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Formulation and evaluation of mucoadhesive buccal patch containing Imipramine hydrochloride

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ABSTRACT

The study was intended to surpass the related problems of reduced oral bioavailability, to reduce the serious side effects associated with other routes and to study the suitability of formulating the drug into a mucoadhesive buccal patch for the systemic drug delivery. The mucoadhesive buccal film of imipramine hydrochloride was prepared using varying concentrations of HPMC E15 and Carbopol 940 by Solvent casting technique, which produced a transparent colourless patch. Physical evaluation, in-vitro drug release studies, ex-vivo drug permeation studies, histopathological studies, stability studies etc. were carried out on these formulated buccal patches and responses such as in-vitro drug release and ex-vivo drug permeation being used for optimization. The average thickness of the patch was found to be in the range of 0.01746 to 0.2488µm. The in-vitro drug release studies showed 93.39% drug release from formulation FVI and the stability studies carried out at two different temperatures showed patches remained stable even after 45 days. This study concludes that mucoadhesive buccal patches that were prepared were safe, convenient and effective formulation for the systemic delivery of an antidepressant drug, imipramine hydrochloride. Besides this, it provides sustained delivery of the drug into the systemic circulation and has least gastrointestinal side effects compared to the marketed formulation.

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INTRODUCTION

There are several routes for the delivery of drugs into the human body. One of the most acceptable routes is the buccal route of drug delivery. Mechanisms like hepatic first-pass metabolism and GI tract enzymatic degradation are the disadvantages of peroral administration; hence this results in prohibiting administration of drugs containing peptides and proteins orally. Mucosal lining is present in various regions of our body like a nasal cavity, vagina, ocular cavity and an oral cavity which are very effective in the delivery of many drugs (Aungst *et al.,* 1988). Among all these buccal route mucosal drug delivery is more easy and simple. First pass effect is bypassing, prevention of presystemic elimination within the GI tract, good enzymatic flora for drug absorption are the advantages of this buccal route.

Benefits of mucoadhesive delivery system on conventional delivery methods are in terms of controlled drug delivery, comparatively large permeability as the mucous membrane is well vascularised which rapidly uptake drug into the systemic circulation and improved bioavailability. Mucoadhesion, the ability to hold on to the mucosal layer for a specific period, is the main part in the design of these novel drug delivery systems. Due to biocompatible and biodegradable properties

Table 1: Composition of formulation

FORMULATIONS	PATCH CODE					
	F I	F II	F III	F IV	F V	F VI
Imipramine hydrochloride (mg)	15	15	15	15	15	15
HPMC E15 (mg)	200	100	150	66	134	50
carbopol 940 (mg)	---	100	50	134	66	150
Glycerine (ml)	2	2	2	2	2	2
ethanol (ml)	7	---	7	---	---	7
tween 80 (g)	---	0.0315	0.0315	---	---	0.0315
water (ml)	---	7	---	7	7	---

natural polysaccharides have been used extensively (Amanpreet and Gurpreet., 2012).

MATERIALS AND METHODS

Materials

Imipramine HCl IP collected from Sigma Aldrich, Mumbai. HPMC E15LV, Carbopol 940 and potassium bromide are taken from Loba Chemie Pvt.Ltd, Mumbai. Glycerine, tween 80 and ethanol from Nice chemicals Pvt.Ltd, Cochin. Disodium hydrogen phosphate from Fisher scientific and potassium dihydrogen orthophosphate from Spectrum, Cochin.

Methodology Preformulation studies

Different preformulation studies like the determination of melting point of drug sample using the open capillary method, determination of absorption maxima of the drug in phosphate buffer pH6.8 & determination of solubility in different solvents were done (Shamika. *et al.,* 2017).

FTIR Studies

Using Infrared Absorption Spectral Analysis (FTIR), compatibility of the drug with individual excipients and physical mixture were established. Then using IR spectral analysis, changes in physical composition.

Partition Coefficient of Drug

The partition coefficient of imipramine hydrochloride in n-octanol and phosphate buffer pH 6.8 was determined (Yogeshwar and Vandana B., 2009).

Compatibility Studies of The Drug With Excipients

Study of compatibility was done to select the excipients which are added to assist administration and provide proper (Amit *et al.,* 2009).

Physical Drug Excipient Compatibility

Both the stability studies at higher temperature and humidity conditions were coupled with physical compatibility studies. The primary states of the mixtures were noted. For the checking of the possible happening of any interactions, evaluation was performed on 15th and 30th day. (Vidya *et al.,* 2015).

Preparation of polymeric films of hydroxypropyl methylcellulose (HPMC E15) and Carbapol 940

By solvent casting method films were prepared using HPMC E15 and Carbopol 940 in different concentrations (Sneh Priya *et al.,* 2011 and Anuj *et al.,* 2011). Higher levels of HPMC E15 gave the films more soft and wet. Glycerol acts as the plasticizer. HPMC E15 polymer made to solution by dissolving in 2ml ethanol. For swelling of the polymer, beaker with polymer solution was kept aside for 4 hrs. To the above polymer solution, 3ml ethanol added and was stirred. Then 1 drop (0.0294g) of glycerol further added. Drug 15mg accurately weighed and dissolved in a separate beaker with 1ml ethanol. Drug solution poured to the polymer solution and mixed with a magnetic stirrer. The whole solution poured into the glass mould.

This was kept at room temperature for 12 hours for drying. After removing from the mould checked for imperfections. With wax paper that was covered and in desiccators they were preserved until evaluation tests performed. Six formulations FI, FII, FIII, FIV, FV and FVI, were prepared using the above method. For drying FI, FII, FIII, FVI kept about 12 hours at. FIV and FV were kept for 72hrs at room temperature for drying. The composition of various films is shown in Table 1.

Evaluation of prepared buccal films

Thickness Uniformity of the Patches

From each formulation, three patches were taken. At three different places micrometre screw gauge was placed and measured patch thickness and mean value calculated (Ashwini *et al.,* 2011).

Folding Endurance

Three patches from each formulation with size 2×2 cm2 cut by means of a sharp blade. Folding endurance was determined. The mean value was calculated.

Uniformity of Weight of the Patches

Three patches from each formulation of size 1×1cm2 taken and weighed on a digital balance. The average weights were calculated.

Percentage Swelling Index

The polymeric patches incise into 1×1 cm2 were weighed exactly and kept immersed in 50ml of phosphate buffer pH6.8. At 5, 10, 30, 60 minutes intervals patches were taken out carefully. Blotted using filter paper for removing buffer solution on their surface and weighed accurately until constant weight was observed weight increase in the film was evaluated at the time, and then percent swelling was calculated with the formula (Krishnapriya *et al.,* 2017).

Drug Content Uniformity of Patches

From each formulation, three patches took into the separate 100ml volumetric flask. The amount of drug in the buccal patches being determined by dissolving 1cm^2 patch in 100ml phosphate buffer saline (pH6.8) and shaken vigorously at room temperature for 24 hours (Surya *et al.,* 2011). Using Whatman filter paper (no. 42) this solution filtered and diluted properly and analysed in UV spectrophotometer at 250 nm. Final reading was taken from averages of the three patches.

Surface PH of Patches

Three patches of each formulation permitted to swell and it was done by allowing it to get contact with 0.5ml of distilled water (pH6.5±0.5) for about 1 hour in room temperature (Anuj *et al.,* 2011). pH was marked down by making electrode touch in with the surface of the patch.

Ex-vivo Mucoadhesive Strength

The mucosal membrane was separated from a fresh buccal mucosa of goat from which fat and loose tissues were removed. Using distilled water and the with isotonic phosphate buffer of pH 6.8 at 37°C, the membrane was cleaned and the bioadhesive strength of the patch was calculated (Varinder *et al.,* 2011).

Method

The exposed surface of the patch then moistened with 50µl of isotonic phosphate buffer of pH6.8 for 30 seconds. To set up the adhesion between the patch and the mucosal tissue the preload about 50gm placed in the right pan and whole gathering was set aside uninterrupted for 3 minutes (preload time). Once 3 minutes over, preload is being removed and weights were added till detachment of patch took place from mucosal surface. Find out the total weights added. Subsequent to each measurement the tissue was quietly washed with isotonic phosphate buffer of pH 6.8, then it was left for 5 minutes prior to the take of reading. Three times the experiment was performed. The mass in grams taken to separate the patch from the mucosal surface gave the measurement of mucoadhesive strength. The force of adhesion (N) and bond strength (Nm^2) were calculated from the bioadhesive strength.

Ex-vivo Mucoadhesion Time

After application of the buccal patch on a fresh piece of goat buccal mucosa, the in-vitro residing time was examined. The buccal mucosa being tied on glass slide and side of each mucoadhesive core of each patch moistened using one drop of phosphate buffer with pH 68 and its been sticked to buccal mucosa with applying a light force by fingertip for 30 seconds To the beaker with 200ml phosphate buffer of pH 6.8 the glass slide was dipped and kept at 37±1°C. After 2 minutes, it was stirred slowly. The time required to detach the patch from buccal mucosa was recorded as mucoadhesion time (John *et al.,* 2013).

Ex-vivo Drug Permeation Studies

The histological and biochemical properties of goat buccal mucosa repetitively prove that it is like that of human skin (Upendra and Siddharth., 2010).

Tissue Preparation

The buccal mucosal membrane washed with Ringer's solution and it was permitted to equilibrate for about 1 hour in receptor buffer for regaining the elasticity that has been lost (Subhashree *et al.,* 2011).

Storage

Membrane surface cleaned with Ringer's solution and was kept to dry (exposed to ambient air conditions for 20 minutes). Then it is being packed in aluminium foil and stored in a polyethylene bag at -2°C (Navneet and Pronobesh., 2011).

Permeation Studies

1 ml sample was withdrawn at a preset time like 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12 hours from the patch that placed in donor compartment of Franz diffusion cell apparatus and the cells refilled filled again with same fresh receptor solution. These samples then analysed spectrophotometrically at 250.5 nm.

The cumulative amount of drug permeated per cm2 versus time was plotted and with the slope of the linear portion, the steady-state flux (Jss) calculated (Arpita *et al.,* 2011).

The apparent permeability coefficient (Kp) were calculated. Time (T) was calculated by the X-intercept of the linear portion of the curve. With a

known thickness of the skin (δ) and the lag time (T), the diffusion coefficient was calculated.

Permeation of Imipramine Hydrochloride from Aqueous Solution

To verify whether the drug is being capable of going through the buccal mucosa and to know the extent of permeation. The drug in the most obtainable form, which is an aqueous solution (15 mg in 5ml of simulated saliva), kept in the donor compartment. At fixed time intervals, the 1ml sample was withdrawn and diluted with pH 7.4 and analysis was carried out using UV spectrophotometer at λmax of 250.5nm (Subhash *et al.,* 2009). Triplicates the same experiments.

Permeation of Imipramine Hydrochloride from Formulated Films

A significant percentage of drug permeated from the solution. Since the drug has good buccal permeability, it is apt for more study from film formulations. To previously moistened mucosal surface, the patch was made to be in contact with. The simulated saliva of pH 6.8 was filled in the donor compartment. As described above the experiment was conducted. The data obtained were statistically analysed.

Skin Deposition Studies

Following the performance of in-vitro permeation study for 12 hours, using methanol, the donor compartment was washed. Using methanol as a receptor solution, buccal mucosa was extracted for an additional period of 12 hours and the quantity of the drug was quantified by spectrophotometry at 250.5nm (Alanazi *et al.,* 2007).

In-vitro Release Studies

Type II (paddle type) USP dissolution apparatus was chosen to perform the in-vitro release studies. The medium was 900ml of isotonic phosphate buffer pH 6.8 as the at 37 ±0.5°C and 50 rpm. For unidirectional liberation, one side of the patch attached to a glass disk and the disk was kept into the bottom of the dissolution vessel, so that patch surface get an exposed medium (Mohammed and Sadath., 2011). At present time intervals, an aliquot of 5ml sample was introverted and comparable volume was added with fresh phosphate buffer pH 6.8 kept at same temperature. These samples then analysed spectrophotometrically and average cumulative percentage drug released was determined. The data obtained were statistically analysed.

Kinetic Analysis of In-vitro Release Data

To analyse the drug release method of each formulation, all the batches dissolution profile were fitted to zero-order, first-order, Higuchi, Hixon-Crowell, Korsmeyer and Peppas models (Rohit *et al.,* 2010).

Statistical Design of Experiment

Using Statgraphics Centurion 16, the statistical design of the experiment was carried out.

ANOVA Study

ANOVA analysis carried out based on P value less than 0.05 which was considerable at a level of significance α =n0.05.

Mass Balance Study

Analysis of the drug content left in the patch was done. Each patch properly diluted by dissolving in water (Khairnar and Sayyad., 2010). The actual drug content was calculated.

Scanning Electron Microscopy

With the help of Scanning Electron Microscopy, film morphology was considered. Using double-backed adhesive tape samples being mounted on round brass slabs (12mm diameter).

Comparison Studies

In-vitro comparative study of the optimized FVI formulation with a marketed formulation.

The optimized buccal patch was compared with a marketed formulation (Imiprin 25 mg tablet) for the in vitro drug release (Rita *et al.,* 2006). For the comparison studies, the cumulative percentage amount of drug released was observed.

Histological Studies

Buccal Mucosa Preparation

The obtained Goat buccal mucosa, which was cleaned after the removal of underlying tissues was then dried and stored in a polyethene bag at -2°C (Kapil *et al.,* 2010).

Preparation of Mucosal Membrane for Morphological Studies

Buccal mucosa which was set in 10% buffered formalin (pH 7.4) was made into vertical sections (Avinash *et al.,* 2011). Each section was hydrated with ethanol, and implanted in paraffin to fix. On glass slides, these Paraffin sections (7 µm) were kept and stained with haematoxylin and eosin (HE). Spoil to the tissue through in vitro permeation sections were observed under light microscope.

Stability Studies

Medicated patches that are chosen for stability testing. For the determination of physical and chemical stabilities patches were positioned in a glass beaker lined with aluminium foil and kept in room temperature (30±2°C) and refrigerator

temperature (4±2°C) for 45 days. Changes in appearance and drug content of the stored patches evaluated each weekend. The data offered were the mean of three determinations.

RESULTS AND DISCUSSIONS

Preformulation studies

Melting Point of the drug was found to 172°C similar to a monograph (170-174°C). The λmax of the drug was found 250.5nm in phosphate buffer pH 6.8 and the results were in accordance with the official standard. The drug was freely soluble in water, phosphate buffer pH 6.8, 95% ethanol and in chloroform.

FTIR Studies

Compatibility of drug excipient was also established by FTIR and the spectra of the drug and drug excipient combinations are shown in figure1 and figure 2.

The characteristic peaks of the drug such as 756 cm^{-1} for the substituted aromatic ring, 1230 cm^{-1} for bending vibrations of methylene group and 1110 cm^{-1} for aliphatic amine were present in the spectrum of the drug-carbopol-HPMC blend, which indicates that the activity of the drug unaffected when it is combined with polymers for making the formulation. From the spectra, it is clear that the activities of the whole compound, as well as the activity of the characteristic groups of imipramine hydrochloride, were unaffected. It concluded that imipramine hydrochloride has no interaction with the ingredients of the buccal patch.

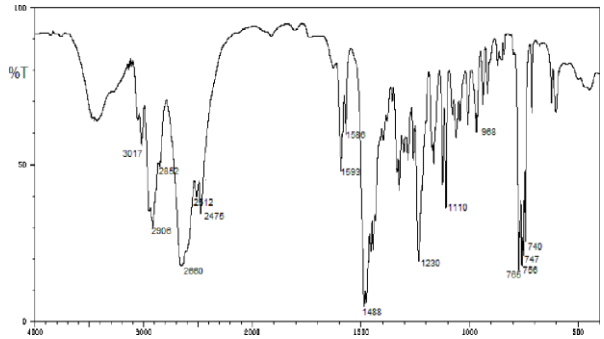

Figure 1: FTIR Spectrum Drug

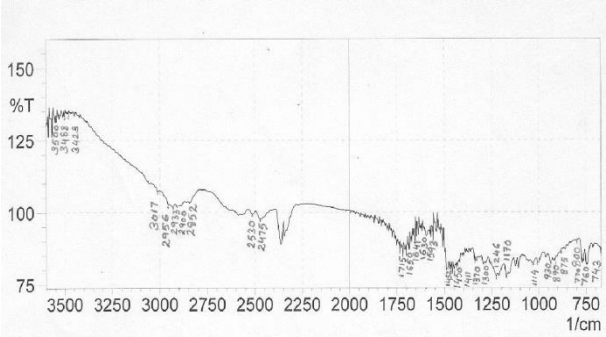

Figure 2: FTIR spectrum of Drug – Carbopol 940 – HPMC E 15 blend

Partition Coefficient of Drug

The drug partition coefficient being observed as found to be 3.9 and it has good lipid solubility. So it can be effectively formulated into a mucoadhesive buccal patch for efficient transport across the buccal mucosa

Compatibility study of the drug with excipients

Physical Drug Excipient Compatibility

The drug excipient compatibility study reveals that there were no incompatibilities. Based on the physical compatibility value, the excipients were taken.

Formulation of Mucoadhesive Buccal Patch

Using solvent casting method mucoadhesive buccal patches were organized. HPMC E 15 and carbopol 940 were used as the polymers for the preparation of patches. HPMC E15 gave soft and fragile patches. Carbopol 940 and HPMC combinations with the highest proportion of carbopol polymer gave good transparent patches. Patches obtained were of matrix type were drug uniformly distributed in the polymer matrix. Tween 80 is used as the non-ionic surfactant for the study, in specific formulations. This helps in finding out the effect of surfactants on drug release from the polymer matrix. Water and ethanol were used as the solvents in the preparation of patches. Carbopol 940 shows higher mucoadhesive property than Hydroxy Propyl Methyl Cellulose. Surface wettability and swelling of the films can be increased by fast disintegration of hydrophilic polymers. These polymers absorb water, swell up and release drug from the matrix. Hence the patch with a higher concentration of carbopol shows good mucoadhesive property and so a sustained drug release pattern when compared to all other formulations. Different proportions of polymers were used in these formulations. Films with a higher percentage of carbopol and ethanol as the solvent could be easily removed from the Petri dish and showed good physical characteristics. Films prepared from HPMC alone using water as the solvent were more fragile, moist and soft in nature. Glycerol was added in all formulations which act as a plasticizing agent. The present study was focused to investigate the various concentration of HPMC E15 and carbopol 940 in the preparation of a good buccal patch and to find out the drug release and permeation through the buccal mucosa. Total six formulations were prepared and were evaluated for a variety of parameters.

Characterisation of mucoadhesive buccal patch

All the buccal patches were evaluated for various parameters. Films formulated from a higher concentration of carbopol were smooth, transparent, whereas those prepared from HPMC alone were

more fragile, soft and moist in nature. All films were translucent and flexible.

Thickness Uniformity of Patches

Results of this test reveal that none of the film samples shows much variation within each formulation. The average thickness was seen to be in the range of 0.1746 to 0.2488. With a micrometre, screw gauge patch thickness was measured.

Folding Endurance

Even subsequent to 295 times folding, no cracks were observed in films, which indicates that they are hard and elastic. Also, there was no much variation in comparison among plain films and drug-loaded films.

Uniformity of Weight of Patches

The patches which were loaded with drugs found to be uniform and the average weight of patch was in the range of 15.357mg to 29.344mg.

Percentage Swelling Index

Swelling has increased the area of each patch. The maximum swelling takes place in the formulations containing a higher proportion of carbopol namely FVI and the least in those containing a higher proportion of Hydroxy Propyl Methyl Cellulose as shown in figure 3. Films with HPMC showed a proportional swelling index and carbopol shows as an inversely proportional swelling index. Carbopol and HPMC are hydrophilic, but carbopol is more mucoadhesive in nature than HPMC.

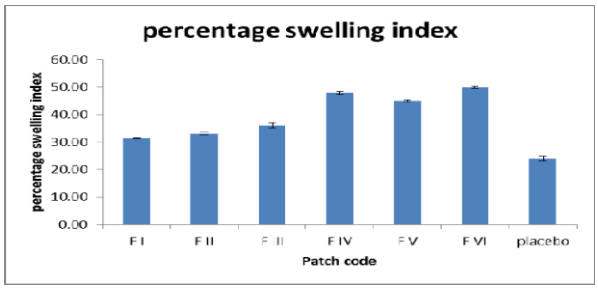

Figure 3: Percentage swelling index of six formulations along with placebo

Drug Content Uniformity of Patches

Film formulations of imipramine hydrochloride with HPMC and carbopol polymers gives uniform drug content.

Surface pH of Patches

Irritation of buccal mucosa can occur by acidic or alkaline pH. The surface pH of the prepared formulations was observed to be close to neutral pH as shown in table 2. This indicates that the potential to irritate mucosa is less and is quite comfortable for buccal use.

Ex-vivo Mucoadhesion Strength

Formulations with higher proportions of mucoadhesive polymer carbopol namely FVI shows higher bioadhesive force. Carbopol and HPMC are hydrophilic in nature but compared to HPMC, Carbopol is more mucoadhesive in nature. So it will hydrate faster attaining utmost swelling at shorter periods which aid interpenetration of a polymer chain within the tissue which is represented in figure 4and 6.

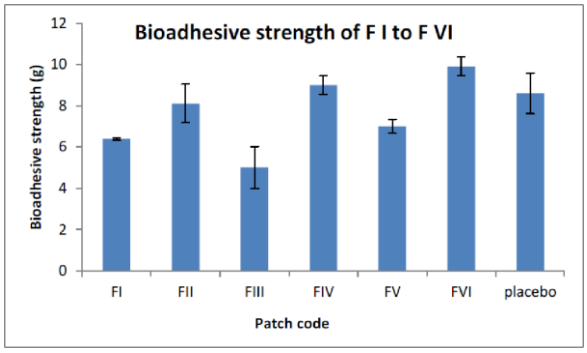

Figure 4: Bioadhesive strength of six formulations along with placebo

Ex-vivo Mucoadhesive Time

In the films with a larger quantity of carbopol, observed the greatest mucoadhesion time, which is apt for buccal drug delivery is represented in figure 5.

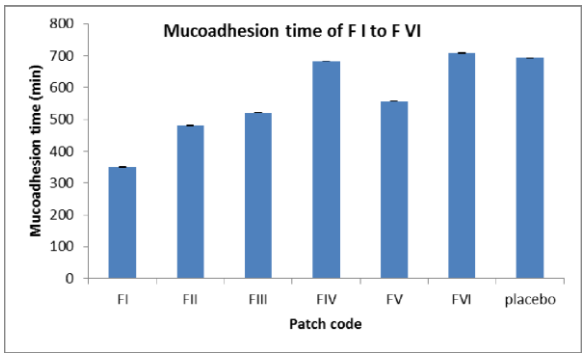

Figure 5: Mucoadhesion time of six formulations along with placebo

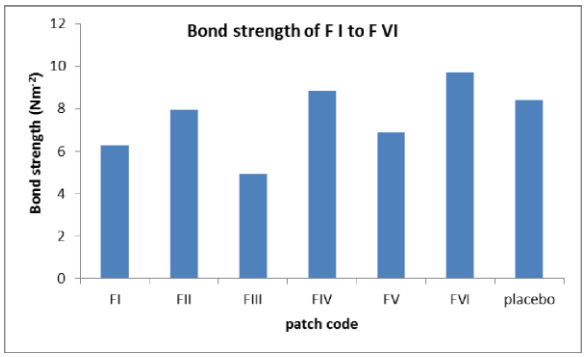

Figure 6: Bond strength of six formulations along with placebo

Table 2: Characterization of mucoadhesive buccal patches

Patch code	TN (mean*±Std)	FE (mean*±Std)	WU (mg) (mean*±Std)	%DC (mean*±Std)	Surface pH (mean*±Std)
I	0.2046±0.015	>295	21.506±0.05	96.70±0.013	6.74±0.98
II	0.1746±0.04	>295	17.493±0.05	92.68±0.04	6.54±0.59
III	0.1826±0.036	>295	15.357±0.92	95.60±0.64	6.85±0.87
IV	0.2486±0.032	>295	28.403±0.04	91.62±0.035	6.85±0.76
V	0.2116±0.026	>295	29.344±0.92	98.89±0.07	6.74±0.45
VI	0.1926±0.023	>295	22.256±0.99	99.0±0.063	6.63±0.63
Placebo	0.1123±0.023	>295	12.74±0.87	------	5.99±0.67

*The values are expressed as Mean ± STDEV; n=3.TN = thickness uniformity of patches, FE = folding endurance, WU = uniformity of weight of the patches, %DC = percentage drug content in the patches

Table 3: Permeation parameters

Patch code	Flux (µg/cm2/h)	KP (cm2/hr.)	Enhancement Ratio
FI	31.54	0.0389	0.1736
FII	51.275	0.06322	0.2822
FIII	61.545	0.07589	0.3387
FIV	46.479	0.05731	0.2558
FV	52.461	0.06468	0.2887
FVI	68.941	0.08500	0.3794
Control	181.7	0.2240	--

Ex-vivo Drug Permeation Studies

The quantity of drug absorbed into the blood is obtained from the data on the quantity of drug permeated. The differences in the concentration of polymers used in the formulations play a role in the permeation of drug a along the buccal mucosa. The hydrophilic polymers used in the formulations can absorb saliva and allows the drug to escape the drug from the polymer matrix. The ex-vivo permeation profile of all film formulation is shown in figure 7 permeation parameter of all the formulations are represented in table 3. The permeation data showed that an increase in the carbopol content cause enhances in the permeation rate. In all the six formulations lag time was not observed and this indicated that only a very small amount of drug was retained on the buccal mucosal surface after the release from the formulation. Hence there was no resistance to the diffusion of the drug across the buccal mucosal surface and therefore there was no diffusion coefficient observed for all the formulations. The mucoadhesive buccal patch with the highest percentage of drug permeation that is formulation FVI showed 0.3794 times enhanced rate of permeation compared to the control that is drug in phosphate buffer pH 6.8 which is represented in figure 8. The non-ionic surfactant present in the formulation, which is necessary for the modification of the structural composition causes the increased enhancement ratio and which results in an increase in the drug permeation across the buccal mucosal layer.

ANOVA Study

From ANOVA analysis of permeation flux P- value is 0.0100. As the P- the value of the F-test is less than 0.05, there is a statistically considerable difference between the means of the 6 variables at the 95% confidence level.

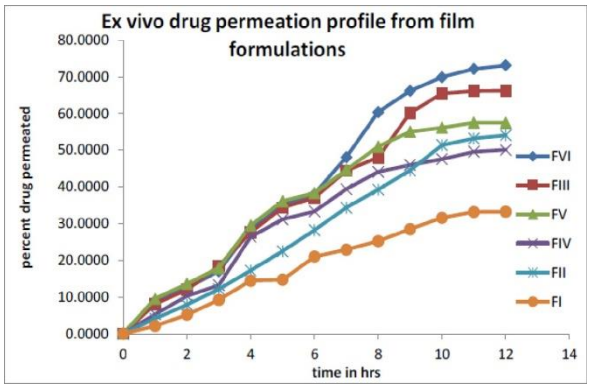

Figure 7: Ex-vivo drug permeation studies of all formulations

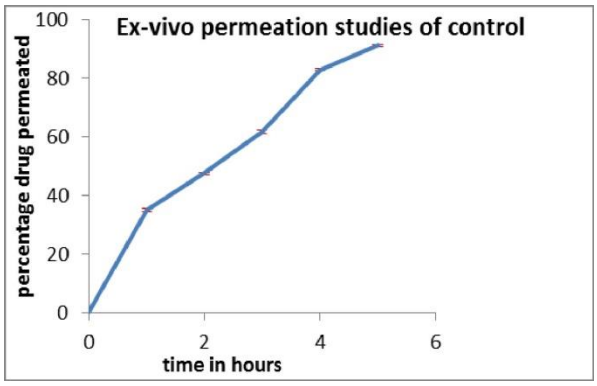

Figure 8: Ex-vivo drug permeation studies of control

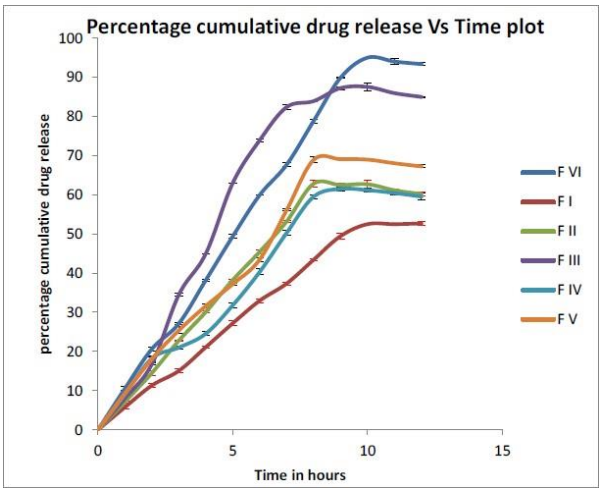

Figure 9: In-vitro release studies of formulations F I to F VI

Mucosal Deposition Studies

Percentage drug deposition in the buccal mucosal layer from various formulations and data obtained from the transmucosal flux of different formulations show a linear relationship between transmucosal flux and percentage drug deposited on the buccal mucosal layer for that formulation. A decrease in transdermal flux leads to the deposition of a higher concentration of drug on to the buccal mucosal layer and this is tabulated in table 4.

Table 4: Skin deposition studies of formulations

Patch Code	Percentage of drug deposited
FI	3.43%
FII	2.30%
FIII	1.53%
FIV	2.65%
FV	1.91%
FVI	1.21%

In-vitro Drug Release Studies

The in-vivo behaviour and function of the delivery system are evaluated from the drug release profile. All these six formulations were selected and underwent in-vitro release studies for 12 hours and it is represented in figure 9.

In-vitro drug release studies in phosphate buffer pH6.8 showed 93.39% drug release for film formulation containing a higher proportion of carbopol polymer that is FVI after 12 hours and formulation FI with HPMC alone showed a lower percentage of drug release and this is due to larger proportion of carbopol 940. Since HPMC is comparatively less mucoadhesive than carbopol, it showed a lower percentage or drug release.

The release profile suggests that the release of the drug occurs slowly and in a sustained manner. When a mucoadhesive buccal patch is applied on the buccal mucosa, it will absorb saliva and this leads to loosening of bonds in the hydrophilic polymer, which leads to the drug release through the polymer matrix. Therefore the release occurs slowly and in a sustained manner. From the release profile, it is clear that the formulation with a higher concentration of carbopol polymer that is formulation F VI shows the highest release of about 95.05% and formulation with HPMC alone that is formulation F I shows the lowest release of about 54.42%.

ANOVA Analysis

ANOVA analysis of in-vitro drug release shows P-value of 0.010. Since P-value of the F- the test is less than 0.05, there is a statistically considerable difference between the means of the 6 variables at the 95% confidence level.

Kinetic Analysis of In-vitro Release Data

For the characterisation of the Imipramine Hydrochloride release mechanism mucoadhesive, the formulations appeared to several kinetic model fitting. In all the six formulations the in-vitro release studies were done and those with highest percentage drug release that if formulation FIII and FVI were subjected to kinetic modelling. The data are represented in the graphs in figure 10 and figure 11 respectively.

The kinetic study reveals that the imipramine hydrochloride loaded buccal patch with highest percentage drug release that is formulation F VI follows zero order kinetics as the regression coefficient approaches unity, indicating the drug release is independent of drug concentration. The n value from the Korsmeyer Peppas model shows that the drug release pattern follows zero order kinetics. The formulation F III with second highest invitro drug release follows Korsmeyer Peppas model as the best fit model where the regression coefficient approaches unity and the n value from the Korsmeyer Peppas model show that the drug release model goes behind zero order kinetics. From drug permeation studies, in-vitro release studies and kinetic analysis of the in-vitro release data we can conclude that the formulation F VI with highest percentage drug release following zero order drug release kinetics can be considered as the optimized formulation.

Kinetic Analysis of Ex-vivo Permeation Data of Optimised Formulation

Ex-vivo permeation study data of the optimized formulation that is formulation F VI was subjected to kinetic data analysis. To characterize the permeation mechanism of Imipramine Hydrochloride from mucoadhesive buccal patches, the optimized formulation was subjected to various kinetic model fitting and is represented in figure 12.

Table 5: Model Fitting Profile of Optimized Formulation

Formulation Code	KINETIC MODELS								Best Fit Model
	Zero Order		First Order		Higuchi		Peppas		
	R	Ko	R	K1	R	Kh	R	N	
F VI	0.9796	6.6341	0.9702	0.0529	0.9173	24.803	0.9932	0.9627	Peppas

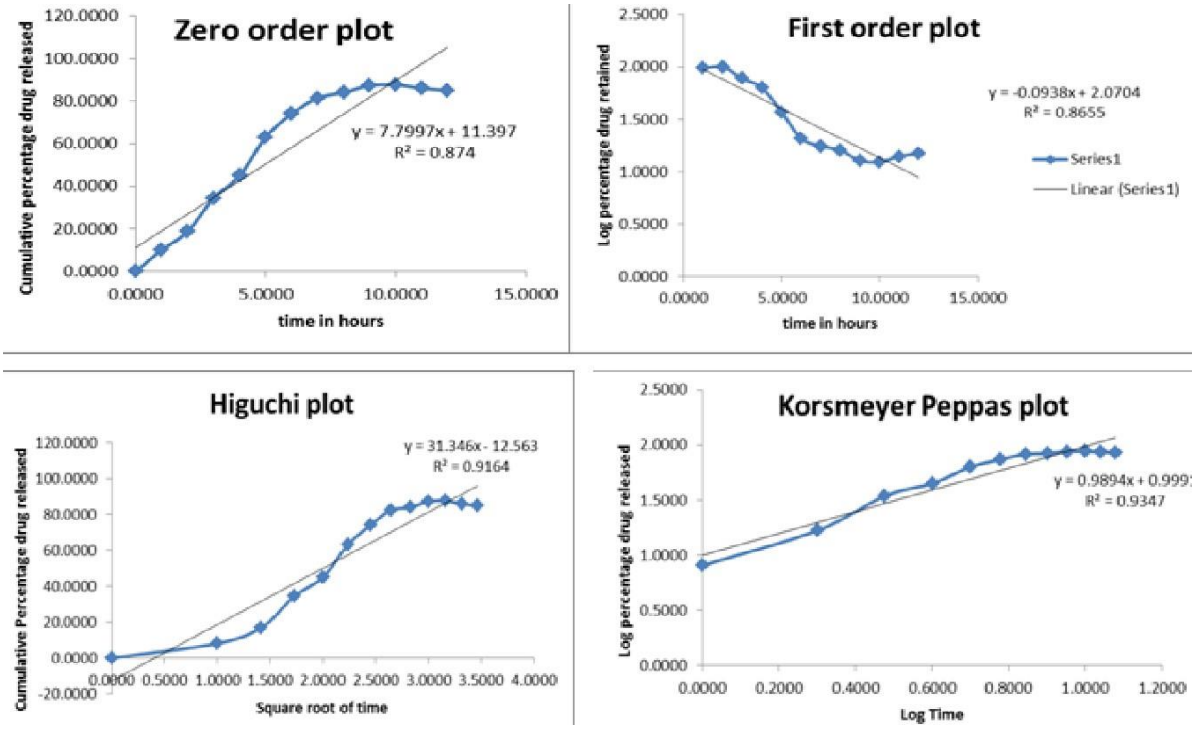

Figure 10: Kinetic plots for F III

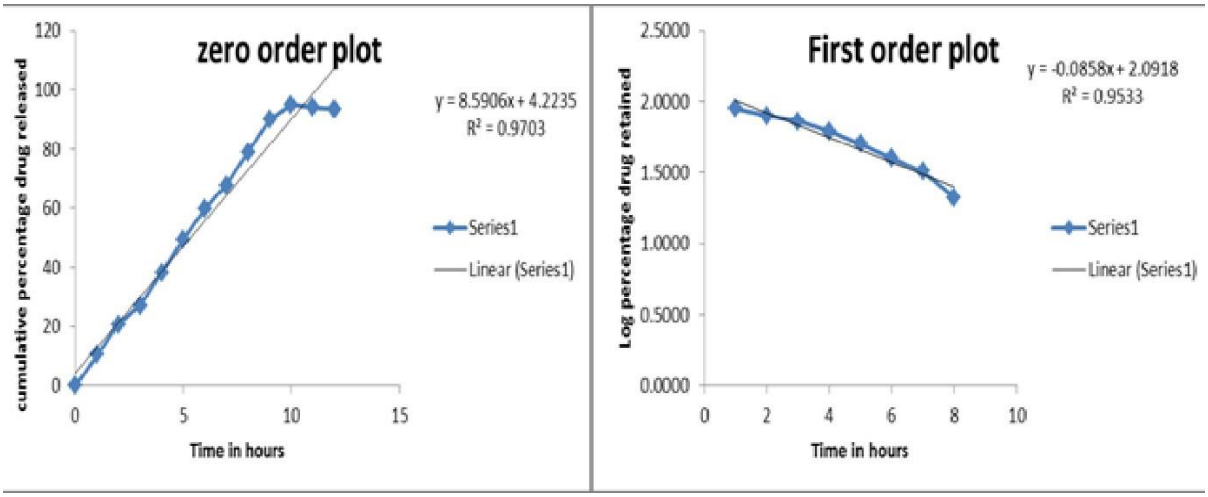

Figure 11: Kinetic plots for FVI

Model Fitting Profile of Optimized Formulation

The kinetic study of ex-vivo drug permeation data reveals that the optimized formulation of imipramine hydrochloride loaded buccal patch with highest percentage drug release that is formulation FVI follows Korsmeyer Peppas model as the best fit model as the regression coefficient approaches unity and it is represented in table 5. The n value from the Korsmeyer Peppas model shows that the drug permeation models track zero order kinetics.

Mass Balance Study

After performing the in-vitro release studies, all the six formulations were taken for mass balance study. Mass balance study is to find out the residual drug content that is left in the films after performing the in-vitro drug release studies. The residual drug content present in each formulation is represented in the figure13.

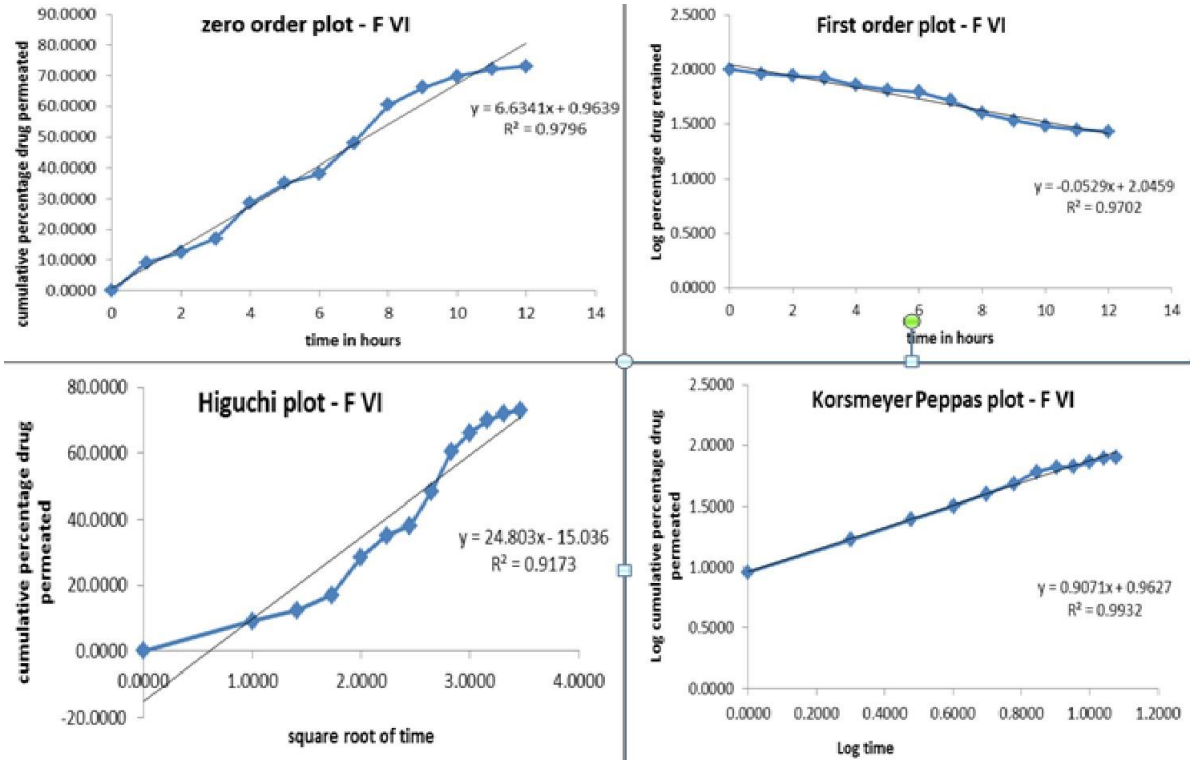

Figure 12: Kinetic plots for optimised formulation

The residual drug content will be less in the case of formulation showing highest percentage drug

release in the in-vitro release studies that is formulation FVI. The formulation FI with lowest percentage drug release shows the highest residual drug content.

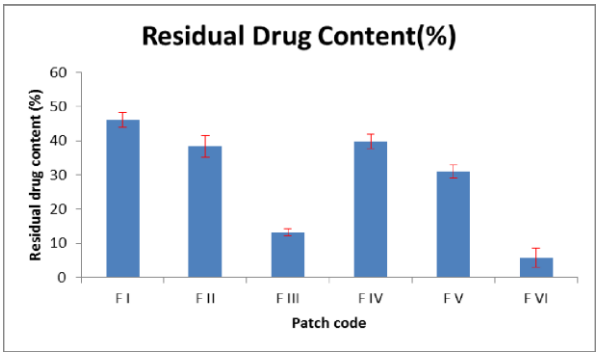

Figure 13: Residual drug content of 6 formulations

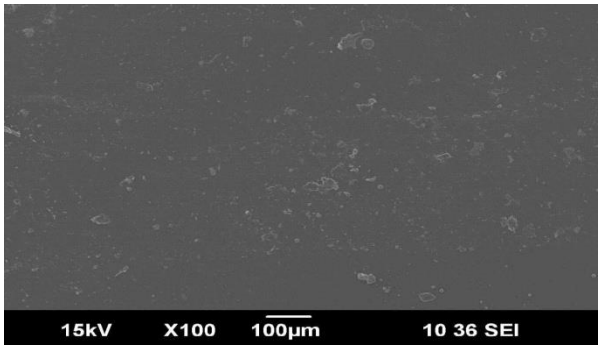

Figure 14: Scanning Electron Microscopy of optimized buccal patch

Scanning Electron Microscopy

Uniform dispersion of polymeric solution with the drug molecule is specified using the SEM photograph Figure 14. The patch (FVI) with the porous and loosely packed surface, is apt for the matrix system.

Comparison studies

Comparative Study of In-vitro Drug Release from the Optimised Gel with the Marketed Formulation of Imipramine Hydrochloride

The in-vitro release data of optimized formulation after 12 hrs was evaluated with a marketed formulation that is Imipramine hydrochloride tablet (Imiprin 25mg tablet). The cumulative percentage amount of drug released was chosen for their comparative studies. The release data of the optimized formulation and the marketed formulation are represented graphically in figure 15. The cumulative percentage amount of liberating drug was found to be 95.05% and 83.12% respectively for the optimized formulation that is FVI and for the marketed formulation at the end of 12 hours. The optimized formulation follows zero order release kinetics. From the information, it is clear that the imipramine hydrochloride mucoadhesive buccal patch showed the highest percentage drug release and the drug release was in a sustained manner for an episode of 12 hours. The drug is incorporated in the polymer matrix to form a mucoadhesive buccal patch and the drug release takes place from the

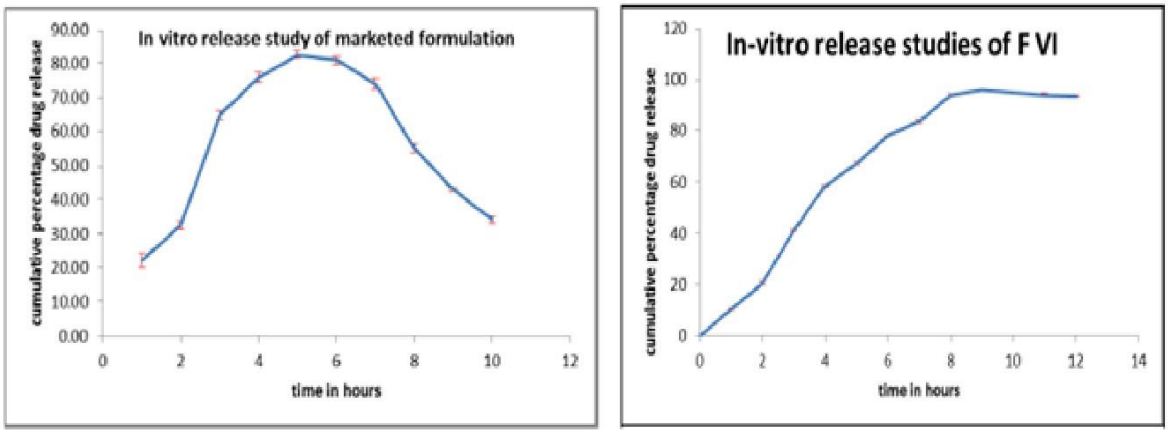

Figure 15: In-vitro release of Imipramine hydrochloride from optimized formulation and marketed formulation

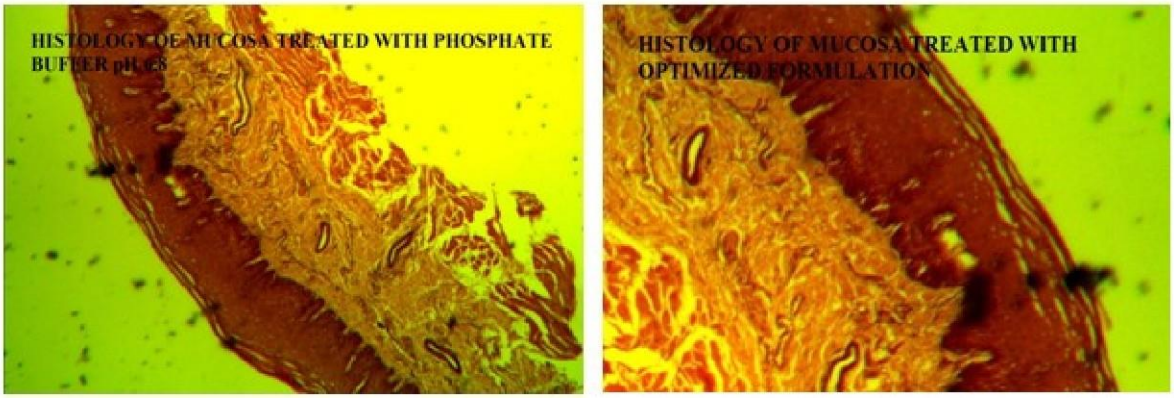

Figure 16: Photograph of histology of mucosa treated with phosphate buffer pH 6.8 and with optimized buccal patch

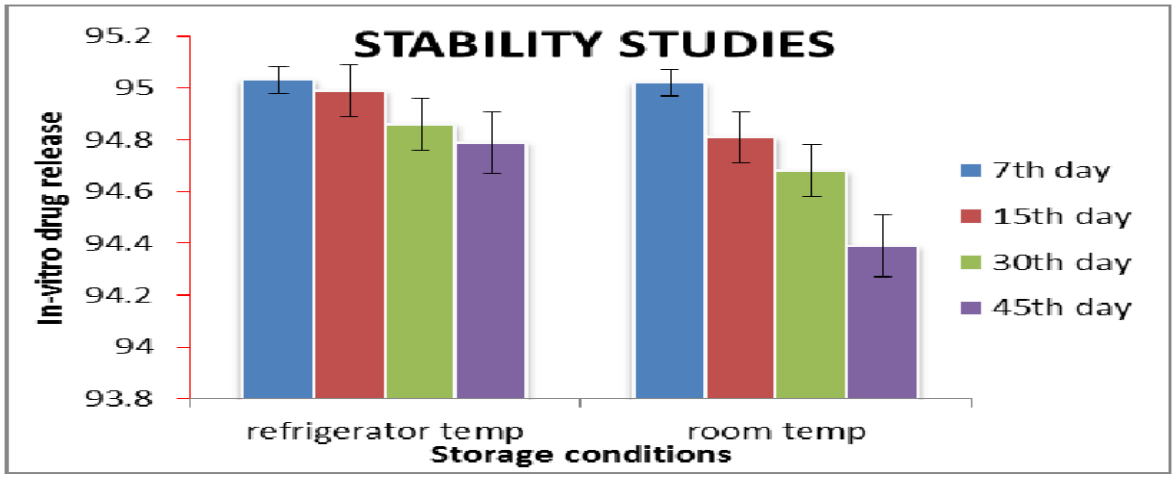

Figure 17: Stability study of optimized formulation

patch at 37°C in a sustained manner. The marketed formulation also showed a controlled pattern and the highest percentage drug release of about 83.12% was during the 5th hour and after that, the drug release was gradually decreasing. The optimized formulation that is FVI showed a sustained drug release pattern where the drug release was for a prolonged time that is for about 12 hours. Here, the highest release was during the 10th hour and then it gradually decreases.

Histological Studies

The histology of goat buccal mucosa treated using phosphate buffer pH 6.8 and those treated with the optimized formulation of the imipramine hydrochloride buccal patch is represented in figure 16 from the microscopic examination it is clear that optimized formulation gives no considerable outcome on the microscopic structure of the goat buccal mucosa. Cell necrosis or exclusion of the epithelium from the buccal mucosa was not observed

after permeation of optimized imipramine hydrochloride buccal patch. When the external part of submucosa was compared with phosphate buffer pH 6.8 treated mucosa, no alterations seen on the epithelial layer. So buccal patch formulations found to be safe. This optimized imipramine hydrochloride buccal patch gives a reduced clearance of the formulation at the spot of application and improved drug absorption. Anionic mucoadhesive polymer carbopol 940 helps in enhanced mucoadhesive potential. By considering all the results mentioned above, formulation F VI was found to be suitable mucoadhesive buccal patch formulation by means of excellent mucoadhesive potential at the same time enhanced permeation achievement. Thus, formulation F VI with 150mg Carbopol 940 and 50mg HPMC E 15 considered being the most excellent formulation for buccal liberation of imipramine hydrochloride.

Stability studies

The physical appearance doesn't be evidence for any changes in comparison with the freshly prepared formulation. The drug content and in-vitro drug release were evaluated on 7th, 15th, 30th, 45th day which was represented in figure 17 and it gives the idea that there is no considerable variation in these parameters during the storage for 45 days in both conditions. But the formulation seems to show better stability in low temperature in comparison to room temperature.

CONCLUSION

Buccal mucosal drug delivery is one of the fastest routes for the release of drugs to the systemic circulation. This study was aimed at developing an appropriate route for the delivery of an antidepressant drug (imipramine hydrochloride), thus overcoming the problems related with it like low oral bioavailability, severe gastrointestinal irritations, high dosing frequency etc. and thereby prolonging the extent of action and in that way decreasing the dosing frequency. From the preformulation studies, it was understandable that it satisfied the whole features for buccal delivery from the partition coefficient of the drug its clear that it is lipid soluble and therefore made sure that the drug can be effectively transported across the buccal mucosa for reaching the systemic circulation. By solvent casting technique, with varying the concentration of carbopol and HPMC, the mucoadhesive buccal patch was prepared, which were used as the factors for optimization and the responses used were in-vitro drug release and ex-vivo drug permeation. The mucoadhesive property, drug release and drug permeation depend upon the concentration of carbopol and HPMC when the concentration of carbopol increases the mucoadhesion property also increases which in turn increases the drug release and permeation. Total six formulations were prepared. All the six formulations were evaluations for its physical properties like thickness, weight uniformity, friability, content uniformity, mucoadhesion time and mucoadhesive strength etc. permeation studies revealed an increase in the permeation flux, and this is due to the presence of permeation enhancer and mucoadhesive polymers present. On the other hand, the in-vitro drug release studies show a sustained manner and track zero order kinetics. The liberation of the drug from the buccal patch does not depend upon the concentration of the drug present in the formulation. The marketed formulation shows a less release than that of the optimised formulation.

The penetration of the drug from the formulation through the buccal mucosa also follows zero order kinetics, which is not dependent on the drug concentration that gets released. The histological studies confirmed that the new formulation does not produce any damage to the mucosal surface, which indicates that the drug can effectively be delivered along the buccal mucosa into the blood. The non-ionic surfactants can increase the fluidity of the drug and increases its permeation across the buccal mucosa. The stability study by two different temperatures for 45 days shows that these were stable formulations even at the end of 45 days. From the present study, it is clear that the higher concentration of mucoadhesive polymer carbopol and the presence of tween 80 can increase the drug release and drug permeation across the buccal mucosa. Thus imipramine hydrochloride buccal patch can effectively deliver the drug into the systemic circulation

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