA3 & FB3) showed more bioadhesive force compared to other formulations. Maximum bioadhesion was observed for PVA patches (maximum with FC, 128.95 kg/m/s^2), probably due to non-ionic and strong adhesive properties of PVA, because of hydrogen bonding or significant chain penetration or both.

Swelling studies

Swelling index of the formulations was found to be in the range of 1.5-2.8, with least for FA1 and maximum for FC3. The purpose of measuring swelling index is to determine the ability of hydrophilic polymers used in formulation to take up water upon hydration. The hydration and swelling behavior of the polymer was reported to be crucial for its character because the former is necessary to initiate intimate contact of the film with the mucosal surface. Adhesion increases with the degree of hydration until a point where over hydration leads to an abrupt drop in adhesive strength due to disentanglement at the polymer tissue interface. The rate and the extent of film hydration and swelling also affect the film adhesion and consequently the drug release from the film. The rate of swelling affects the duration of adhesion with faster swelling resulting in adhesion of shorter duration. Studies have shown that excessive hydration can lead to weakening of the adhesive bond due to dilution of functional groups responsible for the adhesive interaction between the bioadhesive film and mucosa.[22] It was found that among the formulations FA and FB, with more amount of HPMC or HPC, i.e., FA3 and FB3 showed maximum swelling index of 2.08 and 2.17 at the end of 25 and 30 min respectively. The films of PVA and PVP combination showed the slowest uptake of water among all other formulations. Among all PVA-PVP films, FC3 showed maximum swelling index at the end of 240 min, probably due to increased content of PVP.

Tensile strength measurement

The tensile strength gives an indication of the strength and elasticity of the film reflected by the parameters, Tensile Strength (TS) and Elongation at Break (E/B). A weak and soft polymer is characterized by a low TS and E/B; a hard and brittle polymer shows a moderate TS and low E/B; a soft and tough polymer shows a high TS and E/B. The results showed that for the formulations FA and FB, the TS and E/B increased with the increase in the percentage of mucoadhesive polymer, HPMC & HPC respectively. Proportions of PVP higher than that used in these films make them weaker. In the case of PVA films (FC3), TS and E/B is the greatest for FC1 and least for FC3, indicated that the inclusion of PVP decreased the TS. Formulation FC showed high TS compared to FA and FB, which increased with increased amount of PVA, and hence, PVA gives good TS to buccal patches compared to HPMC or HPC. Therefore, such patches are found to be tough and strong enough for use.

Residence time /ex vivo mucoadhesion time

The residence time for the formulation i.e., the time taken for the patch to detach or erode completely from the mucosa was observed was more with formulation containing PVA, the mucoadhesion time increased as the PVA content increased with a maximum of 8.9 h in case of FC1. It was because, as PVA content increased its mucoadhesive force also increased which results in longer mucoadhesion time. Mucoadhesive force also resulted in longer mucoadhesion time; among FA and FB formulations, FA3 and FB3 showed longer mucoadhesion time.

Drug release studies

In vitro drug release studies

Among the formulations FA (Figure 2A) and FB (Figure 2B), those formulations with more amounts of HPMC and HPC i.e. FA3 and FB3 showed slower release extended up to 5 h and 7 h respectively. This may be due to extensive swelling of HPMC and HPC, which created a high viscosity gel barrier for drug diffusion. Among all PVA films (Figure 2C) extent of drug release for both drugs was greater in FC3 films. It was observed that with the increased content of PVP, the rate and extent of drug release was faster. This was because of water soluble polymer PVP, resulting in increased wetting and penetration of water into the film matrices and hence increased diffusion of the drug. Further, PVP is also reported as solubilizing agent.[23] The order of release was FA1>FA2>FA3; FB1>FB2>FB3; FC3<FC2<FC1.

Kinetic analysis of in vitro release data

The release kinetic of the drugs from the formulations was found to be zero order to a greater extent. The release mechanism was best explained by non-Fickian transport as the 'n' values ranged from 0.53-0.78, indicating anomalous (non-Fickian) diffusion. Hence the release of the drugs followed diffusion as well as erosion mechanism from the formulations.[16,17]

Ex vivo permeation studies

Results of *ex vivo* drug permeation studies showed very rapid (within 100 min, Figure 3A) permeation of drugs. However, the permeation of drugs was found to be slower than that of *in vitro* drug release studies by an hour. The slowest permeation of drug was seen in FC1 (Figure 3D) which was extended up to 7 h with a release of 96.5 % for LP and up to 8 h with a release of 91.52 % for HCZ. Among the formulations FA and FB, permeation was found to be faster for FA1 (Figure 3B) and FB1 (Figure 3C) respectively with respect to both the drugs. HPMC (FA3) and HPC (FB3) films showed maximum permeation of both drugs within 6 h. The order of drug permeation in each set of formulation can be given as FA1>FA2>FA3; FB1>FB2>FB3; FC3<FC2<FC1.

CONCLUSION

Buccal delivery of drug combinations have the potential of delivering the drugs as it can resolve the drawbacks like incomplete absorption in the gut thereby possible improvement in bioavailability, apart from controlled release of the drugs which in turn improves patient compliance especially with those patients who have difficulty in swallowing the tablet formulations.

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CONFLICT OF INTEREST

The author declare no conflict of interest.

ABBREVIATION USED

HPMC: Hydroxypropyl methyl cellulose; HPC: Hydroxypropyl cellulose; PVA: Polyvinyl Alcohol; PVP: Polyvinyl Pyrolidine; EC: Ethyl Cellulose; HCZ: Hydrocholorothiazide; LP: Lisinopril; GIT: Gastrointestinal Tract; IR: Infra Red Spectrum; DMSO: Di methyl sulfoxide; SD: Standard Deviation.

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