AAPS PharmSciTech, Vol. 11, No. 1, March 2010 (© 2010)
DOI: 10.1208/s12249-010-9381-4

Research Article

Buccal Drug Delivery of Pravastatin Sodium

Supriya S. Shidhaye,[1,2] Pritesh V. Thakkar,[1] Neha M. Dand,[1] and Vilasrao J. Kadam[1]

Received 19 January 2009; accepted 12 January 2010; published online 19 March 2010

Abstract. The purpose of this study was to develop and optimize formulations of mucoadhesive bilayered buccal tablets of pravastatin sodium using carrageenan gum as the base matrix. The tablets were prepared by direct compression method. Polyvinyl pyrrolidone (PVP) K 30, Pluronic® F 127, and magnesium oxide were used to improve tablet properties. Magnesium stearate, talc, and lactose were used to aid the compression of tablets. The tablets were found to have good appearance, uniform thickness, diameter, weight, pH, and drug content. A 2^3 full factorial design was employed to study the effect of independent variables viz. levels of carrageenan gum, Pluronic F 127 and PVP K30, which significantly influenced characteristics like *in vitro* mucoadhesive strength, *in vitro* drug release, swelling index, and *in vitro* residence time. The tablet was coated with an impermeable backing layer of ethyl cellulose to ensure unidirectional drug release. Different penetration enhancers were tried to improve the permeation of pravastatin sodium through buccal mucosa. Formulation containing 1% sodium lauryl sulfate showed good permeation of pravastatin sodium through mucosa. Histopathological studies revealed no buccal mucosal damage. It can be concluded that buccal route can be one of the alternatives available for the administration of pravastatin sodium.

KEY WORDS: bilayered buccal tablets; carrageenan; mucoadhesive; pravastatin sodium; 2^3 factorial design.

INTRODUCTION

Among the various routes of drug delivery, oral route is perhaps the most preferred by the patient. However, peroral administration of drugs has disadvantages such as hepatic first-pass metabolism and enzymatic degradation within the GI tract that prohibit oral administration of certain classes of drug. Drug buccal administration, on the other hand, is highly acceptable by patients, and the oral mucosa is relatively permeable with a rich blood supply. Furthermore, oral transmucosal drug delivery avoids first-pass effect and provides prompt removal of dosage form in case of need (1).

Buccal route is a promising route of administration for drugs, having high first-pass metabolism, low dose, and log*P* value in the range of 1.60–3.30 and small molecular size. This route, because of the longer contact time and greater flux offered by the drug delivery system, has also tried macromolecular drugs such as proteins, peptides, and steroids. The drugs with variable degree of lipophilicity and molecular size such as acyclovir (2) (log*P* −1.74, mol. wt. 225), atenolol (3) (log*P* −1.82, mol. wt. 266.30), chlorpheniramine (4) (log*P* 0.53, mol. wt. 390.90), diclofenac (5) (log*P* 1.13, mol. wt. 318.10), fentanyl (6) (log*P* 2.98, mol. wt. 528.50), lamotrigine (7) (log*P* 0.076, mol. wt. 256), lidocaine (8) (log*P* 1.62, mol. wt. 288.80), omeprazole (9) (log*P* 2.20, mol. wt. 345.40), propranolol (10) (log*P* 3.60, mol. wt. 295.80), etc. have been tried.

Pravastatin sodium, 3,5-dihydroxy-7- [6-hydroxy-2-methyl-8-(2-methylbutanoyloxy)-1,2,6,7,8,8a-hexahydronaphthalen-1-yl]-heptanoic acid, is a hydrophilic competitive inhibitor of 3-hydroxy-3-methylglutaryl coenzyme A (HMG-CoA), the rate-limiting enzyme of cholesterol synthesis. The drug lowers plasma cholesterol levels in hypercholesterolemia subjects. It is administered orally in doses of 10, 20, or 40 mg as a single dose daily. Approximately 34% of the drug is absorbed orally out of which the average systemic bioavailability of pravastatin sodium is 17% based on the plasma AUC and urinary excretion data (11). These figures indicate that approximately half of the absorbed drug is subjected to pre-systemic metabolism in the liver. The presence of food in the gastrointestinal tract reduces the bioavailability by about 35–40%. Also, drug is reported to be unstable in acidic pH (12).

This justifies a need to develop an effective formulation which allows the drug to directly enter the systemic circulation through internal jugular vein by avoiding first-pass metabolism, thereby increasing bioavailability of pravastatin sodium. Since the buccal route bypasses the hepatic first-pass effect, the dose of pravastatin sodium can be reduced. The physicochemical properties of pravastatin sodium, its suitable log*P* value (1.44), and its molecular weight 446.5 make it a suitable candidate for administration by the buccal route (12).

Buccal route is one such alternative. Also, so far, buccal route has not been explored for the administration of this drug. The drug is currently available in the market as tablet dosage form.

[1] Department of Pharmaceutics, Bharati Vidyapeeth's College of Pharmacy, C.B.D. Belapur, Navi Mumbai, 400 614, Maharashtra, India.
[2] To whom correspondence should be addressed. (e-mail: supriya.shidhaye@rediffmail.com)

The advantages such as excellent accessibility, low enzymatic activity, suitability for drugs or excipients that mildly and reversibly damage or irritate the mucosa, painless administration, easy drug withdrawal, facility to include permeation enhancer/enzyme inhibitor or pH modifier in the formulation and versatility in designing as multidirectional or unidirectional release systems for local or systemic actions, etc. opt for buccal adhesive drug delivery systems as promising option for continued research (13,14).

In the present study, mucoadhesive bilayered buccal tablet of pravastatin sodium for buccal administration was developed and optimized aiming at studying various formulation variables and its effect on release parameters and mucoadhesive strength. Also, attempts were made to improve buccal penetration of the drug. Bilayered design of the tablet coated from three sides, i.e., back and the circumference, was selected to obtain unidirectional release of the drug, greater surface area of contact, and administering the bitter drug without taste masking.

For development of mucoadhesive, bilayered buccal tablets of pravastatin sodium, carrageenan gum was used as mucoadhesive polymer (15). Because of the properties such as hydrophobicity, low water permeability, drug impermeability, and moderate flexibility, ethyl cellulose was used as a backing layer polymer to prevent drug loss.

The present investigation thus aims at establishing the suitability of buccal route for administration of pravastatin sodium and studying influence of matrix polymers like carrageenan gum, Pluronic F 127 and PVP K 30, on the mucoadhesive and drug release properties of the tablet.

MATERIALS AND METHODS

Materials

Pravastatin sodium was received as gift sample from Cipla Pharmaceuticals, Mumbai, India.

λ-carrageenan gum and Pluronic® F127 were received as gift samples from Degussa, Mumbai, India. Lactose (directly compressible grade DCL 11) was received as gift sample from (DMV International, the Netherlands). Polyvinyl pyrrolidone K30, sodium lauryl sulfate, ethyl cellulose (10 cps), magnesium oxide, magnesium stearate, and talc were purchased from S. D. Fine Chemicals, India.

Methods

Mucoadhesive bilayered buccal tablets were prepared by direct compression.

Core Tablet

Mucoadhesive formulation was prepared using 10 mg carrageenan gum, 8 mg Pluronic® F127, and 6 mg PVP K30, with lactose as the diluent, talc as glidant, along with 1 mg of magnesium stearate as lubricant and 2 mg magnesium oxide as drug stabilizer per tablet. Drug (10 mg/tablet) and the excipients for a batch size of 300 tablets were sieved through mesh #80 and homogenously blended for 10 min. This mixture was further lubricated with magnesium stearate by mixing for a further 2 min. The blend was tested for content uniformity, percent compressibility, and angle of repose. Angle of repose was determined by adjusting the height of the funnel 2 cm above the horizontal surface. The blend was allowed to flow from the funnel under the gravitational force until the apex of the pile just touched the apex of the funnel. A boundary was drawn along the circumference of the pile, and an average of six diameters was taken. The values of height and diameter were then substituted in the following equation to get the angle of repose.

$$\text{Angle of repose } (\theta) = \tan^{-1}(2h/d).$$

Subsequently, the blend was compressed into flat-faced tablets (100 mg, 9-mm diameter) using ten station minipress compression machine (Rimek) under constant maximum compression force that could be provided by the machine.

Backing Layer

Ethyl cellulose granules were prepared by wet granulation using isopropyl alcohol as the granulating solvent. The wet mass was passed through mesh #8 and dried at 40°C for 1 h. The granules were then passed through mesh #22 and retained on mesh #44. The core tablet was transferred to the die cavity fitted with 10-mm flat punch. Ethyl cellulose granules (100 mg) were added and subsequently compressed at constant maximum compression force. The tablets were coated from the sides and bottom with ethyl cellulose as backing membrane such that only the top surface remained uncoated.

Optimization of Formulation

A 2^3 randomized full factorial design was used in this study (16). Three factors were evaluated, each at two levels, and experimental trials were performed on all eight possible combinations (Table I). The amounts of Pluronic F 127 (X1), carrageenan gum (X2), and PVP K30 (X3) were selected as independent variables. The mucoadhesive strength and *in vitro* drug release were selected as dependent variables.

Regression polynomials for the individual dependant variables were calculated with the help of Design Expert 7.1 software and applied to approximate the response surface

Table I. Optimization of Formulation of Buccal Tablets

Batch code	Amount of Pluronic F 127 in mg (X1)	Amount of carrageenan gum in mg (X2)	Amount of PVP K-30 in mg (X3)
F-1	8	10	6
F-2	8	8	6
F-3	8	10	4
F-4	8	8	4
F-5	5	10	6
F-6	5	8	6
F-7	5	10	4
F-8	5	8	4

and contour plots. The general model as shown below was generated:

$$Y = B_0 + B_1X_1 + B_3X_2 + B_3X_3 + ... + B_{12}X_1X_2 + B_{13}X_1X_3 + B_{23}X_2X_3 + ... + B_{123}X_1X_2X_3 + \quad (1)$$

where Y is the measured response, X_i is the level of the ith factor, and B_i, B_{ij}, B_{ijk}.... represent coefficients computed from the responses of the formulations in the design.

Physicochemical Characterization of the Tablets

Weight variation (17) was determined on 20 tablets as per the requirement of tablets with average weight <250 mg (limit ± 5% of average weight). Hardness (18) of the tablets was measured on six tablets using Monsanto hardness tester (Dolphin). The dimensions (18) of six tablets were measured using Vernier caliper. Content uniformity (17) of tablets was done by extracting the drug in water and analyzing it spectrophotometrically at 239 nm after appropriate dilution. The mean and standard deviation were calculated.

Surface pH

The surface pH of the prepared tablets was determined to evaluate the possible irritation to buccal mucosa. Tablets were left to swell in 0.2 molar phosphate buffer, pH 6.8 (to mimic the condition in the buccal cavity), in 50-mL beakers, and pH was measured at time intervals of 15, 30, 60, 90, and 120 min by placing the electrode in contact with the microenvironment of the tablet on PHS-3D digital pH meter (Cyberlab, Sanjay biotechnology solutions Pvt. Ltd., India) (19,20).

Swelling Studies

After weighing the tablet (W_1), it was immersed in 0.2 molar phosphate buffer, pH 6.8, solution maintained at 37°C (19,20). The weight at the end of 120 min was reported (W_2). The swelling index was determined from the formula:

$$\% \text{ Swelling index} = (W_2 - W_1)/W_1 \times 100. \quad (2)$$

The experiment was carried out on three tablets.

In Vitro Drug Release Studies

The dissolution test was carried out using USP 24 dissolution testing apparatus II (VEEGO USP dissolution apparatus). The test was performed at a paddle speed of 50 rpm using 500 mL of 0.2 molar phosphate buffer, pH 6.8, as the dissolution medium at 37±0.50°C (19–21). The tablets were stuck on the paddle from the side of backing layer using cyanoacrylate adhesive to mimic unidirectional drug release. An aliquot of 5 mL of the sample solution was withdrawn at the interval of 15, 30, 45, 60, 90, and 120 min, and the absorbance was measured at 239 nm (Shimadzu 1601, Japan) after appropriate dilution with the help of standard curve of the drug (range 2–64 µg/mL, y=0.0483×; r^2=0.9996) in 0.20 molar phosphate buffer, pH 6.8. A test on placebo was performed to eliminate interference of the ingredients of the tablet. The test was performed on three tablets.

Drug Release from Backing Layer

For determination of drug release from the backing layer, Franz diffusion cell was used. A bilayered buccal tablet was placed between donor and receptor compartment. The complete unit was maintained at 37°C; donor compartment (3 mL) was filled with simulated saliva, pH 6.8 (sodium chloride 4.50 g, potassium chloride 0.30 g, sodium sulfate 0.30 g, ammonium acetate 0.40 g, urea 0.20 g, lactic acid 3 g, and distilled water up to 1,000 mL, adjusting pH of the solution to 6.8 by 1 M NaOH solution), and receptor compartment (21 mL) contained phosphate buffer, pH 7.4, with synchronous stirring. At predetermined interval, 2 mL sample was removed from donor compartment and analyzed at 239 nm by UV spectrophotometric analysis (19,20).

Mucoadhesion Strength

The mucoadhesion strength was checked using a modified balance method. The apparatus constitutes of a two pan balance which has been modified by replacing one pan of the apparatus with a Teflon assembly on which the tablet is stuck and which is in turn lowered on another Teflon assembly over which the buccal mucosa is tied. Porcine buccal mucosa was used as the model membrane. The mucosa was stored in phosphate buffer, pH 7.4, at room temperature before use. The mucosal membrane was excised by removing the underlying connective and adipose tissue. It was then equilibrated in 0.2 molar phosphate buffer, pH 6.8, at 37±1°C for 30 min. The tablet was stuck to the Teflon arm using cyanoacrylate adhesive and lowered onto the mucosa under a constant weight of 5 g for a total contact period of 5 min. Mucoadhesion strength was assessed in terms of weight (g) required to detach the tablet from the membrane (19,20,22).

In Vitro Residence Time

The tablet was applied on the porcine buccal mucosa which was fixed on the glass slide with cyanoacrylate glue. The slide was tied to the disintegration apparatus and suspended in the beaker filled with 800 mL simulated saliva, pH 6.8. The slide was allowed to reciprocate in the medium until the tablet got detached or eroded from the mucosa (19,23). The test was performed in triplicate.

Assay

This was carried out by subjecting each tablet to validated, stability-indicating high-performance liquid chromatography (HPLC) method of assay. Tablet was powdered so as to effect the complete extraction of drug in 25 mL of methanol. Methanol is used as the solvent for extraction as the drug is freely soluble in methanol (the solubility of other excipients in methanol is very less). Further dilutions were carried out and analyzed by HiQ Sil C_{18} 4.6×250 mm (5 µm packing) column with acetonitrile and water (1:1) as mobile phase at the flow rate of 1 mL/min and absorbance at 239 nm.

The unknown concentration was determined from the calibration curve (17). This assay helps in determining the exact amount of drug loaded in the tablet. As the analysis is done by a validated HPLC method in lieu of UV–Vis spectrophotometry, the results obtained would be more accurate. The HPLC would also help in identifying any instability or interaction between the drug and the various excipients used.

For HPLC analysis, the method was validated for accuracy, precision, selectivity, and linearity.

Permeation Studies

Diffusion studies were carried out to evaluate the permeability of drug across the porcine buccal mucosal membrane (24) using glass surface Franz diffusion cell. Porcine buccal mucosa was obtained from a local slaughterhouse (R.K. Pork, Mumbai, India) and used within 2 h of slaughter. The tissue was stored in 0.2 molar phosphate buffer (PBS), pH 7.4, solution upon collection. The epithelium was separated from underlying connective tissues with surgical scissors and clamped in between donor and receiver chambers of the diffusion cells for permeation studies. Receptor compartment contained 21 mL of pH 7.4 phosphate buffer, while donor compartment was filled with 3 mL simulated saliva of pH 6.8. The tablet was placed on the mucosal surface in donor compartment, and 2 mL aliquots were removed at suitable intervals from the receptor compartment while the solution was being stirred continuously using magnetic stirrer, replacing it with fresh 2 mL medium each time (20). The experiment was carried out at 37±1°C. The amount of drug permeated was assayed using validated, stability-indicating HPLC method of analysis with the help of standard curve of drug (range 1–16 μg/mL, $y=0.0483\times$; $r^2=0.9990$ in mobile phase). The apparatus used for HPLC analysis was Jasco 200 plus system equipped with a UV detector. Computerized data acquisition and treatment were performed with the Borwin Chromatography Software. Chromatographic conditions applied were flow rate 1.0 mL/min and mobile phase acetonitrile/double distilled water (1:1) separation carried out at 25°C temperature on a 250×4.0 mm using reverse-phase column packed with 5 μm C18 silica particles (Hi-Qsil C18). Absorbance was measured at 239 nm. The graph of percent drug permeated *vs.* time was plotted, and flux, permeability coefficient, and enhancement ratio were determined (25). The experiments were performed in triplicate and average values reported.

Histopathological Evaluation of Buccal Mucosa

Histopathological evaluation of tissue incubated in phosphate-buffered saline solution, pH 6.8, was compared with that treated with buccal tablet for 2 h. The tissue was fixed with 10% formalin, routinely processed, and embedded in paraffin. Paraffin sections were cut on glass slides and stained with hematoxylin and eosin (26). A pathologist blinded to the study to detect any damage to tissue at Haffkins Research Centre, Mumbai, India examined sections on light microscope.

RESULTS AND DISCUSSIONS

During preparation of the tablet, the blend prepared for direct compression was found to be satisfactory with an angle of repose of 18.50±0.82° and compressibility of 18.16±0.55%.

Physicochemical Properties of the Tablets

Physicochemical characteristics of the tablets are shown in Table II. The tablets of all formulation had good appearance: 10.53±0.06-mm diameter, 1.98±0.05-mm thickness, 4.0±0.10-kg/cm^2 hardness, and 199.96±0.74-mg weight. The pH of the formulation was found to be 7.83±0.05 due to presence of magnesium oxide incorporated in the formulation for stabilizing pravastatin sodium. It is reported that the drug degrades in acidic condition.

Optimization of Formulation

As seen from Table III, the model F value of 6,3660,000 implied that the model was significant. There was only a 0.01% chance that a "model F value" this large could occur due to noise. Values of "Prob > F" <0.05 indicated that model terms are significant. In this case, A, B, C, AB, AC, and BC are significant model terms.

The final models for mucoadhesive strength was as follows:

$$\text{Mucoadhesive strength} = [8.75 - 1.25 \times A + 4 \times B - 0.75 \times C - 0.50 \times A \times B - 0.25 \times A \times C + 0.10 \times B \times C \,] \quad (3)$$

where A is the amount of Pluronic F127, B the amount of carrageenan, and C the amount of PVP K-30 ($R^2=0.9998$).

Table II. Physicochemical Characteristics of Formulation from F-1 to F-8

Formulation	Mucoadhesive strength (g)	Time for *in vitro* drug release (for 50% drug release)	Swelling (%)	*In vitro* residence time (min)	Assay (%)
F-1	10	70	85.50	104	99.62
F-2	3	60	112.50	30	98.25
F-3	12	73	83.50	138	101.56
F-4	5	61	106	55	99.51
F-5	14	80	52.50	165	98.67
F-6	5	72	78.50	59	100.05
F-7	15	82	47.50	180	99.37
F-8	6	73	75	68	101.46

Table III. Response 1—Mucoadhesive Strength: Analysis of Variance (ANOVA) for Selected Factorial Model

Source	Sum of squares	*df*	Mean square	*F* value	*p* value Prob > *F*	
Model	147.50	6	24.58	63660000	<0.0001	Significant
A—amount of Pluronic	12.50	1	12.50	63660000	<0.0001	
B—amount of carrageenan	128	1	128	63660000	<0.0001	
C—amount of PVP K 30	4.50	1	4.50	63660000	<0.0001	
AB	2	1	2	63660000	<0.0001	
AC	0.50	1	0.50	63660000	<0.0001	
BC	0	1	0			
Residual	0	1	0			
Cor total	147.50	7				

As seen from Fig. 1, the surface response plot revealed that a corresponding increase in the mucoadhesive strength of tablets was observed with increase in concentration of carrageenan gum. This may be due to contact of the sulfonic acid groups of the potassium, sodium, calcium, magnesium, and ammonium sulfate esters of galactose and 3,6-anhydro-galactose copolymers with amine groups of glycoprotein found in mucous, thus causing mucoadhesion.

It was also revealed that with an increase in the concentration of Pluronic F 127, the mucoadhesion strength decreased. Pluronic F 127, a hydrophilic polymer, has been incorporated in the formulation to improve wetting of tablet by water channeling effect and induce swelling required for mucoadhesion as well as drug release. The decrease in mucoadhesion strength with increase in Pluronic F 127 can be attributed to rapid swelling of tablet, leading to faster erosion and detachment. The results of *in vitro* residence time reported in Table II demonstrated similar effect.

The results also indicated that the effect of concentration of carrageenan gum on mucoadhesion strength was more significant than that of Pluronic F 127 and PVP K30. Hence, for plotting response plots, concentration of PVP K 30 was fixed at constant value.

As seen from Table IV, the model *F* value of 575.67 implies that the model is significant. There is only a 3.19% chance that a "model *F* value" this large could occur due to noise. Values of "Prob > *F*" <0.0500 indicate that model terms are significant. In this case, A and B are significant model terms. Values >0.10 indicate that the model terms are not significant.

The final model of *in vitro* drug release for 50% drug release was as follows:

$$\text{Time for 50 \% release} = [71.375 - 5.375 \times A + 4.875 \times B - 0.875 \times C + 0.625 \times A \times B - 0.125 \times A \times C - 0.375 \times B \times C] \quad (4)$$

(R^2=0.9997). As seen from Fig. 2 (response surface plot for *in vitro* release), the time for 50% drug release appeared to decrease with an increasing amount of the hydrophilic polymer Pluronic F 127, keeping the value of PVP K 30 constant. The increase in the drug release could be explained by the ability of the hydrophilic polymer to absorb water, thereby promoting the dissolution, and hence the release, of the highly water-soluble drug pravastatin sodium. Moreover,

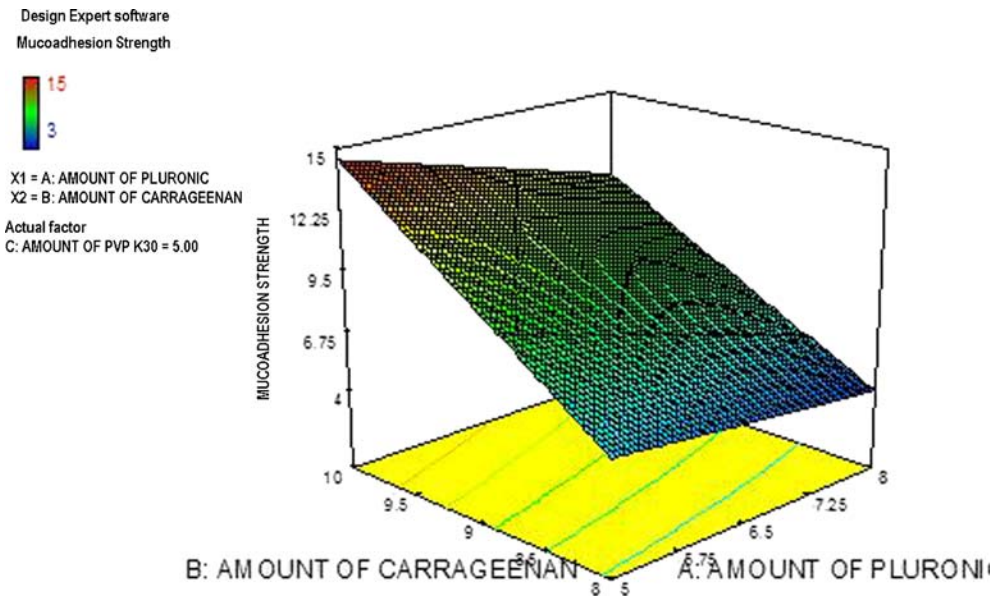

Fig. 1. Surface response plot for mucoadhesive strength

Table IV. Response 2—*In Vitro* Drug Release (Time for 50% Release): Analysis of Variance (ANOVA) for Selected Factorial Model

Source	Sum of squares	*df*	Mean square	*F* value	*p* value Prob > *F*	
Model	431.75	6	71.96	575.67	0.0319	Significant
A—amount of Pluronic	231.13	1	231.13	1849	0.0148	
B—amount of carrageenan	190.13	1	190.13	1521	0.0163	
C—amount of PVP K 30	6.13	1	6.13	49	0.0903	
AB	3.13	1	3.13	25	0.1257	
AC	0.13	1	0.13	1	0.5000	
BC	1.13	1	1.13	9	0.2048	
Residual	0.13	1	0.13			
Cor Total	431.88	7				

the hydrophilic polymers Pluronic F 127 and PVPK30 would dissolve, creating more pores and channels (27,28) for the drug to diffuse out of the tablet, but as concentration of carrageenan increased, drug release was decreased; this could be due to the extensive swelling of the carrageenan gum which created a thick gel barrier, making drug diffusion more difficult. A similar effect of carrageenan gum of retarding the release of drug in buccal formulation has been reported by Ruiz *et al.* (29). The dissolution profiles for the different formulations were as shown in Fig. 3 with the complete release observed in 120 min. Formulation F-2 showed the fastest *in vitro* release among all formulations due to less amount of carrageenan and highest amount of Pluronic F 127, indicating the role of Pluronic F 127 as a release-enhancing agent.

It was concluded that to get desired tablets of having mucoadhesive strength in the range 10–12 g and *in vitro* release time for 50% release in the range 70–75 min, carrageenan amount should be in the range of 9–10% (*w*/*w*) and Pluronic F 127 amount should be in the range of 5.75–8% (*w*/*w*), maintaining level of PVP K30 at 4% (Fig. 4, contour plot). Therefore, formulation containing 10% (*w*/w) of carrageenan, 8% (*w*/*w*) Pluronic F 127, and 4% (*w*/*v*) of PVP K30 (batch no. F-3) was selected as optimized formulation.

In Vitro Residence Time

The *in vitro* residence time with porcine buccal mucosa in simulated saliva (pH 6.8) varied from 30 to 180 min (Table II). It was observed that the effect of concentration of carrageenan on the *in vitro* residence time was significant, with tablets containing low proportion of carrageenan eroding rapidly and giving short residence time (batch no. F-2, F-4, F-6, F-8). Formulations F-1 and F-5 containing the same levels of carrageenan and PVP K30 but different levels of Pluronic F127 demonstrated that decrease in the amount of Pluronic F 127 led to an increase in the *in vitro* residence time. Thus, Pluronic F127 had a negative effect on *in vitro* residence time. This may be due to rapid wetting of the polymer leading to early swelling and detachment of the formulation. Also, PVP K30 was found to have a negative effect on the *in vitro* residence time, with

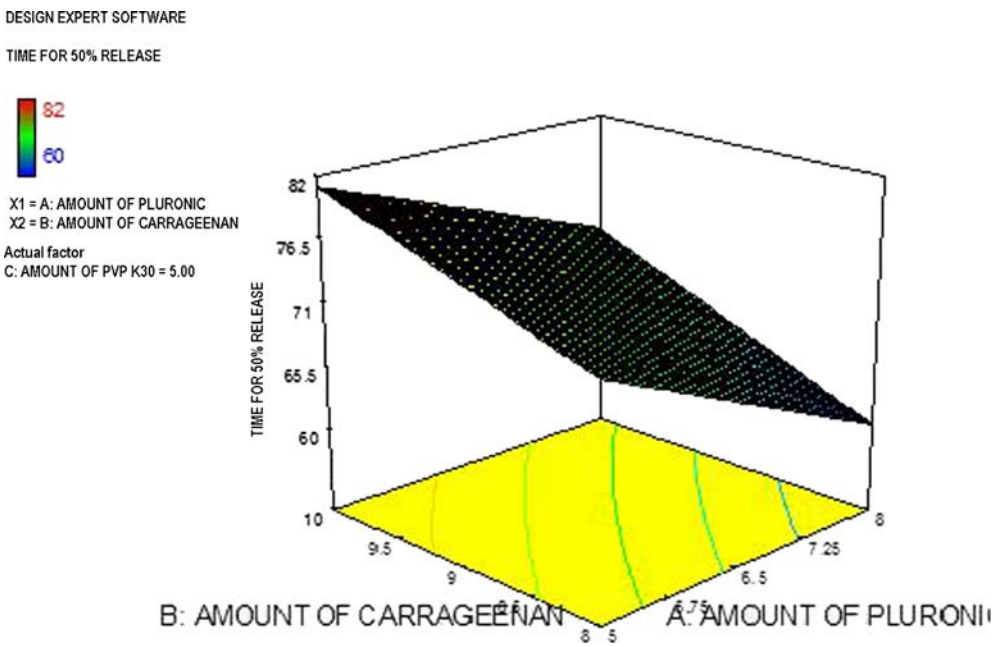

Fig. 2. Surface response plot for *in vitro* drug release for 50% drug release

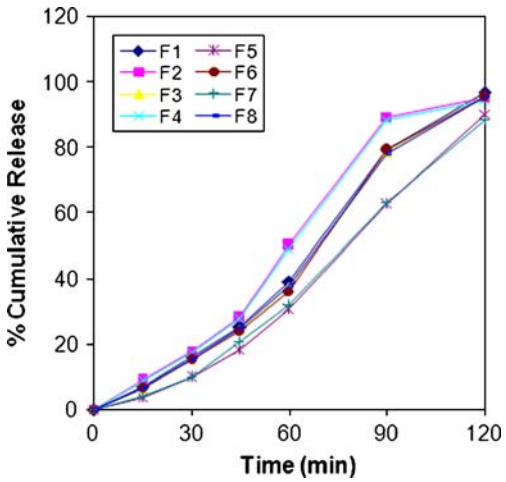

Fig. 3. Dissolution profiles for different formulations

formulations containing highest levels of PVP K30 giving shorter *in vitro* residence time. A similar effect has been demonstrated in the buccal patch of sumatriptan succinate by Shidhaye *et al.* (28).

Swelling of Formulations

Figure 5 depicts the degree of swelling of formulations F1 to F8 in simulated saliva solution of pH 6.8. Swelling of tablets was started within 5 min due to the presence of carrageenan gum, Pluronic F 127, and PVP K-30. Maximum increase in swelling was observed at 30 min. As the amount of carrageenan gum increased in the formulation, erosion of the tablets slowed down and extent of swelling of the tablet also increased.

Drug Release from Backing Layer

To evaluate the performance of backing membrane in avoiding release of pravastatin sodium, a study was conducted using Franz diffusion cell. Results of the study showed that no drug was released in 120 min in the donor compartment of diffusion cell. This indicated that ethyl cellulose membrane was impermeable to pravastatin sodium and the swelling of mucoadhesive layer did not change integrity of backing layer. Hence, tablet was found to be efficient for unidirectional release of pravastatin sodium through buccal mucosa.

The formulation having the best mucoadhesive strength, *in vitro* residence time more than 120 min, and desired drug release was subjected to permeation studies through the buccal mucosa to find out the extent of drug permeability in terms of permeation coefficient and flux.

Permeation Studies

Pravastatin, being hydrophilic with log*P* value of 1.44 (11), exhibits low permeability through buccal mucosa, and there is a need to enhance its buccal permeation with the help of penetration enhancer (30) that causes perturbation and dissolution of paracellular fluid, enhancing its paracellular transport. Based on this fact, different penetration enhancers (sodium lauryl sulfate, malic acid, sodium salicylate, and bile salts) were tried to improve buccal penetration of pravastatin sodium through buccal mucosa.

Figure 6 gives comparison of permeation of pravastatin sodium through porcine buccal mucosa for formulations containing different penetration enhancer. The permeability coefficient was calculated from the graph. These results are listed in Table V.

Results of the trials with malic acid, sodium salicylate, and bile salts showed not much improvement in the permeation of pravastatin as compared to trials with sodium lauryl

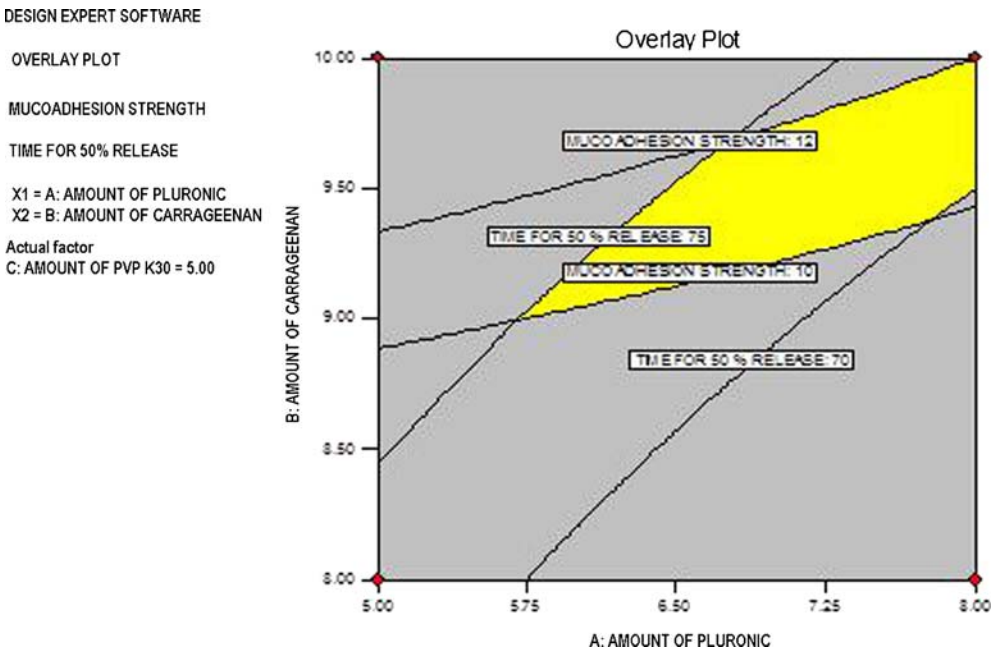

Fig. 4. Overlay plot

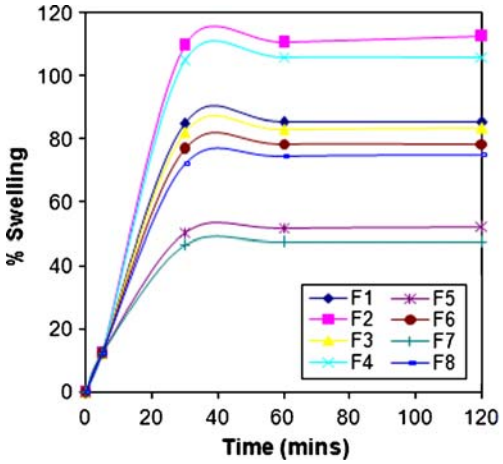

Fig. 5. Degree of swelling of different formulations

Table V. Permeation of Pravastatin Sodium Through Porcine Buccal Mucosa in the Presence of Different Penetration Enhancers

Formulation containing	Permeability coefficient	Flux (mcg/cm^2 s)
No penetration enhancer	0.005	0.019
Malic acid 1%	0.006	0.020
Malic acid 2%	0.006	0.021
Sodium salicylate 1%	0.066	0.022
Sodium salicylate 5%	0.010	0.034
Bile salt 1%	0.006	0.019
Bile salt 5%	0.014	0.046
Sodium lauryl sulfate 0.5%	0.011	0.036
Sodium lauryl sulfate 1%	0.021	0.073
Sodium lauryl sulfate 1.5%	0.022	0.074

sulfate. Sodium lauryl sulfate increased the permeability of drug significantly, with level 1% showing the best results.

When penetration enhancer is added, it is very unlikely that low concentrations of penetration enhancers would influence the physicochemical properties of tablet such as mucoadhesive strength, *in vitro* residence time, and drug release. The formulation optimized for the amount of penetration enhancer was still subjected to physiochemical characterization to confirm that the penetration enhancer did not adversely affect the other physicochemical characteristics.

Histopathological Evaluation of Buccal Mucosa

The microscopic observations indicated that the final formulation containing 1% sodium lauryl sulfate had no significant effect on the microscopic structure of mucosa. As shown in Fig. 7, no cell necrosis was observed. Cellular membrane was intact and no damage was observed to the treated porcine buccal mucosa. Thus, formulation containing 1% sodium lauryl sulfate appeared to be safe with respect to buccal administration.

Conclusion

It may be concluded that buccal route is one of the alternatives available for administration of pravastatin sodium. However, use of penetration enhancer is necessary to achieve permeation of drug through buccal mucosa. The results showed that mucoadhesive bilayered buccal tablets containing 10% (*w*/*w*) carrageenan, 8% Pluronic F 127, 4% PVP K-30, and 1% sodium lauryl sulfate produced buccal tablets having good mucoadhesive strength, 96% drug release over 2 h, and 23% permeation of the drug through buccal mucosa without causing any tissue damage.

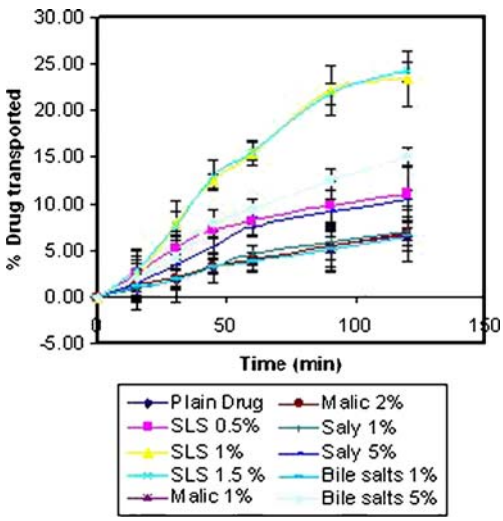

Fig. 6. Comparison of permeation of pravastatin sodium through porcine mucosa in presence of different penetration enhancers

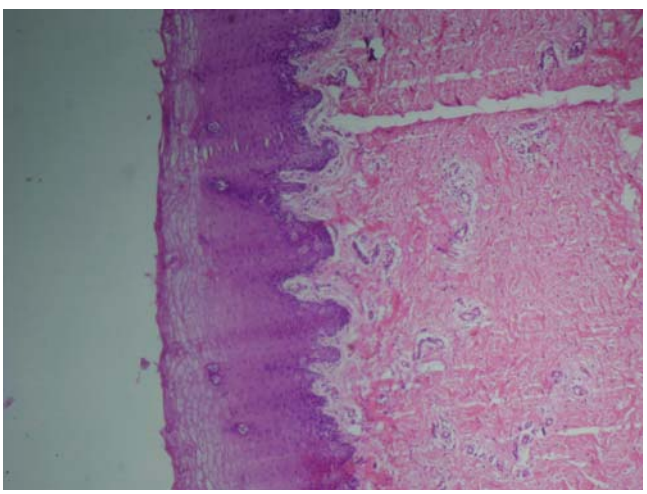

Fig. 7. Histopathological evaluation of sections of porcine buccal mucosa treated with tablet containing 1% SLS

REFERENCES

1. Smart JD. Drug delivery using buccal adhesive systems. Adv Drug Deliv Rev. 1993;11:253–70.
2. Rossi S, Sandri G, Ferrari F, Bonferoni MC, Caramella C. Buccal delivery of acyclovir from film based on chitosan and polyacrylic acid. Pharm Dev Tech. 2003;8(2):199–208.
3. Singh B, Chakkal SK, Ahuja N. Formulation and optimization of controlled release mucoadhesive tablets of atenolol using surface response methodology. AAPS PharmSciTech. 2006;7(1) Article 3.
4. Alur HS, Pather SI, Mitra AK, Johnston TP. Transmucosal sustained delivery of chlorpheniramine maleate in rabbits using a novel, natural mucoadhesive gum as an excipient in buccal tablets. Int J Phar. 1999;188:1–10.
5. El-Samaligy MS, Yahia SA, Basalious EB. Formulation and evaluaton of diclofenac sodium buccoadhesive discs. Int J Pharm. 2004;286:27–39.
6. Gordon D, Schroeder M. Oral transmucosal fentanyl citrate-OTFC (ACTIQ ®). www.eperc.mcw.edu.
7. Mashru S, Sutariya V, Sankalia M, Sankaliya J. Transbuccal delivery of lamotrigine across porcine buccal mucosa: *in vitro* determination of routes of buccal transport. J Pharm Pharmceut Sci. 2005;8(1):54–62.
8. Abu-Huwaij R, Assaf S, Salem M, Sallam A. Potential mucoadhesive dosage form of lidocaine hydrochloride: II. *In vitro* and *in vivo* evaluation. Drug Dev Ind Pharm. 2007;33:437–48.
9. Choi H, Kim C. Development of omeprazole buccal adhesive tablets with stability enhancement in human saliva. J Control Rel. 2000;68:397–04.
10. Munasur AP, Pillay V, Chetty DJ, Govender T. Statistical optimization of the mucoadhesivity and characterization of multipolymeric propranolol matrices for buccal therapy. Int J Pharm. 2006;323:43–51.
11. Dollery CT, Boobis AR, Burley DM. Therapeutic drugs. Edinburgh: Churchill Livingstone; 1999. p. P179–83.
12. Boyong LI, Nangia A, Dely A, Wong D. Stable pharmaceutical compositions containing pravastatin. WO Patent 03/000239, 2003.
13. Shojaei AH. Buccal mucosa as a route for systemic drug delivery: a review. J Pharm Sci. 1998;1:15–30.
14. JAJ Hoogstraate, Philip WW. Drug delivery via the buccal mucosa. PSTT. 1998;1(7):309–16.
15. Piyakulawat P, Praphairaksit N, Chantarasiri N, Muangsin N. Preparation and evaluation of chitosan/carrageenan beads for controlled release of sodium diclofenac. AAPS PharmSciTech. 2007;8(4):Article 97.
16. Sanford B, Charles B. Pharmaceutical statistics. New York: Marcel Dekker; 2005.
17. Indian Pharmacopoeia. Controller of Publication, Delhi, 1996.
18. Lachman L, Lieberman HA, Kanig JL. The theory and practice of industrial pharmacy. India: Verghese Publishing House; 1991.
19. Bortunc B, Ozer L, Uyanik N. Development and *in vitro* evaluation of a buccoadhesive pindolol tablet formulation. Drug Dev Ind Pharm. 1998;24(3):281–8.
20. Parvez N, Ahuja A, Khar RK. Development and evaluation of mucoadhesive buccal tablets of lignocaine hydrochloride. Indian J Pharm Sci. 2002;64(6):563–7.
21. Choi HG, Jung JH, Yong CS, Rhee CD, Lee MK, Han JH, *et al.* Formulation and *in vivo* evaluation of omeprazole buccal adhesive tablet. J Control Rel. 2000;68:405–12.
22. Jain AC, Aungst BJ, Adeyeye MC. Development and *in vivo* evaluation of buccal teblets prepared using danazol-sulfobutylether 7 beta-cyclodextrin (SBE 7) complexes. J Pharm Sci. 2002;91: 1659–68.
23. Patel VM, Prajapati BG, Patel JK, Patel MM. Physicochemical characterization and evaluation of buccal adhesive patches containing propranolol hydrochloride. Current Drug Del. 2006;3:325–31.
24. Nafee NA, Ismail FA. Mucoadhesive drug delivery system, formulation and *in-vitro* evaluation of mucoadhesive tablets containing water soluble drugs. Drug Dev Ind Pharm. 2004;30:995–04.
25. Lalla JK, Gurnaney RA, Narayan S. Permeation of Diclofenac through buccal mucosa. Indian J Pharm Sci. 2002;64(4):373–7.
26. Majithiya RJ, Ghosh PK, Umrethia ML, Murthy RSR. Thermoreversible mucoadhesive gel for nasal delivery of sumatriptan. AAPS PharmSciTech. 2006;7(3):Article 6.
27. Bodmeier R, Paeratakul O. Evaluation of drug-containing polymer films prepared from aqueous latexes. Pharm Res. 1989;6:725–30.
28. Shidhaye SS, Saindane NS, Sutar S, Kadam VJ. Mucoadhesive bilayered patches for administration of sumatriptan succinate. AAPS PharmSciTech. 2008;9(3):909–15.
29. Ruiz G, Ghaly E. Mucoadhesive delivery systems using carrageenan and Eudragit RLPO. Vitae. 2006;13(1):31–9.
30. Senel S, Hıncal AA. Drug permeation enhancement via buccal route: possibilities and limitations. J Control Rel. 2001;72:133–44.