

Development and Evaluation of Chitosan and *Aloe Vera* Gel Mucilage Interpolymer Complex-Based Mucoadhesive Buccal Films of Tramadol Hydrochloride

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Abstract

Aim: To develop and evaluate interpolymer complex based buccal mucoadhesive films of Tramadol Hydrochloride. The interpolymer complex was prepared by using Chitosan and Aloe vera gel mucilage. **Materials and Methods:** In present study the inter polymer complex is formed between mucoadhesive polymer chitosan and aloe vera gel mucilage. The resulted interpolymer complex is used to prepare buccal mucoadhesive films of Tramadol Hydrochloride, an opioid analgesic. Other excipients includes glycerin as plasticizer, a combination of Sodium Dihydrocholate and EDTA sodium salt as permeation enhancers and a backing layer of 1% ethyl cellulose is placed on each film to ensure unidirectional drug release to systemic circulation. The solvent casting method is used to prepare buccal films and evaluated for bioadhesion strength, tensile strength, swelling index, ex vivo diffusion and in vitro dissolution. **Results and Discussion:** Formulations are prepared and optimized by 3² factorial design. Formulation F7 was found to be optimized formulation which contained 50 mg drug, 100 mg IPC and 2% glycerin as plasticizer. Thus, this study suggests that interpolymer complex between chitosan and aloe vera gel mucilage can act as a potential mucoadhesive polymer system for buccal delivery of a drug like Tramadol Hydrochloride. **Conclusion:** In this study novel buccoadhesive film was developed using Inter polymer complex between chitosan and aloe vera gel mucilage. Film was releasing drug over a period of 8hr directly to systemic circulation through buccal mucosa. The extensive first pass metabolism of a drug was prevented to a great extent.

Key words: Alovera gel mucilage, chitosan, factorial design, interpolymer complex, mucoadhesive films, optimization

INTRODUCTION

Among all transmucosal drug delivery systems, mucoadhesive drug delivery systems offer benefits over conventional delivery methods in terms of extended residence time of the drug at the site of application. Other than this it offers advantages such as faster uptake of drug into the systemic circulation and enhanced bioavailability of therapeutic agents, leading to rapid onset of action.^[1] Buccal drug delivery systems release drug in the region of the buccal cavity from which drug directly absorbed through the venous systems drains from the cheek and thus bypass the first pass metabolism.^[2] Buccal mucosa is an attractive route for systemic delivery of drugs since it is relatively permeable, with rich blood supply.^[9] In the case of toxicity buccal drug absorption

can be promptly terminated by removing the dosage form from the buccal cavity. Attempts have been made earlier to formulate various buccoadhesive devices, including tablets,^[10] films,^[11] patches,^[12] disks^[13] and strips.^[14] Among all buccal films are more preferable because of flexibility and comfort.^[15]

Natural polysaccharides are very popular as biopolymers mainly because they are biocompatible and biodegradable

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in nature. In this study chitosan - *A. vera* gel mucilage, interpolymer complex was used as bioadhesive polymer to increase the residence time of the dosage form in buccal cavity. This interpolymer complex swells in aqueous media to form a gel through which the drug has to diffuse thus they can be used to control the rate of drug release.^[1]

Properties such as biocompatibility and biodegradability have attracted many researchers to utilize natural polysaccharides in the development of different drug delivery systems.^[3] In this study *A. vera* gel mucilage, polysaccharide and acetylated-glucomannan are located within the protoplast of the parenchyma cells, and a variety of polysaccharides are present in the cell wall matrix. Aloe leaf holds mainly mannose - Containing polysaccharides, cellulose and pectic polysaccharides, whereas the skin of the leaf contains xylose - Containing polysaccharides.^[16] Apart from being mucoprotective in nature they can be used to prepare highly viscous aqueous solution which remains stable within a wide range of pH. They are noncarcinogenic and can hold high amount of drug.^[4]

Tramadol hydrochloride is a centrally acting opioid analgesic mainly used to treat moderate to severe pain conditions. It is having good absorption after oral administration, but the reason for poor oral bioavailability is extensive first pass metabolism via N and O-demethylation and glucuronidation or sulfation in the liver.^[17]

Interpolymer complex of chitosan and *A. vera* gel mucilage has not been reported for the use of development of buccoadhesive drug delivery systems. An attempt has been made in the present study to utilize *A. vera* gel mucilage which is widely available, mucoprotective in nature and a more economical source of polysaccharides in the development of buccal film of tramadol hydrochloride.

MATERIALS AND METHODS

Materials

Tramadol hydrochloride was obtained as a gift sample from Karnataka Antibiotics & Pharmaceuticals Ltd. (Bengaluru). *A. vera* leaves were procured from local nursery near Surat. Chitosan was purchased from Pure Chem Pvt. Ltd. (Ankleshwar). All other chemicals were purchased from Merck Ltd. (Mumbai).

Experimental design

Optimization of buccal films was done using a 3² randomized full factorial design. This method includes evaluation of two factors individually at three levels shown in Table 1. Different codes such as -1, 0 and +1 were given to lower, medium and higher levels of both variables. Two independent variables were the amount of interpolymer complex in specific ratio of drug (X₁) and the concentration of

glycerin (plasticizer) (X₂). Tensile strength (Y₁), bioadhesion force (Y₂), and % drug release at 8 h (Y₃) were selected as response variables.

Preparations of mucoadhesive buccal films

The compositions of all formulations are shown in Table 2. All mucoadhesive buccal films were prepared by solvent casting method. Chitosan was dissolved in 60 ml 5 M acetic acid to produce 2.5% chitosan solution. To this solution, 10 ml 5 M ammonium solution was added. The drug was dissolved in 40 ml 2.5% carboxymethylated mucilage solution with constant stirring for 15 min using mechanical stirrer. Glycerin was used as plasticizer. This solution was sonicated for 45 min to remove air bubbles. This solution was poured in petri dish of size 8 cm in diameter and was dried in vacuum oven at 55°C for 24 h. The backing layer of 1% ethylcellulose was placed by pouring solution over medicated buccal film and dried in vacuum oven at 55°C for 4 h. Dried films were cut into 1.5 cm² patches containing 50 mg of drug in each patch.

Characterization of buccal films

Thickness and weight

The thickness of film was measured using micrometer screw gage. For each formulation, three films were selected randomly with surface area 1.5 cm². The weight of individual films was noted down using analytical balance. Average weight was then calculated.

Swelling studies

Swelling index study is useful to find out and compare the water absorption characteristics of film polymers. Pre-weighed films (designated as w₁) are placed separately in petri plate having phosphate buffer 6.8 pH. At regular intervals (5, 10, 15, 20, 25, 30, 35, 40, 60 min), films were removed from the petri plate. Excess water was removed carefully using filter paper.^[3] The swollen films were reweighed (w₂). The following formula was used to calculate swelling index.^[5]

$$\text{Swelling index} = \frac{W_2 - W_1}{W_1} \times 100 \quad (1)$$

Measurement of surface pH

Surface pH of film was found out to see whether the film can cause irritation to the mucosa or not. The surface pH study was performed by selecting 3 films randomly. Digital pH meter was used to find out pH. The pH electrode was placed in close contact with the wetted film surface and pH was recorded for each film.^[5]

Folding endurance

The Folding endurance was determined to check flexibility of films. All selected films were folded repeatedly at same place

until they broke to determine folding endurance. The action was repeated until films broke or were folded for 300 times which ever is less.^[6]

Tensile strength

Texture analyzer (CT-3/10,000, Brookfield, USA) equipped with a 10 kg load cell was used to check tensile strength of the formulation. The film of 200 mm² was randomly selected and was fixed between the two clamps of probe TA-DGA and for a hold time of 60 s. The lower clamp was held stationary, and the film was pulled apart by the upper clamp. Film was pulled at a speed of 2.0 mm/s to a distance of 6 mm with trigger load 0.05 N. The force of the film at the point when the film broke was recorded.^[7]

Texture- Pro CT V1.3 Build 14 Software was used for data collection and calculations. The tensile strength break value was calculated using formula:

$$\text{Tensile strength (kg/mm}^2\text{)} = \text{Force at break/initial cross sectional area} \quad (2)$$

In vitro bioadhesion force

Texture Analyzer (CT-3/100, Brookfield, USA) equipped with a 100 g load cell was used to determine the bioadhesion force of buccal patches. The porcine buccal mucosa was used as the model membrane for the measurement of buccal mucosa. The mucosal membrane was isolated by removing the underlying connective tissue. The mucosal membrane was washed thoroughly with phosphate buffer (pH 6.8). Then, the membrane was fixed between two circular discs which were at lower Perspex support. The upper circular disc had a cavity of 12.7 mm diameter through which the mucosal membrane was exposed to the probe. The discs were lowered into the jacketed glass container filled with phosphate buffer (pH 6.8) which was maintained at $37 \pm 1^\circ\text{C}$. The test was started once the membrane was equilibrated at $37 \pm 1^\circ\text{C}$ for 30 min. The buccal film was firmly tight with the help of thread on the lower side of probe. The probe and circular cavity were aligned in such a way that film comes into direct contact with exposed surface of the mucosal membrane. Exposed area of buccal film was moistened with phosphate buffer pH 6.8 before test starts. The probe was lowered at a speed of 0.5 mm/s to contact the tissue with load, 90 g and with contact time 120 s. It was removed at the speed of 2 mm/s.^[8]

Texture-Pro CT V1.3 Build 14 software was used for data collection and processing. The adhesive force and adhesiveness were found out to evaluate the bioadhesive strength of film. Bioadhesion force (N) was calculated using formula:

$$\text{Bioadhesion force (N)} = (\text{Bioadhesive strength [g]/1000}) \times 9.81 \quad (3)$$

RESULTS AND DISCUSSION

Nine different formulations were prepared using 3² randomized full factorial design. Design Expert software 8.0.6 was used to process various data collected by experimental processes. Various models such as linear, 2FI, quadratic and cubic were fitted to the data for two responses simultaneously using and adequacy and good fit of models were tested using analysis of variance. The formulation chart prepared by factorial design is shown in Table 3.

Characterization of buccal films

Physicochemical characteristics of the bilayer films are shown in Tables 3 and 4.

Thickness and weight

The average thickness of buccal films is found out ranging from 0.35 to 0.45 mm. the weight variation values for films are ranging from 106 to 413 mg. Thus, it can be concluded that the increase in weight is well supported by the increase in thickness of films. This data indicates that the film was casted uniformly.

Swelling studies

Swelling characteristics of films shows that as the concentration of IPC increases more swelling was observed in films. Thus, this study confirms that increase in swelling index is mainly because of increase in the concentration of IPC. Swelling index of film is directly associated with the release of the drug.

Measurement of surface pH

Surface pH for formulation F1-F9 was found to range from 6.67 to 7.21. Since range of film is near to the salivary pH, no mucosal irritation was expected.

Folding endurance

The folding endurance of films was found to increase with an increase in glycerin concentration. The values range from 245 to 318 which show that all films have high mechanical strength. This is highly desirable because it would not allow

Table 1: Translation of coded levels in actual units

Variable levels	Low (-1)	Medium (0)	High (+1)
IPC: Drug (X ₁)	0.5:1	1:1	2:1
% Plasticizer glycerin (X ₂)	2	4	6

Table 2: Composition of various buccal film formulations

Ingredients	Formulations and quantity								
	F1	F2	F3	F4	F5	F6	F7	F8	F9
Tramadol Hydrochloride (mg)	50	50	50	50	50	50	50	50	50
Interpolymer complex (mg) (CH:M, 60:40)	25	50	100	50	100	25	100	25	50
Glycerin (% w/v)	2	4	6	2	4	6	2	4	6
Sodium dihydrocholate (% w/v)	1.25	1.25	1.25	1.25	1.25	1.25	1.25	1.25	1.25
EDTA disodium salt (% w/v)	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5
Ethyl cellulose (1% w/v)	Backing layer on F1-F9 formulations								

EDTA: Ethylenediaminetetraacetic acid

Table 3: Factorial design with corresponding 9 formulations

Batch no	Variable levels in coded form		Tensile strength (kg/mm ²) Y ₁	Bioadhesive force (N) Y ₂	Drug release (%) Y ₃
	X ₁	X ₂			
F1	-1	-1	10.76±0.13	0.39±0.08	84.62±0.6
F2	-1	0	13.05±0.18	0.46±0.07	86.46±0.4
F3	-1	+1	13.82±0.20	0.51±0.09	88.56±0.8
F4	0	-1	11.94±0.15	0.78±0.08	94.52±0.2
F5	0	0	14.88±0.15	0.82±0.10	95.45±0.4
F6	0	+1	16.24±0.12	0.91±0.11	93.28±0.6
F7	+1	-1	14.45±0.15	1.10±0.12	90.36±0.5
F8	+1	0	16.23±0.18	1.19±0.13	89.22±0.4
F9	+1	+1	18.33±0.11	1.23±0.11	88.39±0.4

Table 4: Result of physicochemical characteristics of the bilayer films

Code	Thickness (mm)	Weight uniformity (mg)	Swelling index	Surface pH	Folding endurance	Permeation study (%)
F1	0.35±0.02	145.31±7.56	32.80±0.3	6.88±0.2	307±1.55	82.63±0.5
F2	0.36±0.01	244.46±7.36	38.32±0.2	6.67±0.4	294±2.15	85.32±0.8
F3	0.41±0.01	413.03±5.17	36.88±0.3	6.79±0.3	245±1.45	83.21±0.5
F4	0.35±0.02	161.05±5.25	36.74±0.3	7.13±0.4	251±1.85	86.67±0.7
F5	0.45±0.01	469.16±7.11	38.18±0.2	7.18±0.7	262±2.35	87.18±0.3
F6	0.39±0.02	303.03±7.14	33.30±0.3	7.10±0.4	255±2.25	88.35±0.5
F7	0.41±0.02	323.73±5.75	45.94±0.2	7.21±0.8	267±1.65	94.42±0.8
F8	0.36±0.01	106.12±6.46	46.53±0.3	7.18±0.2	318±2.70	92.12±0.7
F9	0.41±0.01	322.89±5.38	44.65±0.2	7.13±0.7	263±1.55	90.73±0.4

easy dislocation of the film from the site of application or breaking of film during administration.

Effect of formulation variables on tensile strength

Tensile strength test data for all formulations show that films are sufficiently strong to withstand wear and tear occurring during handling, packaging, and transportation. The Tensile strength of formulations is in the range of 10.76-18.33 kg/mm². Results indicate satisfactory mechanical strength.

The constant and regression coefficient for Y₁ (tensile strength) are shown below:

$$Y_1 = 14.88 + 1.56X_1 + 2.21X_2 + 0.71X_1X_2 - 0.25X_1^2 - 0.80X_2^2 \quad (4)$$

The quadratic model was found to be significant with F=3226.66 ($P < 0.0001$) which shows that the model is significant. Figure 1 represents the contour plot showing the effect of different proportions of independent variables

on the response Y_1 . The increase in glycerin concentration at the same concentration of IPC is responsible for the increase in tensile strength. The combined effect of factor X_1 (IPC) and X_2 (Glycerin) can be further understood with the help of response surface plot [Figure 1]. High level of factor X_2 gave a high value of tensile strength at all levels of factor X_1 which shows that the factor X_2 has significant positive effect on tensile strength. Increase in concentration of glycerin and IPC were responsible for increase in tensile strength of buccal films.

Effect of formulation variables on *in vitro* bioadhesion force

Bioadhesion force is necessary to hold drug delivery system at the site of application during the course of treatment. Bioadhesion force is directly related to the swelling index. Higher the swelling index of polymer greater will be the

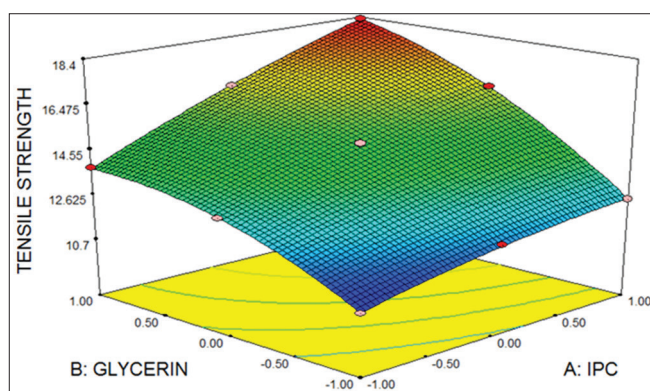


Figure 1: Two-dimensional contour plot and three-dimensional response surface plots for tensile strength

bioadhesion force. Formulations F7, F8, and F9 show higher values of bioadhesion force to its good swelling index. Increase in concentration of IPC mainly responsible in higher bioadhesion force of buccal films.

The constant and regression coefficient for Y_2 (bioadhesion force) are as follows:

$$Y_2 = 0.83 + 0.36 X_1 + 0.06 X_2 + 2.50 X_1 X_2 - 0.01 X_1^2 + 7.14 X_2^2 \quad (5)$$

The quadratic model was found to be significant with $F=575.59$ ($P < 0.0001$) that shows that the model is significant. Figure 2 represents the contour plot showing the effect of different proportions of independent variables on the response Y_2 . Increase in Bioadhesion force of buccal films is mainly because of increase in IPC concentration. The combined effect of factor X_1 (IPC) and X_2 (Glycerin) can be further elucidated with the help of response surface plot [Figure 2]. High level of factor X_1 gave high value of bioadhesion force at all the levels of factor X_2 which indicates that the factor X_1 has significant positive effect on bioadhesion force.

Effect of formulation variables on *in vitro* release of tramadol hydrochloride from buccal film

No significant release of drug was observed in any formulation until polymers swell completely, i.e., for 60 min. Formulations with a higher concentration of IPC show good swelling index values, greater hydration rates, which would permit faster and ready disentanglement of individual chains, thus increasing the porosity of the film and gives good release. Formulation F7 showed the highest drug release (94.42%) in

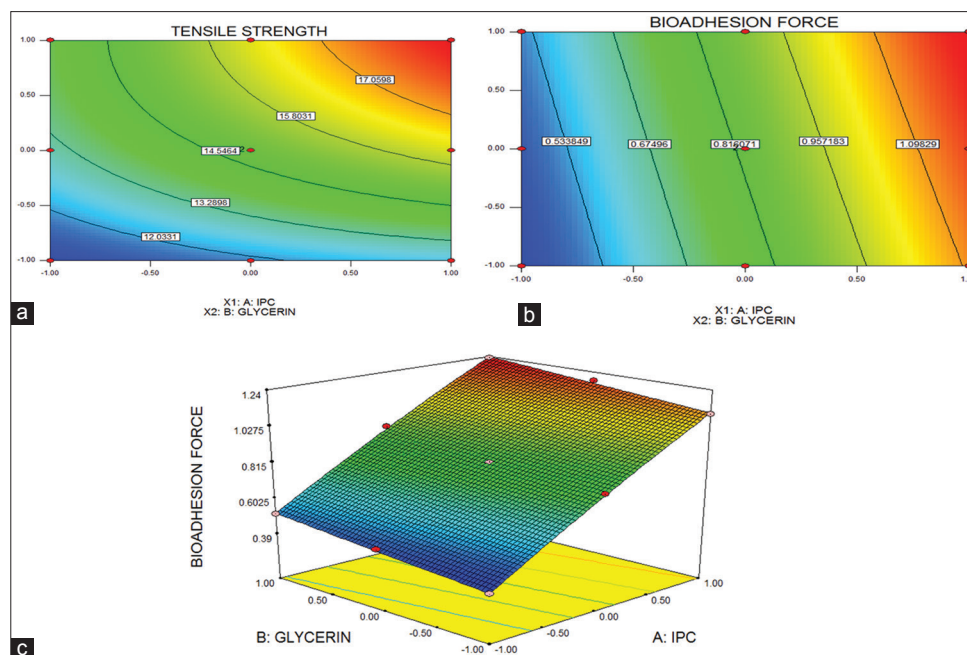


Figure 2: (a-c) Two-dimensional contour plot, three-dimensional response surface plots for bioadhesion force

8 h. This optimized formulation (F7) was subjected to various mathematical models to understand the release pattern. The value of coefficient of regression (R^2) was found to be 0.9631 for Korsmeyer's - Peppas and release exponent (n) was 0.5210 indicating that drug transport mechanism is mainly anomalous transport, i.e., drug release is being governed by both diffusion as well as erosion.

The constant regression coefficient for Y_3 (% drug release) is as follows:

$$Y_3 = 94.96 + 1.388X_1 + 0.12X_2 - 1.47X_1X_2 - 6.64X_1^2 - 0.58X_2^2 \quad (6)$$

The quadratic model was found to be significant with $F=31.69$ ($P < 0.0001$) which shows that the model is significant. Figure 3 represents the contour plot displaying the effect of different quantities of independent variables on the response Y_3 . Increase in cumulative percentage release was because of increase in the concentration of IPC and then declined. The combined effect of factor X_1 (IPC) and X_2 (Glycerin) can be further understood with the help of response surface plot. Medium level of factor X_2 gave high value of drug release which shows that the factor X_1 has significant positive effect on drug release.

Ex vivo permeation studies

All films have shown satisfactory results for *ex vivo* permeation of tramadol hydrochloride. Films containing higher amount of IPC have shown good permeation of drug compare to other formulations. Highest diffusion of around 94.42% was shown by formulation F7 followed by F8 and F9. Drug diffusion of formulation F1, F2 and F3 was less than other formulation. It may be because of poor swelling due to the lowest concentration of IPC. The rate limiting factor here is swelling index and which is directly related to concentration of IPC in individual formulation.

Optimization

The computer optimization technique by setting desirable values was selected to find out the optimum formulation.

The process was optimized for the response variables Y_1 - Y_3 . The optimized formula was found out by setting maximum percentage drug release at 8 h. The values for bioadhesion force were set in the range of 0.8-1.2 N and the tensile strength greater than 14 kg/mm². Formulation F7 was emerged as optimized formulation with 50 mg drug, 100 mg IPC and 2 % glycerin.

CONCLUSION

In this study, novel buccoadhesive film was developed using Interpolymer complex between chitosan and *A. vera* gel mucilage. The film was releasing drug over a period of 8 h directly to systemic circulation through buccal mucosa. The extensive first pass metabolism of a drug was prevented to a great extent. Formulation chart was prepared by 3² level factorial design. The effect of formulation variables on bioadhesion force, drug release and tensile strength were studied and analyzed with the help of computer-based optimization method. After analyzing, all data and results formulation F7 designed based on the quadratic model was selected as optimal formulation.

Thus, an Interpolymer complex based mucoadhesive buccal films of Tramadol Hydrochloride was developed by optimization technique. The main objective of developing buccal films was to deliver tramadol hydrochloride to systemic circulation without any painful procedures. Interpolymer complex is more suitable for the preparation of buccal film as it exhibited good film forming ability and satisfactory bioadhesion force in comparison to chitosan alone. The study also shows that economical and widely available *A. vera* gel mucilage can be a promising excipient for systemic drug delivery of a water-soluble drug like tramadol hydrochloride via oral mucosal route. The *in vitro* dissolution studies confirmed that drug released at satisfactory rate from buccal films which is very much important for achieving therapeutic targets.

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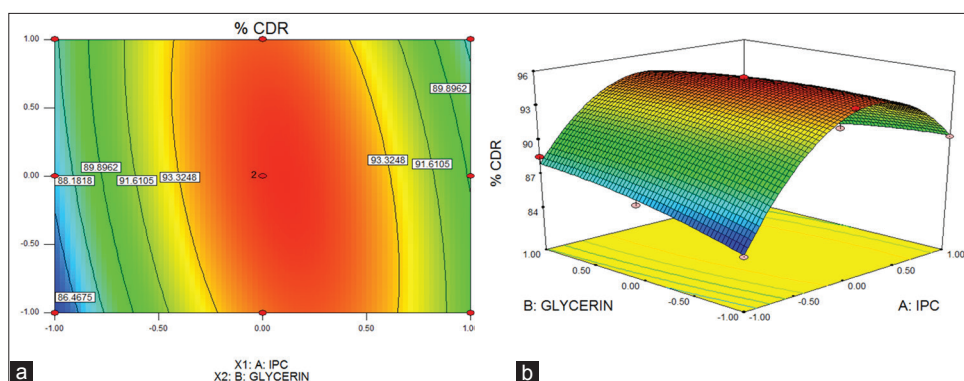


Figure 3: (a and b) Two-dimensional contour plot, three-dimensional response surface plots for % drug release

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