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Research Article

DESIGN AND EVALUATION OF BUCCAL PATCHES OF LOSARTAN POTASSIUM USING NATURAL GUM

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ABSTRACT

In the present study, an attempt was made to develop a novel mucco-adhesive drug delivery system in the form of the buccal patches for the release of Losartan potassium in a unidirectional manner, to maintain constant therapeutic levels of the drug for long time using natural polymer. Buccal formulations containing Losartan potassium mucco adhesive patches were developed to a satisfactory level in term of drug release, bio adhesive strength, content uniformity, moisture content, surface pH, thickness and stability study. The patches were prepared by solvent casting method. Five formulations were developed and formulations with varying concentrations of natural polymers. FTIR studies have shown no indication of interactions between drug and polymer. USP type II apparatus was used to perform *in-vitro* release study under perfect sink condition. All the formulations followed zero order kinetics. Although all bucco adhesive formulations exhibited satisfactory drug release, best results have been obtained for formulation with higher polymer concentration in terms of evaluations performed. From the results obtained F5 was found as best formulation.

Keywords: *Manilkara Zapota* Seed Gum, Buccal patches, Mucco adhesion, Solvent casting method, Losartan potassium

INTRODUCTION

Mucco adhesive polymers, which are synthetic or natural in nature, are capable of adhering to the mucosa. Oral route of drug delivery system is most acceptable when compared with otherroutes[1]. Administration of the drug via the mucosal layer is a novel method that can render treatment more effective and safe, not only for the tropical diseases but also for systemic ones. The mucosal layer lines a number of regions of the body including the gastrointestinal tract, buccal cavity, airways, ear, nose, eye, urogenital tract, vagina and rectum are covered[2].

In this modern era, lots of mucco adhesive devices have been developed. Nevertheless, compared to tablets buccal patches provide greater flexibility and comfort. Moreover, it can reside on mucosa for more time compared to gels. In addition, it should possess sufficient mucco adhesive strength thereby it will be retained in the mouth for a long duration[3].

Numerous natural polymers such as pectin, guar gum, chitosan, etc. are used as mucco adhesive polymers which are economical, safe, stable and gel-forming in nature. The polymer which is extracted from fruits of *Manilkara zapota* belongs to the family Sapotaceae can be used as a sustained release and mucco adhesive polymer[4,5].

Losartan potassium is a drug used to treat high blood pressure belongs to the category of angiotensin II receptor antagonists. The drug entirely absorbed from GIT because of its high first-pass metabolism, its bioavailability is about 25-30%[6]. Upon buccal administration, higher bioavailability of Losartan potassium was observed and it indicates that its oral availability can be improved by formulating it in mucco adhesive dosage form. So buccal patches will maintain a therapeutic concentration for a long time and thereby enhance patient compliance and also decrease the frequency of administration. In the current study, buccal patches of Losartan potassium using zapota polymer has been developed and evaluated[7].

MATERIALS AND METHODS

Manilkara zapota seeds were naturally collected from Neyyar forest area of the district, Thiruvananthapuram. The plant and seeds of *Manilkara zapota* was authenticated by Dr. Sheeba M.S, Assistant Professor, Department of Botany, Govt. College for Women, Thiruvananthapuram. The herbarium voucher specimen number WC/118/2018 sample voucher specimen of plant was deposited for future reference. Losartan potassium was a gift sample (Sangrose Laboratories, Alapuzha, India). HPMC K-100M, sucralose, ethylcellulose, and isopropyl alcohol were obtained from Yarrow chem., Mumbai.

Isolation and Purification of Gum

The maceration technique was used to isolate polymer from seeds. Zapota seeds were collected and dried in sunlight; its kernels were crushed into a fine powder. Petroleum ether was added to 100 gm seed powder and wait for 5 hours. Then it was poured into 500 ml of cold water and the slurry was heated and kept aside for one day. After one day it was filtered through a muslin cloth and the filtrate was centrifuged at 3000 rpm for 10 minutes. Then the supernatant was added with an excess volume of acetone to precipitate the gum. The formed precipitate was washed with chloroform and is then dried in a hot air oven at 40-45^0 C. It is then collected, grounded, passed through a no. 120 sieve and stored in a desiccators until use[8].

Identification of Drug and Drug-Polymer Interaction Study

FTIR was carried on the drugs, the physical mixture of the drug and different polymers. Here spectral changes in the mixture are the basis for the determination of compatibility.

Preparation of Standard Curve of Losartan Potassium

Accurately weighed 100 mg of Losartan potassium and transferred into 100 ml volumetric flask and dissolved in small quantity of methanol and diluted with 6.8 phosphate buffer up to the mark to give stock solution 1 mg/ml. 1 ml was taken from the stock solution in another volumetric flask and diluted up to 100 ml to give a stock solution 10 µg/ml. Further dilutions were made from 2-10 µg/ml with 6.8 phosphate buffer and absorbance was measured at 235 nm[9].

Preparation of Mucco adhesive Buccal Patches

The buccal mucco adhesive patches of Losartan potassium were prepared by the solvent casting method. The calculated quantity of zapota gum was added to 10 ml of warm water and stirred to produce a solution. It is then kept aside for one day to form a clear solution. To the above polymer solution add 10 ml solution of HPMC K-100M in acetone with stirring. The drug solution was prepared by dissolving Losartan potassium (397.40 mg), citric acid (0.5 mg) and sucralose in 10 ml of water by stirring on a magnetic stirrer for one hour. Both drug solution and polymer solution were mixed and stirred for one hour. The prepared viscous solution was then poured onto a petridish of 9 cm diameter and placed it in an oven at 45^0 C for one day. The backing layer of ethyl cellulose was then prepared by slowly pouring a solution containing 500 mg of ethyl cellulose and 0.2 ml dibutyl phthalate in 10 ml of acetone to the above-formed drug-containing mucco adhesive layer. It was air-dried for one hour. Then the patch was removed from petridish, packed in aluminium foil and stored in a desiccators. The compositions of the different formulations were represented in Table 3 and Table 4[10].

Evaluation of Buccal Patches of Losartan Potassium

Evaluation of Physical Parameters

Physical Appearance and Surface Texture

Physical Appearance and Surface Texture includes visual inspection of patches and evaluation of texture by feel or touch.

Weight Variation Test

The patches of each formulation were cut into 2 X 2 cm^2 and five patches from each batch were weighed. Then the weight variation was calculated.

Thickness

The thickness of the patch was measured using a screw gauge at 10 different spots for each batch. From this mean value was calculated.

Folding Endurance

Folding endurance is used to determine the flexibility of patches. It was measured by folding patches at the same place till it broke.

Measurement of surface pH

For the determination of surface pH, patches were placed on the surface of the agar gel plate for 2 hours. pH paper was placed over the surface of the patch. A mean value of three readings was taken as a result[11].

Determination of Moisture Content and Moisture Absorption

Anhydrous calcium chloride was taken in a desiccator and accurately weighed buccal patches were placed on it. After 3 days, it was taken out and weighed. The moisture content (%) was determined by:

$$\text{Moisture content (\%)} = \frac{\text{Initial weight} - \text{Final weight}}{\text{Initial weight}} \times 100$$

For maintaining humidity 100 ml saturated solution of aluminium chloride was taken in a desiccator and placed with accurately weighed patches. It was then taken out and weighed after 3 days. The percentage of moisture absorption was calculated using the formula[12]:

$$\text{Moisture absorption (\%)} = \frac{\text{Final weight} - \text{Initial weight}}{\text{Initial weight}} \times 100$$

Swelling Studies

Agar gel plate (2% w/v) was prepared and previously weighed patches were placed over it. It was then kept in an incubator at 37^0 C. A definite time intervals of 1, 2, 3, 4, 5, 6, 7 and 8 hours the weight of patches were determined. The swelling index was found out by using the formula[8]:

$$\text{S.I} = \frac{W2 - W1}{W1} \times 100$$

Where- S.I. - swelling index, W1- weight of buccal patch before dipping into beaker, W2- weight of buccal patch after dipping in beaker and wiped

Evaluation of Perfomance Parameters

Drug Content Uniformity

Patches of 2 x 2 cm were dissolved by homogenization in a mixture of 5 ml ethyl alcohol and 2 ml of dichloromethane for 5 hour with occasional shaking and diluted to 50 ml with distilled water. It is then filtered to remove insoluble residue from the filtrate 1 ml was taken and was diluted to 10 ml with a buffer of pH 6.8 using U V spectrophotometer at 235 nm absorbance was measured[13].

***Ex-vivo* Bio adhesive Strength**

This method is used to measure *in vitro* bio adhesive capacity of different polymers. It is a modified method developed by Mertti Marvole[14]. Goat buccal mucosa was collected from slaughter house and it was stored in a buffer of pH 6.8. Within 3 hours the experiment was performed. Then the mucosa was washed and tied to a glass slide with the help of a thread. This portion was put in the petridish with a 6.8 buffer solution. During the experiment, the solution was kept at 37°C. The patch was stuck on to the glass stopper by using cyanoacrylate adhesive and that stopper tied with a thread. The other portion of that thread was tied to the plastic beaker. That patch was put on mucosa by applying finger pressure for 30 seconds. After making contact between the tablet and mucosa for a fixed time of 3 seconds; the water was added through a pipe connected to a burette containing water in a drop

wise to that plastic beaker. Then the weight of water required to detach buccal patch was measured.

***Ex-vivo* Bio adhesion Time**

By using cyanoacrylate glue, goat buccal mucosa was fixed inside the beaker above 2.5 cm from the bottom. The buccal patch was then pasted on to the mucosa by a light force by fingertip for 30 seconds. The beaker was filled with 500 ml of 6.8 buffer solution and maintained at $37^0c \pm 1^0c$. After 2 minutes, to simulate the buccal environment it was stirred at 50 rpm for 6 hours. Then the time required by the patch to detach from the buccal mucosa was calculated as mucco adhesion time[15].

***Ex-vivo* Permeation Study**

Ex-vivo drug permeation was performed using Franz diffusion. Goat buccal mucosa was placed between donor and receptor compartments. The temperature was maintained at 37 ± 0.5°C. The patch was then placed over the membrane and the receptor compartment was filled with 15 ml of phosphate buffer 6.8, stirred at 50 rpm. At definite time intervals (1, 2, 3, 4, 5, 6, 7 and 8 hours) 1 ml aliquot was withdrawn and replaced with the same volume of fresh medium. The collected samples were analyzed by U V spectrophotometer at 235 nm after proper dilution[16].

***In-vitro* Dissolution Study**

Rotating paddle apparatus was used to study drug release from patches. Temperature maintained at 37 ± 0.5°C, with a speed of 50 rpm using pH 6.8 phosphate buffer. The backing layer of the patch adhered to a glass slide by cyanoacrylate glue. Then it was placed at the bottom of the vessel. 5 ml of samples were withdrawn at specific time intervals and replaced with a fresh medium. After appropriate dilution, it was filtered and analyzed by U V spectrophotometer[8].

Kinetic Study

In short, the results obtained from *in vitro* release studies were plotted in four kinetics models of data treatment as follows: Zero order rate kinetics by plotting Cumulative percentage drug release Vs. Time, First order rate kinetics by plotting Log cumulative percentage drug retained Vs. Time, Higuchi's classical diffusion equation by plotting Cumulative percentage drug release Vs. √T and Korsmeyer-Peppas by plotting Log of cumulative percentage drug release Vs. log Time[18,19].

Stability Study

5 ml human saliva was taken in a petridish and patches were placed over it in a temperature-controlled oven at $37^0C \pm 0.2^0C$ for 8 hours. At definite time intervals, morphological and physical changes such as appearance, colour, shape, etc. were observed[17].

RESULTS AND DISCUSSION

Identification of Drug and Drug-Polymer Interaction Study

The IR spectrum of drug alone and in combination with natural mucilage, Ethyl cellulose and HPMC K100M suggested that the characteristic peak of drug was undisturbed and also the characteristic peak of polymer was unaffected. Hence the IR study reveals that drugs were in the free form and no drug-polymer and polymer-polymer interaction took place during formulation development. The IR spectra of drugs alone and in combination with the polymers are given in Figure 1 and Figure 2.

Standard Curve of Losartan Potassium

The standard curve of Losartan potassium was constructed in pH 6.8 buffer as the solvent system. Table 1 shows the absorbance readings of drug solutions containing 2-10 µg/ml of the drug in pH 6.8 buffer. Figure 3 shows standard curve for Losartan potassium.

Isolation and Purification of Gum

The mucilage from Zapota seeds isolated using appropriate extraction procedures (Figure 4).

Characterization of Mucilage

Macroscopic properties of mucilage such as colour and odour were performed. It was soluble in water and insoluble in Ethanol, Acetone and chloroform. pH, angle of repose, bulk density, tapped density, Hausner's ratio and Carr's index were calculated. The results are shown in Table 2.

Physical Appearance

All the buccal patches were visually inspected for clarity, flexibility and surface texture. They are having good physical appearance.

Weight Variation Test

Weight uniformity of all the patches were determined by weighing three 2 x 2 cm^2 sections of each patch and then average weight was calculated. All batches were uniform in weight and there was no significant difference. From the results shown in Table 5, the values were ranged from 87.36 ± 0.28 to 102.63 ± 0.46 mg.

Thickness

Average and standard deviation of all three readings were calculated and recorded in Table 5. It was found in the range of 0.09 ± 0.02 - 0.15 ± 0.06 mm. Thickness increases with increase in the concentration of polymers.

Folding Endurance

The recorded folding endurance of all the formulations was above the 200 and most of them are above 300, which indicate good flexibility. Table 5 shows the folding endurance value of all the formulations.

Measuring of Surface pH

Table 5 shows the result of average surface pH values for all formulations. These values represent the mean of three replicate determinations. They were found to be in the range of 6.10 ± 0.19 - 6.35 ± 0.10. The results were within the limit of acceptable salivary pH range of 5.69-6.34.

Determination of Moisture Content and Moisture Absorption

The moisture content (%) study was done for 3 days. The percentage of moisture content (%) is varied between 1.21 ± 0.03 to 1.31 ± 0.04% **(**Table 6). The moisture uptake values varied between 5.21 ± 0.05 to 6.13 ± 0.04% **(**Table 6). Low moisture content protects from microbial contamination and low moisture uptake (%) helps to retard hydrolytic degradation.

Table 1: Absorbance values of Losartan potassium

S. No.	Paramaters		Zapota polymer
1	Macroscopic Properties	Colour	Yellow
		Odour	Odourless
2	Solubility	Water	Soluble
		Ethanol	Insoluble
		Acetone	Insoluble
		Chloroform	Insoluble
3	pH		6.8
4	Angle of Repose (^{0})		27.34
5	Bulk density (g/cc)		0.36
6	Tapped density (g/cc)		0.45
7	Hausner's ratio		1.36
8	Carr's index (%)		26.11

Table 2: Pre formulation study of isolated gum

Concentration (µg/ml)	Absorbance (nm)
0	0
2	0.1099
4	0.2246
6	0.3378
8	0.4398
10	0.5532

Table 3: Composition of Losartan potassium mucoadhesive buccal Patches

Formulation code	Ingredients (mg)						
	Losartan potassium	Zapota polymer	HPMC K-100M	Citric acid	Sucra-Lose	Propylene glycol (ml)	Dist. water (ml)
F1	397.40	50	300	0.5	0.5	1	10
F2	397.40	100	300	0.5	0.5	1	10
F3	397.40	150	300	0.5	0.5	1	10
F4	397.40	200	300	0.5	0.5	1	10
F5	397.40	250	300	0.5	0.5	1	10

Table 4: Composition of backing membrane

Ingredient	Quantity
Ethyl cellulose	1.5 gm
Acetone	19 ml
Isopropyl alcohol	11 ml
Dibutyl phthalate	2 ml

Table 5: Evaluation of Physical parameters- Average weight, thickness, folding endurance and surface pH of Losartan potassium mucco adhesive buccal patches

Formulation code	Average weight (mean ± S.D.) (mg)	Thickness (mm)	Folding Endurance	Surface pH
F1	87.36 ± 0.28	0.09 ± 0.02	268	6.10 ± 0.19
F2	90.15 ± 0.72	0.11 ± 0.01	280	6.25 ± 0.08
F3	95.26 ± 0.26	0.12 ± 0.02	>300	6.22 ± 0.15
F4	99.79 ± 0.51	0.13 ± 0.07	>300	6.26 ± 0.42
F5	102.63 ± 0.46	0.15 ± 0.06	>300	6.35 ± 0.10

Table 6: Evaluation of Physical parameters - Moisture Content (%) and Moisture Uptake (%) of Losartan potassium mucco adhesive buccal patches

Formulation code	Moisture content (%)	Moisture uptake (%)
F1	1.31 ± 0.04	5.21 ± 0.05
F2	1.28 ± 0.01	5.48 ± 0.08
F3	1.10 ± 0.03	5.66 ± 0.14
F4	1.06 ± 0.03	6.06 ± 0.10
F5	1.21 ± 0.03	6.13 ± 0.04

Table 7: Evaluation of Physical Parameters – Swelling Index of Losartan potassium mucco adhesive buccal patches

Formulation code	Time (Hours)					
	1	2	3	4	5	6
F1	20.65	24.98	41.67	48.15	55.74	54.51
F2	23.74	35.51	43.17	50.65	53.19	58.74
F3	27.38	40.78	46.29	54.40	55.12	58.98
F4	31.74	45.96	44.86	50.14	58.15	60.75
F5	35.40	38.34	52.61	57.40	60.19	68.65

Table 8: Evaluation of performance parameters – Drug content uniformity, Measurement of bio-adhesion time and bio-adhesion strength of Losartan potassium mucco adhesive buccal patch

Formulation code	Drug content Uniformity	*Ex-vivo* bio adhesion strength (gram)	*Ex-vivo* bio adhesion time (hours and minutes)
F1	99.74 ± 0.25	20.25 ± 0.17	7 hr 35 min
F2	99.26 ± 0.04	24.65 ± 0.09	7 hr 45 min
F3	99.45 ± 0.15	32.76 ± 0.31	7 hr 55 min
F4	100.36 ± 0.22	33.54 ± 0.42	9 hr 20 min
F5	99.48 ± 0.17	34.45 ± 0.19	10 hr 25 min

Table 9: Evaluation of performance parameters – *Ex-vivo* permeation study of Losartan potassium mucco adhesive buccal Patches

Form. Code	Time (min)								
	0	60	120	180	240	300	360	420	480
F1	0	16.24	24.56	36.17	45.62	57.41	66.53	77.62	87.23
F2	0	20.19	29.46	38.46	46.74	58.32	70.26	76.24	83.55
F3	0	19.76	31.25	41.27	52.45	61.76	72.77	79.24	81.08
F4	0	17.45	28.26	39.68	47.51	55.01	63.26	71.52	81.85
F5	0	14.19	26.20	33.20	41.74	53.11	64.57	71.39	79.22

Table 10: *In-vitro* release data of Losartan potassium mucco adhesive buccal patches

Form. Code	Time (min)										
	0	60	120	180	240	300	360	420	480	540	600
F1	0	17.55	24.98	38.79	54.58	67.23	76.56	87.66	99.09	-	-
F2	0	18.29	30.55	46.17	57.7	66.64	79.28	90.69	99.23	-	-
F3	0	16.51	27.58	39.74	50.4	61.85	72.86	81.52	90.83	99.74	-
F4	0	18.43	28.75	40.86	52.37	63.39	75.57	84.32	92.28	98.74	-
F5	0	15.54	26.55	35.41	47.45	56.24	65.42	74.16	83.78	91.5	99.26

Table 11: Regression analysis of the *in-vitro* release data of Losartan potassium mucco adhesive buccal patches according to various release kinetic models

Formulation code	Zero order R^2	First order R^2	Higuchi R^2	Korsmeyer-Peppas	
				R^2	N
F1	0.9955	0.9388	0.9308	0.9858	0.8712
F2	0.9917	0.9162	0.954	0.9985	0.8188
F3	0.9946	0.9177	0.9513	0.9989	0.8271
F4	0.9926	0.9204	0.9577	0.9966	0.789
F5	0.9942	0.914	0.9542	0.999	0.8115

Table 12: Stability data of formulation F5 in Human saliva

Time (Hours)	Colour change	Thickness	Change in pH	Collapsing
0	No change	0.16	No change	No change
1	No change	0.17	No change	No change
2	No change	0.20	No change	No change
4	No change	0.22	No change	No change
6	No change	0.25	No change	No change
8	No change	0.26	No change	No change

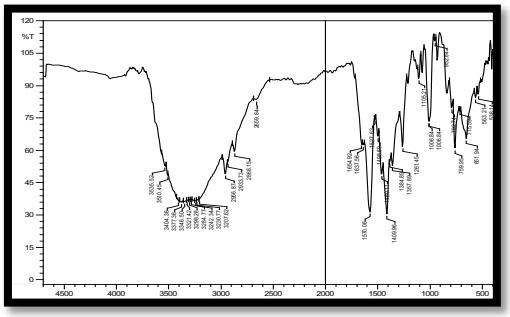

Figure 1: FTIR Spectrum of Losartan potassium

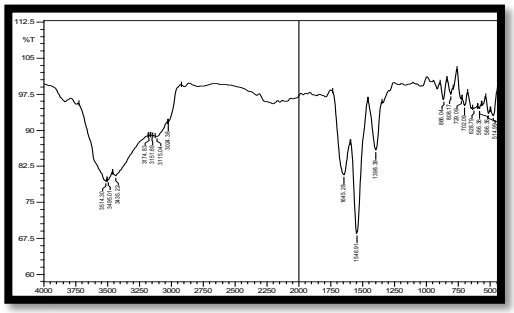

Figure 2: FTIR Spectrum of Losartan potassium + Zapota polymer + HPMC K 100M+ Ethyl cellulose

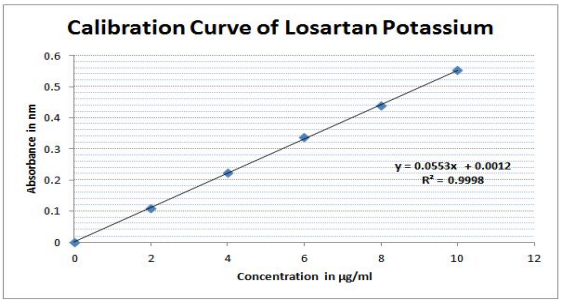

Figure 3: Calibration Curve of Losartan potassium

Figure 4: Zapota polymer

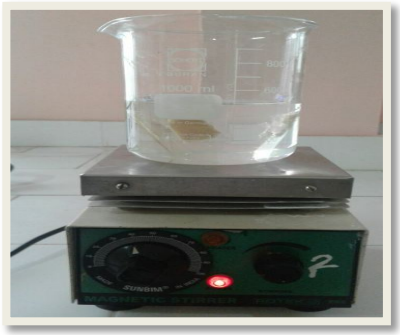

Figure 5: Measurement of bio adhesion time

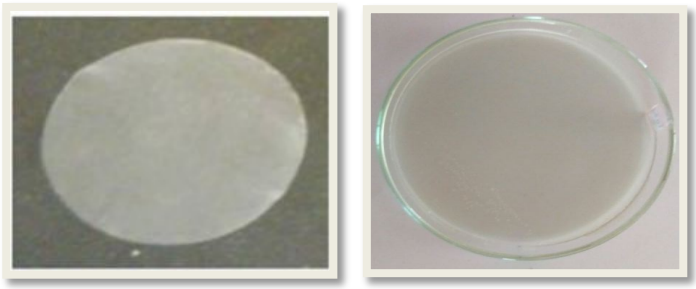

Figure 6: Mucoadhesive buccal patches

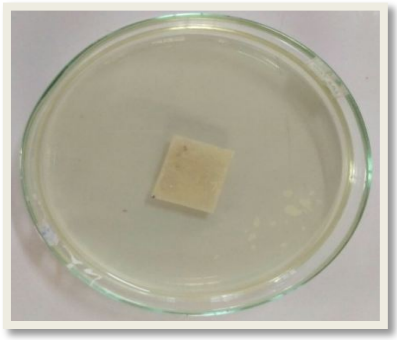

Figure 7: Swelling study

Figure 8: *Ex-vivo* bio adhesion strength

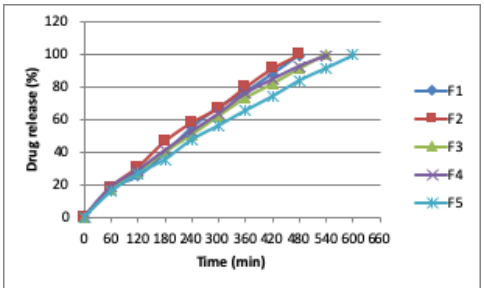

Figure 9: *In-vitro* Release graph

Swelling Studies

Swelling studies were performed to investigate the performance of the dosage form, swelling capacities and patch integrity after swelling. Maximum swelling of 68.65% (F5) and minimum swelling of 54.51% (F1) was observed for patches. Table 7 shows the folding endurance value of all the formulations.

Drug Content Uniformity

Drug content of all the formulations was determined using UV-Visible spectrophotometer. Table 8 represent the values. Drug content was found to be in the range of 99.74 ± 0.25 to 100.36 ± 0.22%.

***Ex-vivo* Bio adhesion Strength**

All the batches showed good mucco adhesive strength. Results were shown in Table 8. Mucco adhesive strength of the formulation increases with increase in concentration of the Mucco adhesive polymer. Maximum strength bio adhesion was observed for F5 (34.45 ± 0.19 g) and minimum for F1 (20.25 ± 0.17 g).

***Ex-vivo* Bio adhesion Time**

The bucco adhesion time was evaluated and reported in Table 8. Maximum bucco adhesion time was shown by formulation F5 which was 10 h 25 min. and minimum bio adhesion time was 7 h 35 min by F1.

***Ex-vivo* Permeation Study**

The *ex-vivo* drug permeation studies were performed using sheep buccal mucosa as a model membrane using Franz diffusion cell. The study was conducted at 37 ± 2 °C for 8 h. The result of *ex-vivo* drug permeation study is shown in Table 9.

***In-vitro* Dissolution Study**

The data obtained from *in-vitro* drug release study performed up to 8 h gives a clear indication that prepared patches showed necessary controlled release profile. The results for release studies are shown in Table 10 and Figure 9.

***In-vitro* Drug Release Kinetics**

For all the formulations, various kinetic models were applied and results were interpreted. On the basis of kinetic assessment, the values were obtained and the best fitted model was decided. Data is shown in Table 11.

Stability Studies

The stability of the optimized buccal formulation in human saliva was performed. The results are shown in the Table 12. The following parameters like colour change, thickness, change in pH and collapsing were determined for an interval of about 0, 2, 4, 6 and 8 hours.

CONCLUSION

The patches of Losartan potassium have appropriate folding endurance, moisture content and moisture uptake, swelling index, bio adhesion strength and time. The above study demonstrated the possibility of designing and developing mucco adhesive patches of Losartan potassium using zapota gum, which will more ideal than conventional drug delivery. It can also be chosen as a system for the controlled delivery of antihypertensive drugs.

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