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Buccal Bioadhesive Delivery System of 5-Fluorouracil: Optimization and Characterization

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The objective of this work was to apply the response surface approach in the development of buccal bioadhesive tablets of 5-fluorouracil (5-FU). Experiments were performed according to a 3^2 factorial design to evaluate the effects of two polymers, Gantrez MS-955 (X_1) and hydroxypropylmethyl cellulose (HPMC) K15M (X_2) on the bioadhesive force, percentage drug release in 8 h ($Rel_{8\ h}$), time taken for 50% drug release ($t_{50\%}$), and diffusion coefficient (n). The effect of the two independent variables on the response variables was studied by response surface plots and contour plots generated by the Design Expert® software. The compatibility between 5-FU and the tablet excipients was confirmed by differential scanning calorimetry (DSC) and Fourier transform infrared (FTIR) studies. Both the polymers were found to have synergistic effect on bioadhesion but the effect of Gantrez was more pronounced. A nonlinear twisted relationship was obtained for $Rel_{8\ h}$ at the intermediate and high levels of the polymers, which indicated an interaction between them at the corresponding factor levels. Kinetic treatment to the dissolution profiles revealed that the drug release ranged from Fickian to anomalous transport, which was mainly dependent on both the independent variables. The desirability function was used to optimize the response variables, and the observed responses were in agreement with the experimental values.

Keywords 5-fluorouracil; Gantrez; HPMC; bioadhesive tablets; factorial design; response surface methodology; contour plots

INTRODUCTION

Buccal mucosa is an attractive route for systemic delivery of drugs as it is relatively permeable with a rich blood supply (Hoogstraate et al., 1996; Voorspoels, Remon, Eechaute, & De Sy, 1996). Moreover, it has high robustness and accessibility. A drug can be easily applied and localized at the application site, and can also be removed from there if necessary (Tsutsumi, Obata, Nagai, Loftsson, & Takayama, 2002). The buccal mucosa has been investigated for local and systemic delivery of therapeutic peptides and other drugs that are subjected to first-pass metabolism or are unstable within the rest of the gastrointestinal tract (Ahuja, Khar, & Ali, 1997; Chidambaram & Srivastava, 1995; Smart, 1993). Buccal delivery for the transmucosal absorption of drugs into the systemic circulation offers a number of advantages over oral delivery, especially for those drugs that have poor oral bioavailability and/or those drugs that suffer from extensive first-pass metabolism in the liver. Conceivably, buccal delivery systems provide ease of administration and thereby increase patient compliance. Various types of buccal delivery systems have been developed, including sprays, solutions, erodable or nonerodable multilayer adhesive films, adhesive tablets, gels, disks, and lollipops (Alur, Beal, Pather, Mitra, & Johnston, 1999; Benes et al., 1997; Desai & Kumar, 2004; Hoogstraate & Wertz, 1998; Jay, Fountain, Cui, & Mumper, 2002; Li, Bhatt, & Johnston, 1997; Nafee, Ismail, Boraie, & Mortada, 2003; Patel, Prajapati, & Patel, 2007; Schechter, Weisman, Rosenblum, Bernstein, & Conard, 1995; Schwagmeier, Alincic, & Striebel, 1998; Shin, Bum, & Choi, 2000; Voorspoels et al., 1996).

Oropharyngeal cancer develops in the part of throat just behind mouth, called the oropharynx (http://www.cancer.gov). 5-Fluorouracil (5-FU) has been used for the treatment of oropharyngeal cancer (Dollery, 1999). The oral bioavailability of 5-FU is fairly poor because of its high first-pass effect and poor membrane permeability (Diasio & Harris, 1989). Hence, orally administered 5-FU shows marginal efficacy. An alternative route of administration that avoids injection but leads to a better efficacy (i.e., buccal route) would be advantageous for the patients.

In the development of extended-release dosage form with appropriate dissolution rate, an important issue is to design an optimized pharmaceutical formulation in a short time period with minimum trials. For this purpose, computer optimization techniques, based on response surface methodology (RSM) embodying the use of appropriate experimental designs and

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utilizing polynomial equation has been widely used (Bouckaert, Massart, Massart, & Rremon, 1996; Huang et al., 2004; Huang, Tsai, Yang, Chang & Wu, 2004; Singh, Dodge, Durrani, & Khan, 1995). Factorial designs, where all the factors are studied in all possible combinations, are considered to be most efficient in estimating the influence of individual variables and their interactions using minimum experiments (Singh & Ahuja, 2002). The application of factorial design in pharmaceutical formulation development has played a key role in understanding the relationship between the independent variables and the responses to them (Vandervoort & Ludwig, 2001). The independent variables are controllable whereas responses are dependent. Contour plots give a visual representation of the values of the response. This helps the process of optimization by providing an empirical model equation for the response as a function of the different variables. Factorial designs have been applied earlier for optimization of formulation of buccal bioadhesive drug delivery systems (Mehta, Yadav, & Sawant, 2007; Narendra, Srinath, & Prakash Rao, 2005; Prakobvaitayakit & Nimmannit, 2003; Singh & Ahuja, 2002; Singh, Chakkal, & Ahuja, 2006).

Hydroxypropylmethyl cellulose (HPMC) is a well-known polymer, which acts as a release retardant as well as bioadhesive (Attia, El Gibaly, Shaltout, & Fetih, 2004; Shin, Cho, & Yang, 2004). HPMC is widely used to prepare extended-release dosage forms of both water-soluble and water-insoluble drugs because the drug release rates from HPMC matrix formulations are generally independent of process variables such as compaction pressure, drug particle size, and the incorporation of a lubricant (Ford, Rubinstein, & Hogan, 1985; Lahdenpaa, Niskanen, & Yliruusi, 1997; Sako, Sawada, Nakashima, Yokohama, & Sonobe, 2002; Zuleger &Lippold, 2001).

Gantrez MS, a novel polymer composed of poly(methyl vinyl ether-co-maleic anhydride) mixed calcium/sodium salt, which is reported to be used as a key ingredient in denture adhesive preparations, was chosen for its bioadhesive properties (Gantrez Brochure, 2005). This study was initiated with the intention of developing a new dosage form, a buccal adhesive tablet system, to improve absorption and bioavailability of 5-FU, and to enhance patient compliance. The formulation optimization was performed by application of factorial design.

MATERIALS AND METHODS

Materials

5-Fluorouracil was obtained as a gift sample from BDH Labs Ltd. (Mumbai, India). HPMC K4M and K15M were kindly provided by Alembic Ltd. (Baroda, India). Gantrez MS -955 was received from International Specialty Products (ISP) Ltd. (Mumbai, India) as a gift sample. Tablettose-100 (Lactose Monohydrate, E.P.) was generously gifted by Meggle (Deerlijk, Germany). All other chemicals and solvents used were of AR grade.

Experimental Design

A 3^2 factorial design was employed in which the amount of two carrier(s) (factors) was varied at three levels as required by the design. The amounts of Gantrez (X_1) and HPMC K15M (X_2) were selected as factors and studied at three levels each. Table 1 summarizes the nine experimental runs studied, their factor combinations, and the translation of the coded levels to the experimental units employed during the study. Bioadhesion (bioadhesive force or Y_1), release until 8 h ($Rel_{8\ h}$ or Y_2), time taken to release 50% of drug ($t_{50\%}$ or Y_3), and diffusion exponent (n or Y_4) were taken as the response variables.

Preparation of Bilayered Buccal Adhesive Tablets

Bilayered tablets (consisting of a backing layer and adhesive drug reservoir layer) were prepared by covering one side of single layer tablet with a layer of ethyl cellulose (EC). EC was selected as the backing material because this hydrophobic polymer has very low water permeability, thus providing an impermeable backing layer that can prevent drug loss in oral cavity (Miyazaki et al., 2000).

Drug-containing layer of the tablets was prepared by direct compression of drug blended with HPMC, Gantrez MS, and other excipients (on 8-station rotary machine, D-Tooling, Jaguar, General Machinery, Mumbai, India) using 9.5-mm flat-faced punches at a lower hardness (2–3 kg/cm^2). Then the

TABLE 1
Factor Combinations as per 3^2 Factorial Design

	Coded Factor Levels	
Trial No.	Factor 1	Factor 2
1	−1	−1
2	−1	0
3	−1	1
4	0	−1
5	0	0
6	0	1
7	1	−1
8	1	0
9	1	1

Translation of Coded Levels in Actual Units

Coded level	−1	0	+1
X_1: Gantrez MS 855 (mg)	10	20	30
X_2: HPMC K15M (mg)	4	10	16

All batches contained 20 mg 5-FU, HPMC K4M (4 mg), HPMC K15M (4–16 mg), Gantrez MS 955 (10–30 mg), magnesium stearate (2 mg), and quantity sufficient of Tablettose-100. Weight of drug containing layer—155 mg. Backing layer contained ethyl cellulose 20 cps (70 mg).

backing layer was compressed (consisting of Ethyl cellulose [EC; 20 cps]) on the drug-containing layer to obtain bilayered tablets with final hardness at 4–5 kg/cm^2.

Evaluation of the Buccal Adhesive Tablets

Twenty tablets from each batch were weighed individually and their average weight and standard deviation were calculated. The hardness and friability were measured by taking 3 and 10 tablets, respectively, from each batch. The diameter and thickness of 10 tablets from each batch were also determined.

Content Determination

Five tablets were accurately weighed and powdered. Drug content of the formulations was determined by dispersing and shaking an accurately weighed quantity of formulation equivalent to 10 mg of 5-FU in 50 mL of phosphate buffer (pH 6.8). This dispersion was filtered and quantitatively transferred to volumetric flask and appropriate dilutions were made with the same buffer solution. The resulting solution was then filtered through 0.45-μm membrane filter and subjected to UV spectrophotometric analysis at 267 nm (Jonnalagadda & Robinson, 2000).

Measurement of Bioadhesion

The tensile strength required to detach the bioadhesive tablet from the mucosal surface was taken as a measure of the bioadhesive performance. The apparatus used was locally assembled on the lines of a previously described apparatus (Agrawal & Mishra, 1999).

A circular piece (diameter 3.6 cm) of porcine buccal mucosa was cut, separated from connective tissue, and glued with cyanoacrylate adhesive on the ground surface of a tissue holder made of plexiglass. Similarly, the tablet was glued from the backing membrane containing side to another tissue holder of the same size. Thereafter, the two tissue holders carrying mucosa and tablet were put in contact with each other under uniform and constant pressure for 5 min (preload time) to facilitate adhesion bonding. The tissue holder with mucosa was allowed to hang on an iron stand with the help of an aluminum wire attached with a hook provided on the back side of the holder. A preweighed lightweight polypropylene bottle was attached to the hook on the back side of the formulation holder with an aluminum wire. After a preload time of 5 min, water was added to the polypropylene bottle through an intravenous infusion set at a rate of 1 drop/s until the tablet detached from the mucosal tissue. The water collected in the bottle was measured and expressed as weight (g) required for the detachment and finally converted into dynes/cm^2 as per a reported formula (Varma, Singla, & Dhawan, 2004). All experiments were carried out in triplicate.

In Vitro Release Study

The tablets were stuck onto the bottom of the dissolution vessel from its backing side with a drop of adhesive (Araldite®). A volume of 500 mL of prewarmed (37 ± 0.5°C) dissolution medium (phosphate buffer, pH 6.8) was gently poured into the vessel and dissolution was conducted as per United States Pharmacopoeia (USP) method II. The paddle speed was kept at 50 rpm. The 5-mL samples were collected at predetermined intervals and replaced with 5 mL of fresh buffer after each sample collection. The samples were filtered through a 0.45-μm membrane filter, suitably diluted, and analyzed at 267 nm using double-beam UV–Visible spectrophotometer (UV-1601, Shimadzu, Tokyo, Japan). The drug content was calculated using the equation generated from the standard curve. All experiments were carried out in triplicate, average values plotted, and the release kinetics were calculated.

Optimization Data Analysis

Various RSM computations for the current optimization study were performed employing Design Expert® software (version 7.1.2, Stat-Ease Inc., Minneapolis, MN, USA). Polynomial models including the interaction and quadratic terms were generated for all the response variables using multiple regression analysis (MLRA) approach. The general form of MLRA model is represented as

$$Y = B_0 + B_1X_1 + B_2X_2 + B_3X_1^2 + B_4X_2^2 + B_5X_1X_2 + B_6X_1^2X_2 + B_7X_1X_2^2, \quad (1)$$

where B_0 is the intercept representing the arithmetic average of all quantitative outcomes of nine runs; B_1 to B_7 are the coefficients computed from the observed experimental values of Y; and X_1 and X_2 are the coded levels of the independent variable(s). The terms X_1X_2 and X_i^2 (i = 1, 2) represent the interaction and quadratic terms, respectively. The main effects (X_1 and X_2) represent the average result of changing one factor at a time from its low to high value. The interaction terms (X_1X_2) show how the response changes when two factors are changed simultaneously. The polynomial terms (X_1^2 and X_2^2) are included to investigate nonlinearity. The polynomial equation was used to draw conclusions after considering the magnitude of coefficients and the mathematical sign it carries, that is, positive or negative. A positive sign signifies synergistic effect, whereas a negative sign indicates an antagonistic effect.

Statistical validity of the polynomials was established on the basis of ANOVA provision in the Design Expert® software. Level of significance was considered at $p < .05$. The best-fitting mathematical model was selected based on the comparison of several statistical parameters, including the coefficient of variation (CV), the multiple correlation coefficient

(R^2), the adjusted multiple correlation coefficient (adjusted R^2), and the predicted residual sum of squares (PRESS), provided by the software. PRESS indicates how well the model fits the data, and for the chosen model, it should be small relative to the other models under consideration (Huang, Tsai, Lee, Chang, & Wu, 2005). The 3-D response surface graphs and the 2-D contour plots were also generated by the Design Expert® software. Subsequently, the desirability approach was used to generate the optimum settings for the formulations (Huang et al., 2005; Narendra et al., 2005).

Investigation of Drug–Excipient Interactions

Physicochemical interactions between drug and excipients were investigated using differential scanning calorimetry (DSC) and Fourier transform infrared (FTIR) techniques. DSC study was performed using a Differential Scanning Calorimeter (Shimadzu DSC 60) at a heating rate of 10°C/min in nitrogen atmosphere.

FTIR spectra were recorded using a Shimadzu-8300 FTIR Spectrophotometer (Tokyo, Japan). Samples were prepared in KBr discs by means of a hydraulic press. The scanning range was 500–4,000 cm^{-1} and the resolution was 4 cm^{-1}.

RESULTS AND DISCUSSION

Evaluation of the Matrix Tablets

The weight of tablets varied between 223.1 and 228.9 mg, diameter between 9.5 and 9.6 mm, thickness between 2.05 and 2.15 mm, and hardness ranged between 4 and 5 kg/cm^2. The content of 5-FU varied between 96.7 and 99.8% and friability ranged between 0.1 and 0.4%. Thus, all formulations were found to comply with compendial requirements.

Effect of Formulation Variables on Bioadhesion Force

Table 2 lists the values of various response parameters of the nine optimization batches .The constant and regression coefficients for Y_1 (bioadhesion force) are as follows:

$$Y_1 = +10.766 + 1.975X_1 + 1.348X_2 - 1.11E - 016X_1X_2 + 0.851X_1^2 + 0.4175X_2^2 - 0.0965X_1^2X_2 - 0.0485X_1X_2^2, \quad R^2 = .9998. \tag{2}$$

The polynomial model was found to be significant with an F value of 4,792.24 (p = .0111). The value of correlation coefficient was found to be 0.9998, indicating a good fit. Equation 2 reveals that both the factors (X_1 and X_2) affect bioadhesion force, Y_1. The low value of X_1X_2 coefficient suggests that the interaction between X_1 and X_2 is not significant.

The combined effect of factors X_1 and X_2 can further be elucidated with the help of response surface and contour plots (Figure 1A and B), which demonstrate that Y_1 varies in a linear fashion with the amount of both the polymers. However, the steeper ascent in the response surface with Gantrez (X_1) than with HPMC K15M (X_2) is clearly discernible from both the plots, indicating that the effect of Gantrez is comparatively more pronounced than that of HPMC K15M. From this discussion, one can conclude that the bioadhesion may be changed by appropriate selection of the levels of X_1 and X_2. Figure 1C shows a linear relationship between the observed response values and the predicted values indicating the correctness of the model.

Effect of Formulation Variables on the Percentage 5-FU Release at 8 h

When the model terms for Y_2 (5-FU release at 8 h [$Rel_{8\,h}$]) were fitted in the polynomial model, they were found to be

TABLE 2
Response Parameters for 5-FU Buccal Bioadhesive Tablets Prepared as per 3^2 Factorial Design

Formulation Code	Formulation Composition: Gantrez MS-955 (mg)	Formulation Composition: HPMC K15M (mg)	Bioadhesion ($\times 10^3$ dyn/cm^2) [Y_1]	Release till 8 h ($Rel_{8\,h}$) [Y_2]	Time for 50% Drug Release, $t_{50\%}$ (min) [Y_3]	Diffusion Coefficient, n [Y_4]
F1 (–1, –1)	10	4	8.86	99.60	72.00	0.3807
F2 (–1, 0)	10	10	9.63	99.22	96.00	0.4225
F3 (–1, 1)	10	16	11.36	97.17	156.00	0.4795
F4 (0, –1)	20	4	9.82	98.83	114.00	0.4546
F5 (0, 0)	20	10	10.78	96.44	168.00	0.5516
F6 (0, 1)	20	16	12.52	86.56	186.00	0.5753
F7 (1, –1)	30	4	12.71	100.01	60.00	0.3234
F8 (1, 0)	30	10	13.58	99.12	74.00	0.3560
F9 (1, 1)	30	16	15.21	98.14	120.00	0.4493

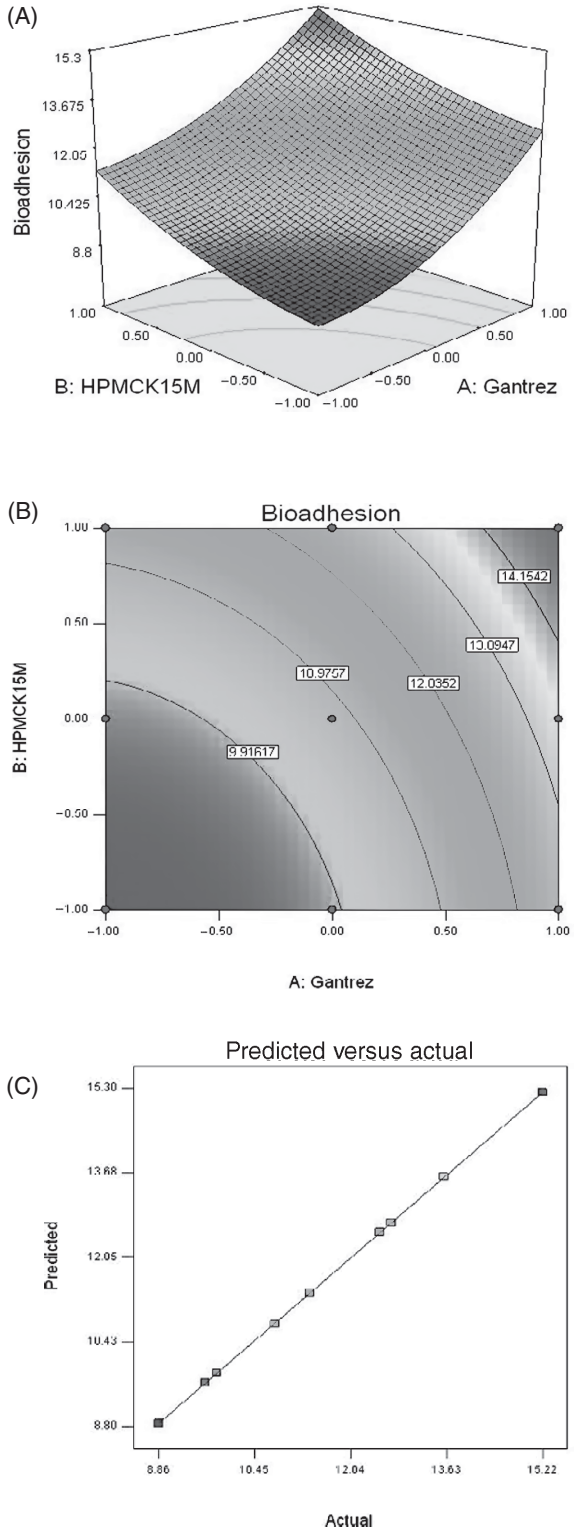

FIGURE 1. (A) Response surface plot showing the influence of Gantrez and hydroxypropylmethyl cellulose (HPMC) K15M on the bioadhesive force (Y_1). (B) Corresponding contour plot showing the relationship between various levels of the two polymers. (C) Plot between observed and predicted values of Y_1.

nonsignificant (p = .3693) with an F value of 3.96 and R^2 value of .9652. However, on reducing the model to a quadratic one, the F value was 1.54 (p = .3839), all the factors were found to be nonsignificant and the R^2 value reduced significantly to .7194. As per the literature reports, low PRESS values indicate adequate fitting of model (Huang et al., 2005). Therefore, the quadratic model with the lower PRESS value of 455.14 was selected, as the PRESS value of polynomial model was very high (887.44). The quadratic model describing the percentage 5-FU release at 8 h ($Rel_{8\,h}$) can be written as

$$Y_2 = +94.92 + 0.223X_1 - 2.77X_2 + 0.16X_1X_2 + 4.956X_1^2 - 1.543X_2^2, \quad R^2 = .7194. \tag{3}$$

Figure 2A displays a nonlinear twisted relationship for $Rel_{8\,h}$ at intermediate and high levels of the polymers. This can be attributed to the occurrence of potential interaction between the two polymers at the corresponding factor levels, construing that each polymer tends to modify the effect of the other toward the drug release. The contour plot (Figure 2B) shows that Gantrez has a comparatively greater influence on the response variable than HPMC K15M. A high value of $Rel_{8\,h}$ was observed in the formulation having a low value of either of the independent variables (Table 2), which may be because of the weakened gel strength of the matrix at lower polymer concentration. A high level of X_2 gave a low value of $Rel_{8\,h}$ at all the three levels of X_1 which might be because of the increased thickness of the gel layer, which forms on contact with the dissolution medium and in turn retards drug diffusion.

An increase in $Rel_{8\,h}$ was observed at higher levels of X_1. This can be explained by the fact that when a soluble polymer like Gantrez is used, the voids formed in the matrix due to dissolution initially contain a rather viscous solution, characteristic of dissolved polymers. The high viscosity in the pores serves to retard the drug diffusion at the early stages of release. At later stages, the polymer solution becomes dissipated and resistance to diffusion is decreased (Korsmeyer, Gurny, Doelker, Buri, & Peppas, 1983). Tablets with a high percentage of Gantrez exhibited visible erosion. From the results, it can be concluded that both the independent variables have pronounced effect on $Rel_{8\,h}$. Figure 2C represents the observed response values compared with that of predicted values indicating the correctness of the model.

Effect of Formulation Variables on $t_{50\%}$

The quadratic model for $t_{50\%}$ (Y_3) was found to be significant (p = .0199) with an F value of 17.49. In this case, both the factors X_1 and X_2 as well as the interaction factor X_1X_2 were found to be significant. Thus, the model becomes

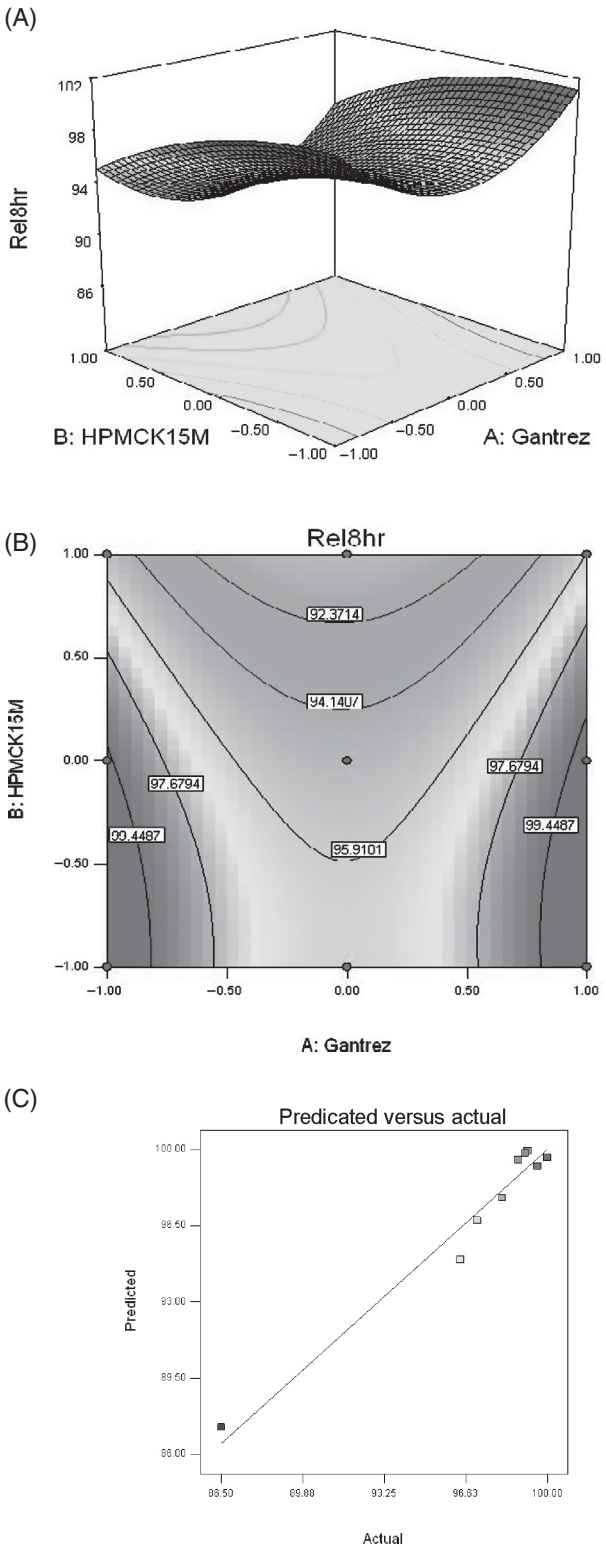

FIGURE 2. (A) Response surface plot showing the influence of Gantrez and hydroxypropylmethyl cellulose (HPMC) K15M on $Rel_{8\ h}$ (Y_2). (B) Corresponding contour plot showing the relationship between various levels of the two polymers. (C) Plot between observed and predicted values of Y_2.

$$Y_3 = +152.44 - 11.666X_1 + 36.00X_2 - 6.00X_1X_2 - 59.66X_1^2 + 5.33X_2^2, \quad R^2 = .9668. \tag{4}$$

Figure 3A and B shows the "region of maximum" lying between intermediate-to-high levels of both the polymers. The variables had a significant effect on $t_{50\%}$ ($p < .05$, Table 2). A linear relationship was obtained between the fraction of HPMC K15M and $t_{50\%}$, and it was observed that as the fraction of HPMC K15M increased, the value of $t_{50\%}$ increased, at all the three levels of Gantrez (Figure 3A and B). On increasing the amount of Gantrez, a nonlinear trend was observed in the response curve, that is, the "region of maximum" for $t_{50\%}$ at intermediate levels of X_1 at all the three levels of X_2. The contour lines (Figure 3B) depict a nonlinear relationship between the factors with region of maximum for $t_{50\%}$. It may be because a high level of HPMC K15M will form a highly viscous gel that will increase the diffusion barrier, decrease the release rate, and in turn increase the $t_{50\%}$. The nonlinear effect of Gantrez on $t_{50\%}$ could be due to the erosion of the polymer at higher concentrations. Similar results have been reported by Owens, Dansereau, and Sakr (2005). Figure 3C represents the observed response values compared with that of the predicted values depicting a good fit.

Effect of Formulation Variables on Diffusion Exponent (*n*)

Dissolution profiles were fitted to the power-law equation (Korsmeyer et al, 1983; Peppas, 1985) to calculate the values of diffusional exponent (n). Using an aspect ratio (diameter/thickness) of 4.41, the critical values of n for declaring Fickian diffusion, non-Fickian difusion, and zero-order release were found to be 0.45, $0.45 < n < 0.89$, and 0.89, respectively (Peppas, 1985).

The quadratic model for n or Y_4 was found to be significant with an F value of 21.16 ($p = .0152$). In this case, factors X_1 and X_2 along with interaction factor X_1X_2 were found to be significant. Thus, the model then becomes

$$Y_3 = +0.5268 - 0.025X_1 + 0.0575X_2 + 0.0067X_1X_2 - 0.125X_1^2 + 0.00043X_2^2, \quad R^2 = .9724. \tag{5}$$

Equation 5 shows that both the factors X_1 and X_2 affect the values of release exponent n. The value of correlation coefficient was 0.9724, indicating a good fit.

Figure 4A and B shows the "region of maximum" for n, lying between intermediate-to-high levels of both the polymers. The values for n ranged from 0.32 to 0.57, which

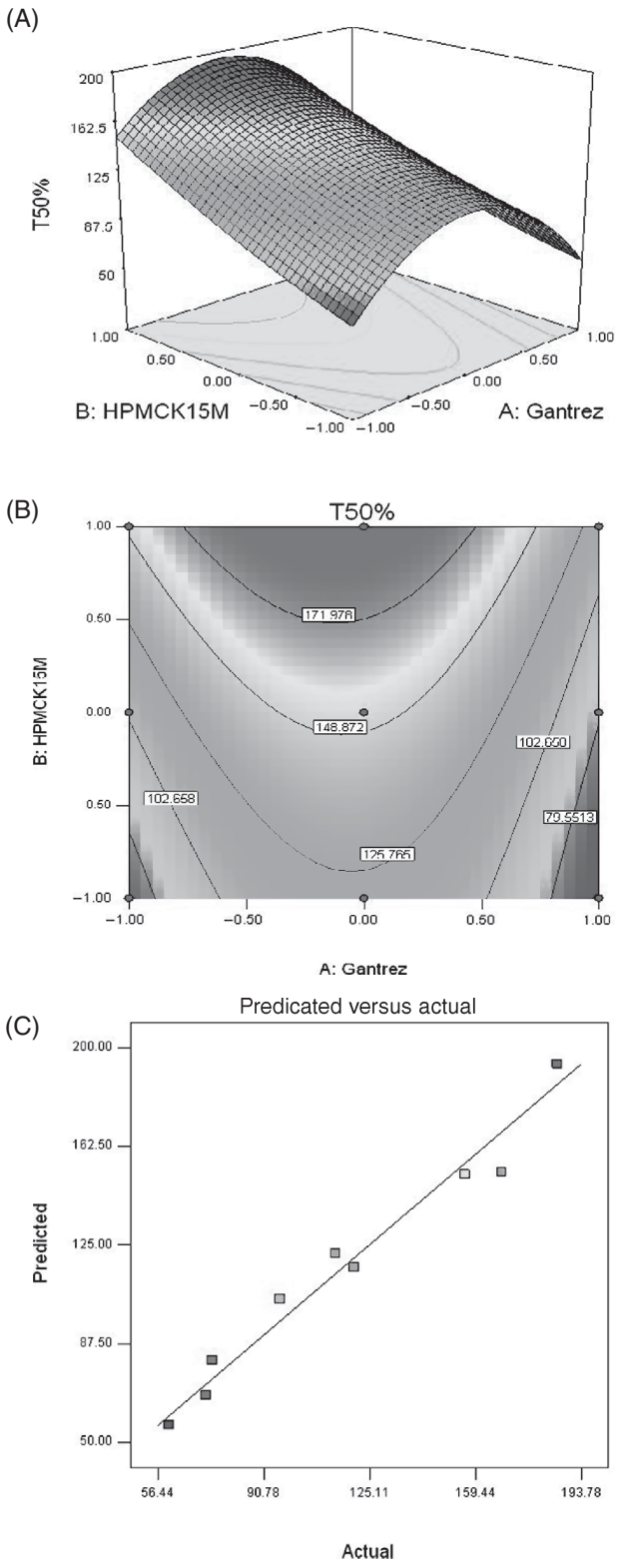

FIGURE 3. (A) Response surface plot showing the influence of Gantrez and hydroxypropylmethyl cellulose (HPMC) K15M on $t_{50\%}$ (Y_3). (B) Corresponding contour plot showing the relationship between various levels of the two polymers. (C) Plot between observed and predicted values of Y_3.

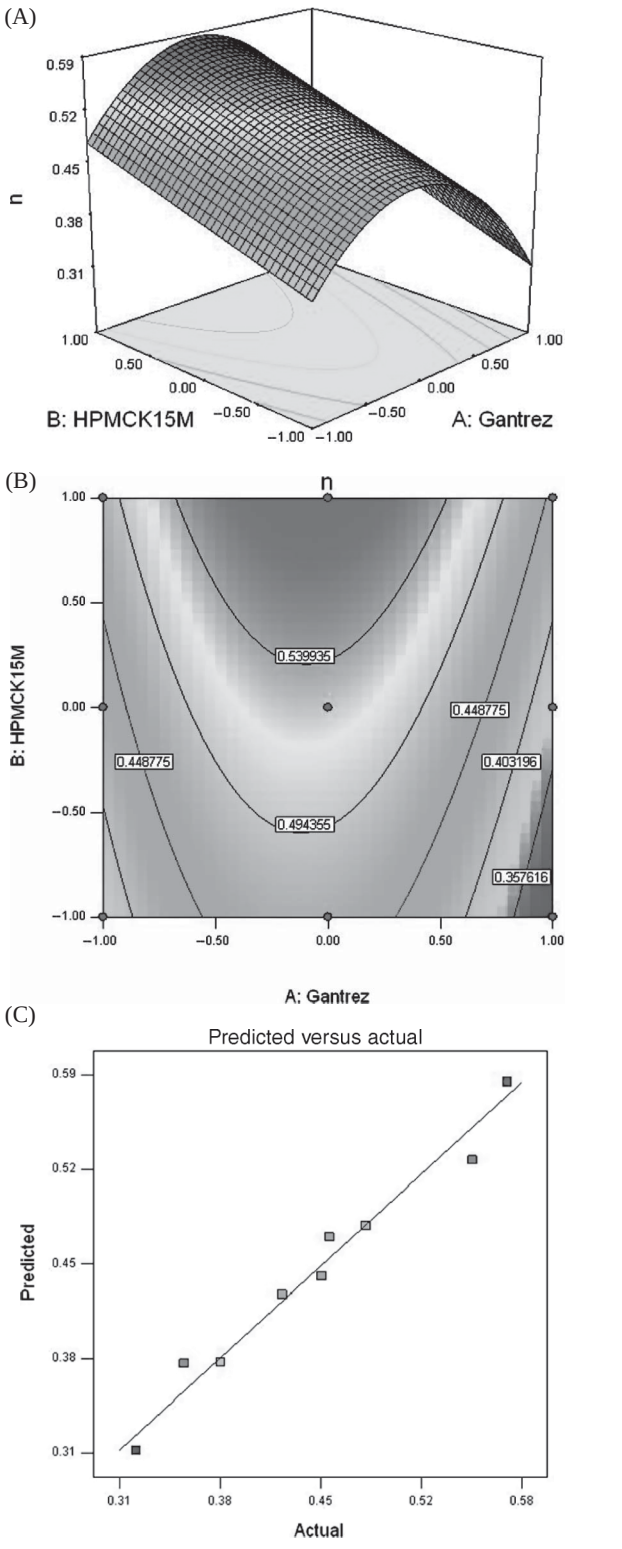

FIGURE 4. (A) Response surface plot showing the influence of Gantrez and hydroxypropylmethyl cellulose (HPMC) K15M on diffusion coefficient n (Y_4). (B) Corresponding contour plot showing the relationship between various levels of the two polymers. (C) Plot between observed and predicted values of Y_4.

indicates that the independent variables have a significant effect on the mechanism and kinetics of drug release. A linear relationship was obtained between HPMC K15M and *n*, and as the fraction of HPMC K15M increased, the value of *n* increased, at all the three levels of Gantrez (Figure 4A and B).

The response values of *n* varied analogously to $t_{50\%}$ values with respect to variation in the levels of Gantrez. On increasing the amount of Gantrez, a nonlinear trend in the response curve was observed, that is, the "region of maximum" for *n* was seen at intermediate levels of X_1 at all the three levels of

TABLE 3
Multiple Regression Output for Dependent Variables

Parameters	B_0	B_1	B_2	B_3	B_4	B_5	B_6	B_7	R^2	*p*
Coefficient of Regression Parameters										
Bioadhesion	10.766	1.975	1.348	−1.11*E* – 016	0.851	0.4175	−0.0965	−0.0485	.9998	.0111
$Rel_{8\,h}$	94.92	0.223	−2.77	0.16	4.956	−1.543	–	–	.7194	.3839
$t_{50\%}$	152.44	−11.666	36.00	−6.00	−59.66	5.33	–	–	.9668	.0199
n	0.5268	−0.025	0.0575	0.0067	−0.1252	0.00043	–	–	.9724	.0152

$t_{50\%}$ indicates the time required for 50% of drug release; $Rel_{8\,h}$ the percentage drug release at 8 h; *n* is the diffusion exponent.

TABLE 4
Results of Analysis of Variance (ANOVA) for Measured Responses

Parameters	*df*	*SS*	*MS*	*F*	Significance *F*
Bioadhesion					
Model	7	34.35	4.91	4,792.24	0.0111
Residual	1	0.001	0.001	–	–
Total	8	34.35	–	–	–
$Rel_{8\,h}$					
Model	5	100.56	20.11	1.54	0.3839
Residual	3	39.23	13.08	–	–
Total	8	139.79	–	–	–
$t_{50\%}$					
Model	5	15,913.78	3,182.76	17.49	0.0199
Residual	3	545.78	181.93	–	–
Total	8	16459	–	–	–
n					
Model	5	0.055	0.011	21.16	0.0152
Residual	3	1571*E*–003	5.236*E*–004	–	–
Total	8	0.057	–	–	–

df, degrees of freedom; *SS*, sum of squares; *MS*, mean sum of squares; *F*, Fischer's ratio.

TABLE 5
The Predicted and Observed Response Variables of the Optimal 5-FU Buccal Bioadhesive Tablets

	Y_1	Y_2	Y_3	Y_4
Predicted	10.77	94.93	152.44	0.53
Observed	10.31 ± 0.89	98.21 ± 1.5	161.22 ± 4.6	0.55 ± 0.09
Predicted error (%)	4.27	3.45	5.75	3.77

Predicted error (%) = (observed value − predicted value)/predicted value × 100%.

X_2. The release mechanism changed from Fickian to anomalous (non-Fickian) up to maxima and then again reverted to Fickian diffusion on increasing the amount of Gantrez to the maximum level. The above outcome could be because of the water-soluble nature of Gantrez. Figure 4C shows a linear relationship between the observed response values compared with the predicted values indicating the correctness of the model.

Regression analysis was carried out to obtain the regression coefficients (Table 3). The results of ANOVA for the dependent variables (Table 4) demonstrate that the model was significant for all response variables except for Y_2, which was further analyzed by PRESS analysis for appropriate model fitting.

Optimization

A numerical optimization technique by the desirability approach was used to generate the optimum settings for the formulation. The process was optimized for the dependent (response) variables Y_1 to Y_4 and the optimized formula was arrived at by keeping the bioadhesion force greater than 10 dyn/cm^2. The 5-FU release at 8 h was targeted to greater than 90% with $n > 0.5$ and $t_{50\%}$ was kept between 150 and 175 min. The formulation F5 fulfilled all the criteria set from the desirability search. To gainsay the reliability of the response surface model, a new optimized formulation (as per formulation F5) was prepared according to the predicted model and evaluated for the responses. The results in Table 5 illustrate a good relationship between the experimental and predicted values, which confirms the practicability and validity of the model. The predicted error for all the response variables was below 6% indicating that the RSM optimization technique was appropriate for optimizing the 5-FU bioadhesive buccal tablets.

Investigation of Drug–Excipient Interactions

The DSC thermogram gives information regarding the physical properties like crystalline or amorphous nature of the samples (Clas, Dalton, & Hancock, 1999). The DSC thermogram of 5-FU shows an endothermic peak at 283.5°C corresponding to its melting temperature, which was also detected in the thermogram of the physical mixture of tablet components (5-FU, Gantrez, HPMC, Tablettose-100, magnesium stearate) at the same temperature, signifying no interaction between 5-FU and other tablet components (Figure 5).

The FTIR spectra of pure 5-FU showed sharp characteristic peaks at 812 (C–H out-of-plane), 1,243 (C–H in-plane), 1,425, 1,655 and 1,718 (C=O, C=N stretch), and 3,122 cm^{-1} (N–H stretch). All the above characteristic peaks appeared in the spectra of the physical mixture of tablet components at the same wave numbers indicating no modification or interaction between the drug and the tablet components.

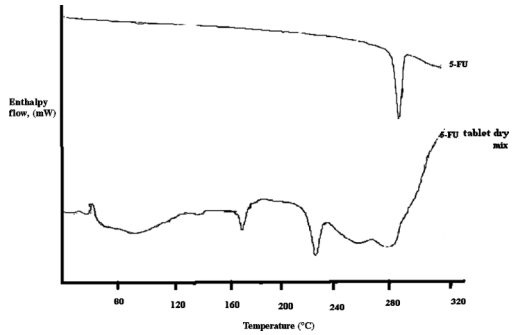

FIGURE 5. Differential scanning calorimetry (DSC) thermograms of 5-flurouracil (5-FU) and 5-FU tablet dry mixture.

CONCLUSION

The study suggests that the hydrophilic bioadhesive tablets of 5-FU can be designed using HPMC and Gantrez to provide extended release. The matrices demonstrated adequate bioadhesion with buccal mucosa. The 5-FU containing buccoadhesive tablets could provide an alternative to the conventional dosage form for the treatment of oropharyngeal cancer.

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