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Effect of Formulation Parameters on Enalapril Maleate Mucoadhesive Buccal Tablet Using Quality by Design (QbD) Approach

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Abstract The buccal route has great prospects and possible benefits for the administration of drugs systemically. The present study involves designing, developing and optimising the buccal tablet formulation of Enalapril Maleate (EM) by using the QbD approach. We prepared the EM buccal tablets using the dry granulation method. In the QTPP profile, the CQAs for EM buccal tablets are Mucoadhesive strength, swelling index and drug release (dependent variables); the CMAs identified for EM buccal tablets were Carbopol 934P, HPMC-K100M and chitosan (independent variables). Diluent quantity, blending time and compression force were selected as CPPs; the Box-Behnkentdesign was used to evaluate the relationship between the CMAs and CPPs. Based on the DoE, the composition of the optimised formulation of EM BT-18 consists of 20mg of EM, 15 mg of carbopol 934p, 17 mg of HPMC-K100M, 10mg of chitosan, 30 mg of PVP K-30, 1 mg of magnesium stearate, 16 mg of Mannitol, 1 mg of aspartame, and 50 mg of Ethyl cellulose. The optimised formulation of EM BT 18 was found to have a Mucoadhesive strength of 24.32±0.30g. The swelling index was 90.74±0.25% and drug release was sustained up to 10 hours 98.4±3.62% compared to the marketed product, whose release was up to 8 hours. We attempted to design a buccal tablet of Enalapril Maleate for sustained drug release in the treatment of hypertension. Patients who cannot take oral medication due to trauma or unconscious conditions could receive the formulation. Development of a newly P.ceutical product is very time-consuming, extremely costly and high-risk, with very little chance of a successful outcome. Hence, this study showed EM tablets are already available on the market but we have chosen a buccal drug delivery system using a novel approach using QbD tools to target the quality of the product accurately.

Keywords Enalapril Maleate, Mucoadhesive Buccal Tablet, Quality by Design (QbD), 3D Response, *Ex-vivo* permeation

INTRODUCTION

Millions of people worldwide suffer from hypertension, a chronic medical condition that significantly increases the risk of cardiovascular illnesses. Because of its high effectiveness and low

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toxicity, Enalapril Maleate, an ACE inhibitor, is a mainstay in the treatment of hypertension. Regular oral dose forms of Enalapril maleate, on the other hand, have problems like low bioavailability, changing plasma levels, and having to take doses often, which makes people not follow through with their treatment plans. Buccal drug delivery has become a viable substitute for traditional oral administration, including benefits such as prolonged release, reduced hepatic first-pass metabolism, and enhanced patient compliance. Additionally, Mucoadhesive buccal tablets provide controlled release and medication retention, which improves therapeutic results even more. Mucoadhesive buccal tablet development, despite its promise, needs a methodical approach to guarantee product quality, effectiveness, and patient safety. The systematic method of Quality by Design (QbD), which integrates quality management with pharmaceutical development, provides a structured framework for the design, formulation, and optimisation of drug delivery systems. Using a QbD methodology, this study intends to investigate the formulation, optimisation, and assessment of Mucoadhesive Enalapril maleate buccal tablets. This research aims to improve the quality of the final product and the knowledge of the formulation process by methodically examining crucial formulation factors and their influence on product performance (Shaikh R et al., 2011).

The aim of present study to develop Mucoadhesive buccal tablets of Enalapril maleate by using different mixture of polymers Carbopol-934P, HPMCK100M, Chitosan By using box behnken design for formulation table between dependent & independent variable. The research aims to provide a thorough methodology for the creation of buccal formulations. The first step in developing a formulation is choosing the right medications and polymers. We then conduct preformulation investigations to understand the physicochemical characteristics of the selected components. The research then attempts to establish the Quality Target Product Profile (QTPP) and identify critical quality attributes (CQAs) that are essential to the formulation's performance using the Quality by Design (QbD) methodology. The next step is to identify and optimise critical material attributes (CMAs) in order to guarantee raw material quality. Critical process parameters (CPPs) are identified and optimised in order to achieve further optimisation. A risk assessment is done in order to foresee and reduce any problems. After that, prototype formulations are created using Design of Experiment (DoE) tools, and optimisation is carried out in response to the results of the experiments. Numerous in vitro and ex vivo investigations, including drug release and permeability evaluations, as well as in vivo experiments in rabbits, are used to thoroughly evaluate the optimised formulation. Ultimately, in order to verify the resilience of the optimised formulation, stability experiments are carried out in accordance with ICH criteria (Tiwari G et al., 2012).

MATERIAL AND METHOD

Materials

Enalapril Maleate was procured from G.R. Scientific, Varanasi Uttar Pradesh, HPMC K 100M, Chitosan, Carbopol-934P, PVP-K30, Ethyl cellulose, Mannitol was purchased from G.R. Scientific, Varanasi Uttar Pradesh, and manufacturer are OXFORD Lab Fine Chem LLP, All reagents used were of analytical grade. The Mucoadhesive buccal tablet was prepared by direct compression method.

Risk assessment Matrix of critical material and process attributes

A risk assessment matrix was carried out to identify potential risks associated with material attributes and process parameters that could impact CQAs after the Quality Target Product Profile (QTPP) was defined and the Critical Quality Attributes (CQAs), Critical Material Attributes (CMAs), and Critical Process Parameters (CPPs) for buccal tablet formulation were determined. High, medium, and low risk categories were assigned based on the results of an extensive literature study (Vora and Shah, 2019). Table 1 presents the complete findings from the risk assessment matrix for buccal tablets (Vora R and Shah Y, 2019).

Optimization of Mucoadhesive Buccal Tablet

For the optimisation of experiments, the Box-Behnken statistical technique (Design Expert software version 13) was used. For buccal tablets, the primary variables influencing Mucoadhesive strength, swelling index, and drug release were Carbopol 934p (A), HPMC K100 (B), and chitosan (C). To look into the effects of the polymers, which were chosen for the study's high, medium, and low level based trials as independent variables. Buccal tablet formulations were made and their dependent variables, namely drug release, swelling index, and Mucoadhesive strength i.e. R1, R2, R3, were characterised Table- 2. Once we created the final formulation of the dependent and independent variables, we examined the specific factors and

responses. Table-3 displays the recommended outputs of 17 runs, which align with the DoE investigations.

Formulation of EM Mucoadhesive Buccal Tablet using Experimental design

In Table-4 listed the ingredients of the EM BT (Enalapril maleate Buccal Tablet), which had 20 mg of Enalapril maleate per tablet. Before being compressed directly, each component was screened using a No. 60 sieve. Using a tablet compression machine, an 8.0 mm flat-faced punch was used to compress the backing layer (EC). Enalapril maleate was manually combined with several ratios of polymers, such as Chitosan, Carbopol 934p, and HPMC K-100. The mixture was then mixed for ten minutes with the addition of PVP K30 (as a binder), Mannitol (as a diluent), and Aspartame (as a sweetening agent). After this blending period, three more minutes of mixing were spent adding the lubricant magnesium stearate. The final mixture was then crushed using the direct compression technique into tablets using an 8.0 mm flat-faced punch on a sixteen-station CEMACH rotary tablet-punching machine.

Table 1. Details of risk assessment matrix for EM Buccal Tablets

Drug Product CQAs	Risk Assessment Matrix					
	Critical Material Attributes (CMAs)			Critical Process Parameters (CPPs)		
	HPMC K100	Carbopol 934-P	Chitosan	Blending time	Lubricating time	Compression force
Mucoadhesive strength	High	High	High	High	Medium	Medium
Swelling index	High	High	High	Medium	Medium	Medium
Drug release	High	High	High	Medium	Medium	Medium

Table 2. Variables and their levels in Box-Behnken Design

Factors : Critical Formulation and Process		Levels	
Variables (Independent)	**-1**	**0**	**+1**
(A) Carbopol-934-P	5	7.5	10
(B) HPMC-K-100M	15	17.5	20
(C) Chitosan	0	7.5	15.00
Responses (Dependent)	**Goal**	**Acceptance criteria**	
(R1) Mucoadhesive strength	In Range	4.00-24.0g	
(R2) Swelling index	In Range	16.00-88.00%	
(R3) Drug release	In Range	95 - 99.5%	

*Design Expert 13 .0 software was used; No. of factors = 3, No. of levels = 3 Replicates = 0, No. of centre points = 5; Total number of runs=17runs

Table 3. Box-Behnken design for optimization of Mucoadhesive Buccal Tablet

Independent Variable	Run																
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
Carbopol-934-P	1	0	1	-1	-1	0	-1	0	0	0	0	0	1	0	1	0	-1
HPMC-K-100M	0	0	1	1	0	1	0	0	1	-1	-1	0	-1	0	0	0	-1
Chitosan	-1	0	0	0	-1	-1	1	0	1	-1	1	0	0	0	1	0	0

Characterization of EM Mucoadhesive Buccal Tablet

(Kadam PB et al.,2008; Lodhi M. et al.,2013)

FTIR Analysis of EM

Enalapril Maleate's molecular structure and functional groups may be found and examined using an FTIR analysis of the chemical. Through the measurement of the sample's absorption of infrared light at various wavelengths, Fourier transform infrared spectroscopy (FTIR) offers important insights into the chemistry and structure of Enalapril Maleate and identification, purity, and stability verification are crucial for maintaining the effectiveness and quality of pharmaceutical compositions that include this active component. Result are shown in table no.5 and figure no. 1.

Mucoadhesive strength measurement (Kadam PB et al.,2008)

We used a modified physical balancing approach to compute the Mucoadhesive force. Male pigs' freshly excised porcine buccal mucosa was used as the model substrate, and PB solution, which has a pH of 6.8, was used as the moistening agent. After the mucosal membrane was obtained, it was in contact with the EM BT for five minutes. Following this first time of contact, weights were placed on the right side of the pan with the intention of separating the tablet from the membrane. The weights were applied at a steady force of 100 milligrams for a continuous five-minute interval. The ex vivo Mucoadhesive strength was defined as the weight (in grams) needed to accomplish separation. The weight increase stopped at the same instant the pill separated from the porcine buccal mucosa, and this weight was recorded at that time. The temperature was kept at 37°C throughout the experiment. We used the following formula to get the Mucoadhesive force: Weight needed for separation (in grams) = Mucoadhesive force (in grams) This computation offers a numerical depiction of the tablet's Mucoadhesive potency.

$$N=W \times g/1000$$

Where, the Mucoadhesive force is represented by N, the acceleration caused by gravity is represented by g,

Table 4. Formulation Table of EM Mucoadhesive Buccal Tables

Ingredients (mg)	F1	F2	F3	F4	F5	F6	F7	F8	F9	F10	F11	F12	F13	F14	F15	F16	F17
Enalapril Maleate	20	20	20	20	20	20	20	20	20	20	20	20	20	20	20	20	20
Carbopol-934P	15	10	15	7.5	7.5	10	7.5	10	10	10	10	10	10	10	15	10	7.5
HPMC-K100M	15	15	20	15	15	20	15	15	20	10	10	15	10	15	15	15	10
Chitosan	5	7.5	7.5	7.5	5	5	10	7.5	10	5	10	7.5	7.5	7.5	10	7.5	7.5
PVP K-30	30	30	30	30	30	30	30	30	30	30	30	30	30	30	30	30	30
Magnesium Stearate	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1
Aspartame	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1
Ethyl Cellulose	50	50	50	50	50	50	50	50	50	50	50	50	50	50	50	50	50
Mannitol	23	25.5	15.5	28	30.5	23	25.5	25.5	18	33	28	25.5	30.5	25.5	18	25.5	33
Total weight	160	160	160	160	160	160	160	160	160	160	160	160	160	160	160	160	160

Table 5. Results of FTIR values of Pure drug (EM)

Characteristic peak	Standard range (cm^{-1})	EM peeks (cm^{-1})
N-H stretching amide	3500-3310	3210.02
C=O carboxylic acid stretching	1760-1720	1751.28
C=O amide stretching	1690-1630	1647.07
C=O Stretching ester	1750-1735	1726.81
COOH	3300-2500	3024.19

and the weight in gram needed to separate the tablet from the porcine buccal mucosa is indicated by W. result are shown in Table no, 6.

Swelling index study (Sunitha M et al., 2014)

In order to assess the swelling properties that are essential for the best adhesion and drug release in buccal tablets, each tablet was weighed separately before testing, and this initial weight was noted as W1. Then, to make sure the tablets were completely submerged, they were put on petri plates with 5 mL of PB solution (pH 6.8). Using forceps, the buccal tablets were gently removed from the petri dishes at regular intervals of 1, 2, 3, 5, 6, 7, and 8 hours. Blotting with Whatman filter paper helped to gently remove any extra PB solution that was around the pills. The tablets were reweighed after swelling as a result of the buffer solution's absorption, and this weight was noted as W2. Using the following formula, the extent of swelling was calculated and result are shown in Table no, 6.

Swelling degree =.(W2-.W1) / W1 *100

Where, W_1=Initial weight of Tablet (g) & W_2= Final weight of Tablet (g)

In-vitro drug release studies (Dash SK et al., 2013)

EM release from Buccal Tablets was examined using the USP II dissolving apparatus's rotating paddle method. The dissolving medium included 900 mL of PB pH 6.8 and was maintained at a constant temperature of 37±0.2°C at 25 rpm. The backing layer of the tablet was adhered to the glass slide-using adhesive. A glass slide was placed at the base of the vessel to enable unidirectional drug release from the buccal tablet. At predetermined intervals, two milliliter samples were removed, and the same amount of buffer was replaced. Following the appropriate dilution, the material was subjected to UV spectroscopic analysis and filtered using 2µm Whatman filter paper. result are shown in Table no, 6.

Optimization and validation of formula of EM BT-18

With the aid of Design Expert software version 13, the Box-Behnken design was optimized and the formulation was validated. To evaluate the optimal formulation, a regression model was built using the necessary values derived from three-dimensional graphs, as shown in Figures 4 to 6. In this procedure, coded values were used to reflect the in vitro drug release (Section 6.1.5.2.3), swelling index (Section 6.1.5.2.1), and Mucoadhesive strength (Section 6.1.5.2.1). In order to assess the precision of the anticipated results, a triple experiment was conducted using brand-new circumstances, enabling a comparison between the projected and observed values. The formula that follows was then used to get the percentage prediction error.

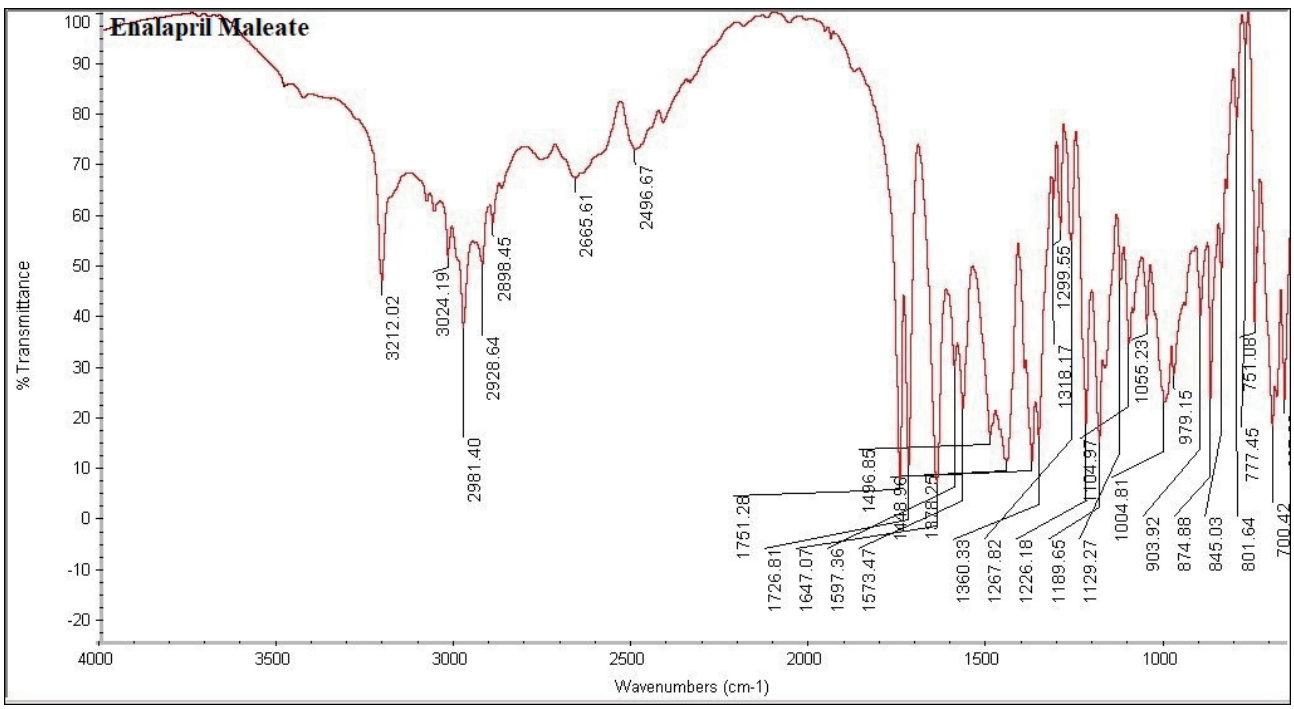

Figure 1. Results of FTIR of working standard

Table 6. Result of data obtained from experiment & DoE Study of EM BT (Batch: 1-17)

Run	Independent variable			Dependent Response		
	Factor-1 (A) Carbopol-934P	Factor-2 (B) HPMC-K 100M	Factor-3 (C) Chitosan	Response-1 Mucoadhesive Strength (g)	Response-2 Swelling index (%)	Response-3 Drug release (%)
1	1	0	-1	23.56	45.35	98.5
2	0	0	0	5.65	45.16	72.42
3	1	1	0	15.32	57.54	98.34
4	-1	1	0	9.63	86.97	99.3
5	-1	0	-1	22.62	65.43	96.12
6	0	1	-1	6.54	23.86	88.24
7	-1	0	1	21.23	25.24	95.62
8	0	0	0	2.89	51.32	66.58
9	0	1	1	6.73	88.56	73.25
10	0	-1	-1	6.86	38.65	81.29
11	0	-1	1	7.65	18.67	89.56
12	0	0	0	6.89	47.65	72.46
13	1	-1	0	13.62	52.37	98.42
14	0	0	0	6.58	49.08	80.2
15	1	0	1	22.65	81.24	92.54
16	0	0	0	6.74	52.65	76.54
17	-1	-1	0	7.94	16.35	97.15

Percentage prediction error (%PE) = [(Measured value−Predicted value) /Measured value] x 100

The acceptance criteria for the satisfactory prediction, the percentage PE should be less than 5 %. The results was shown in Tables 15.

Statistical analysis

To fit polynomial equations and establish the relevance of the independent variables, the collected data were examined using multiple regression analysis. We used analysis of variance (ANOVA) to check whether the regression model and its coefficients were statistically significant. In order to find the best formulation conditions and see the interaction effects, response surface plots were created. To ensure the precision and dependability of the model's predictions, the improved formulation underwent experimental validation.

Formulation of EM BT for the Optimized Batch EM-BT-18

As described in formulation section heading, Enalapril maleate was manually mixed with various ratios of polymers, such as Carbopol 934P, HPMC K-100M, and Chitosan. Then, for ten minutes, PVP K-30 (as a binder), mannitol (as a diluent), and aspartame (as a sweetener) were mixed together. After that, the mixture was stirred for an extra three minutes with lubricant magnesium stearate. A powder flow test was performed after this mixing procedure to assess the formulation's flow characteristics.

Ex-vivo permeation study of EM-BT-18 (Campisi G et al., 2010; Velmurugan S et al., 2010)

Drug absorption kinetics via biological membranes may be better understood with the use of ex vivo permeation studies. Most of the time, the physiological barrier and the drug molecules themselves dictate how a drug is transported over a membrane. Drug permeation tests were conducted on porcine buccal mucosa since it is the most structurally and chemically similar to pigs' freshly excised porcine buccal mucosa.

Table No. 17.

RESULTS AND DISCUSSION

Fourier Transform Infra-Red (FTIR) spectroscopy

EM was described using FT-IR; spectral data from the scanned sample at 4000-400 cm-1 was used to compare the EM working standard with the EM reference standard. Characteristic peaks of the EM that were identical to those of the working sample were found to include N-H stretching amide, C=O carboxylic acid stretching, C=O amide stretching, C=O ester stretching, and COOH stretching. EM showing that there were similarities between the working and reference standards of EM and the result were shown in table no. 5 and figure no. 1. (Shama Parveen et al., 2019 and Swamy et al., 2012)

Results of analytical methods for estimation of EM by UV (Thabet Y et al.,2018)

The spectrum and calibration curve of EM was prepared using methanol, the absorbance values were found in the range of 2-10 µg/ml. The standard spectrum and calibration curve of EM was shown in Fig. 2, 3. From the least square regression analysis, a linear response was obtained over a range of 2 to 10 µg/ml with a regression coefficient (R2) value of 0.9998. The spectra showed a sharp peak at 215 nm The best-fit linear equation obtained was y = 0.0991x + 0.004, y is the absorbance (AU) and x is the concentration of EM in µg/ml.

Results of EM-BT by QbD Approach Using DoE Tool (Wable AJ et al.,2013)

Mucoadhesive strength for EM BT 1-17 through 3D contour plot

From EM Mucoadhesive BT 1 -EM Mucoadhesive BT 17, the Mucoadhesive strength of the prepared BT from the DoE testing ranged from 5.65 to 23.56g. Using a modified physical balance, we measured the optimized formulation's Mucoadhesive strength by placing the tablet on top of the porcine buccal mucosa for 5 minutes without moving it, and then slowly adding 100 mg of weights to the right side of the pan to separate it. Maximum Mucoadhesive strength for the optimum formulation (EM-BT-1) was at 23.56 g. As seen in Fig No. 4 and table no. 6, the program created the 3D graphs. From the above 3D images of contour plots of Fig. 4(A) it was observed that, with increased in polymer ratio of HPMC K100M and Carbopol 934P from 7.5 to 20 mg the Mucoadhesive strength was also increased from 5.65 to 23.56 g as shown in Table 6. Similarly, Fig.4 (B) demonstrates that with increased in ratio of Chitosan and Carbopol 934p the Mucoadhesive strength were also increased but there is no much significant impact. The Fig.4 (C), shows there was no much significant impact on the polymer ratio between HPMC K100 and Chitosan. The final

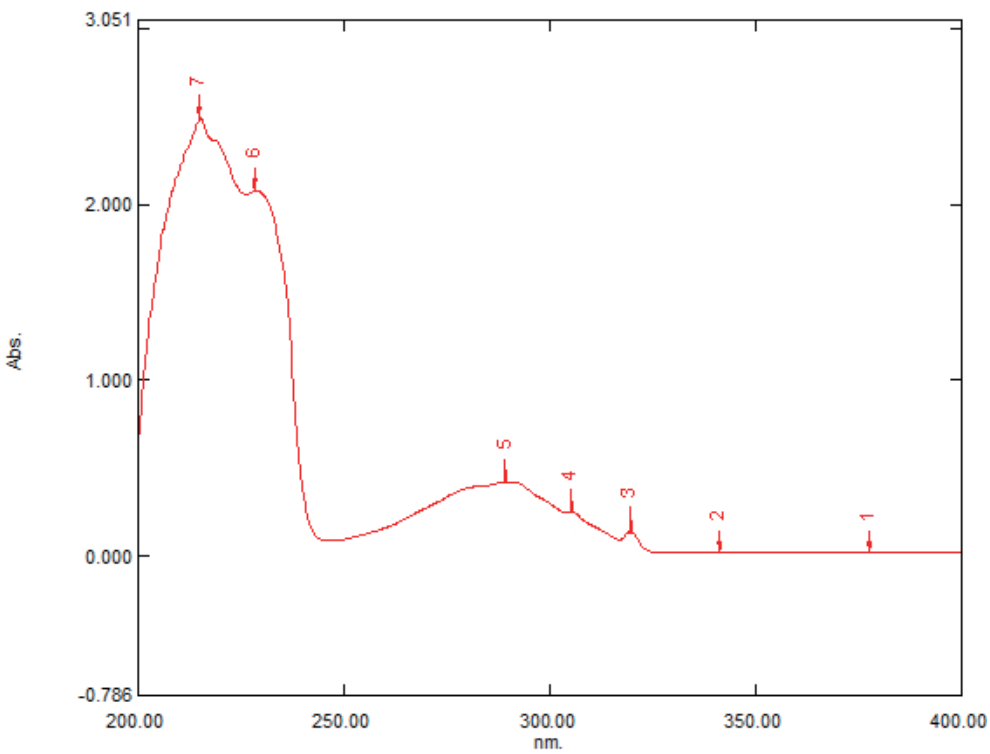

Figure 2. UV Spectrum scanning for EM

equation was generated in the coded form by software was given below.

$$R1=+5.75+1.72A++0.26B-0.16C+0.002AB+0.12AC-0.15BC+10.72A^2-4.85B^2+6.04$$

Result of swelling index for EM BT 1-17

Batch numbers EM BT 1–17 of the produced EM BT from the DoE studies exhibited a swelling index ranging from 17.24% to 88.56%. After weighing each buccal tablet in the submerged buffer solution for 1–8 hours, we used forceps to extract them from the petri dish and calculated the swelling index of the optimal formulation. With its maximal swelling property, the swelling index of the optimum formulation (EM BT 6) ranged 86.65%. Fig. 5 displays the software-generated 3D graphs. From the above 3D images of contour plots of Fig. 5 (A) it was observed that with increased in polymer ratio of HPMC K100M and Carbopol 934P the swelling index also increased 16.35 to 88.56 % but there was no significant impact, as shown in the Table no 6. similarly, Fig.5(B) demonstrates that increased in ratio of Chitosan and Carbopol 934P the Mucoadhesive strength also increased. In Fig.5(C), shows that as the polymer of ratio between HPMC K100 and Chitosan increases there was increased in swelling properties. The final equation was generated in the coded form by software was given below.

$$R2=+49.77+5.31A+16.36B+5.05C-16.36AB+19.02AC+21.17BC$$

Result of drug release for EM BT 1-17

For batch numbers EM BT 1–EM BT 17, the drug release ranged from 66.58 to 99.30% in the produced EM BT derived from the DoE experiments. The optimal

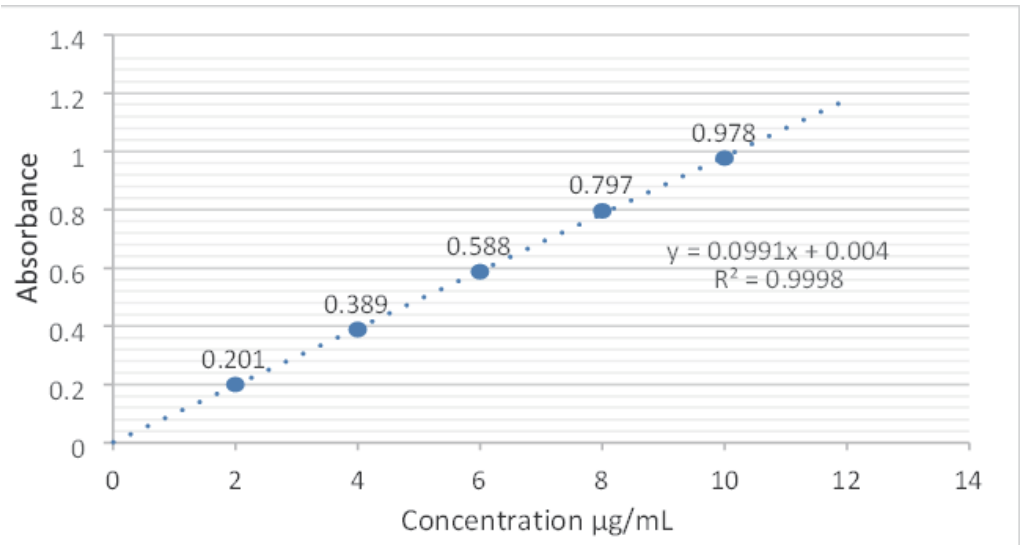

Figure 3. Results of standard graph of EM in Methanol

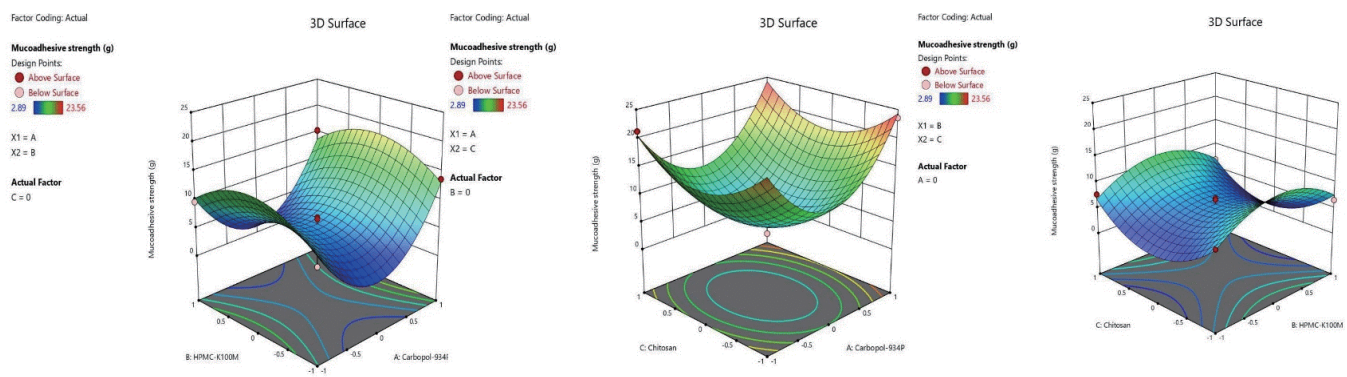

Figure 4. 3D response surface plot unveiling the simultaneous influence of independent variables on Mucoadhesive strength in Formulation

formulation's drug release was ascertained by use of a USP type II apparatus spinning paddle. The optimal formulation, EM-BT-4, had a maximum drug release of 99.30%. Fig.6. displays the 3D graphs produced by the program. From the above 3D images of contour plots Fig.6 (A) it was observed that with increased in polymer ratio of HPMC K100M and Carbopol 934p the drug release also increased from 67.86 to 99.5 % as shown in Table 6. Similarly, Fig.6 (B) demonstrates that with increased in ratio of Chitosan and Carbopol 934p the drug release increased. In Fig 6 (C), it shows that as the polymer ration between HPMC K100 and Chitosan there was no change in drug release. The final equation was generated in the coded form by software was given below.

R3=+73.64-0.04A-0.09B-1.65C-0.55AB-1.36AC-5.81BC+18.64A2+6.03B2+3.42C2

Statistical analysis Result of EM-BT

Table No. 7-9, display the results of the ANOVA analysis, which demonstrated that the developed linear model was highly significant (p value was < 0.05). Table no. 10-12 display the R2 values for the swelling index (0.8905), drug release (0.9414), and Mucoadhesive strength (0.9677). Additionally, low values of the coefficient of variation (CV) for the swelling index (at 18.48%), drug release (at 4.78%), and Mucoadhesive strength (16.75%) demonstrated a high degree of experimental accuracy. For the chosen variables, a signal-to-noise ratio (SNR) higher than 4 was ideal for navigating design space, which is what the appropriate precession measures. The tables no. 10-12 shows. Mucoadhesive strength, swelling index, drug release, and formulation variables: a connection study In order to evaluate the response's individual interaction, 3D graphs were created. Table no. 6 shows that the trials were conducted randomly with the

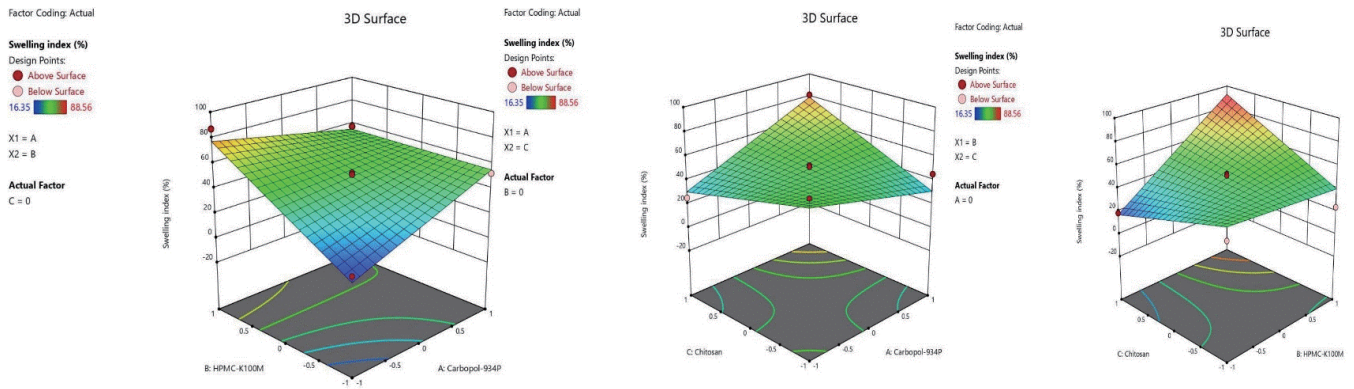

Figure 5. 3D response surface plot unveiling the simultaneous influence of independent variables on Swelling Index in Formulation

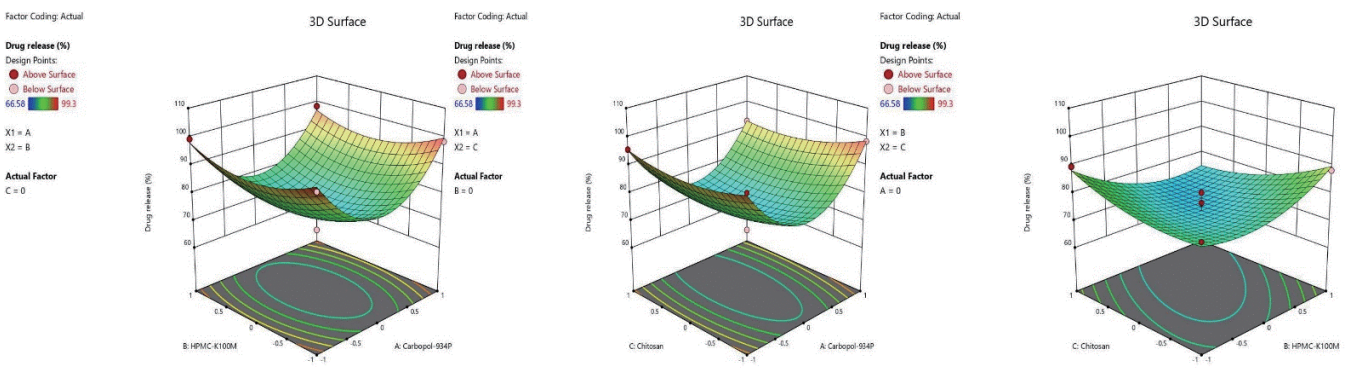

Figure 6. 3-D response surface plot unveiling the simultaneous influence of independent variables on Drug Release in Formulation

elimination of mistakes, and all the results fell within the range.

Response 1: Mucoadhesive strength Factor coding is coded, as Table No. 7 demonstrates. The sum of squares is partial (Type III). The model is deemed significant based on its F-value of 23.27. A significant F-value like this has a 0.02% probability of being caused by noise. Model terms are considered significant when P-values are less than 0.0500. A, A^2, B^2, and C^2 are important model terms in this instance. The model terms are not important if the value is bigger than 0.1000. Model reduction might make your model better if it has a large number of unimportant model terms (apart from those needed to maintain hierarchy). The 1.69 Lack of Fit F-value indicates that the Lack of Fit is not statistically significant in comparison to the pure error. A significant Lack of Fit F-value has a 30.50% probability of being caused by noise. It is desirable for there to be a non-significant lack of fit in the model.

Response 2: Swelling index Factor coding is coded, as Table No. 8 demonstrates. The sum of squares is partial (Type III). The model is deemed significant based on its F-value of 13.55. The probability that an

Table 7. Response 1: Mucoadhesive strength

Source	Sum of Squares	df	Mean Square	F-value	p-value	
Model	757.86	9	84.21	23.27	0.0002	significant
A-Carbopol-934P	23.56	1	23.56	6.51	0.0380	
B-HPMCK100M	0.5778	1	0.5778	0.1597	0.7014	
C-Chitosan	0.2178	1	0.2178	0.0602	0.8132	
AB	0.0000	1	0.0000	6.909E-06	0.9980	
AC	0.0576	1	0.0576	0.0159	0.9031	
BC	0.0900	1	0.0900	0.0249	0.8791	
A^2	484.21	1	484.21	133.81	< 0.0001	
B^2	98.89	1	98.89	27.33	0.0012	
C^2	153.67	1	153.67	42.47	0.0003	
Residual	25.33	7	3.62			
Lack of Fit	14.17	3	4.72	1.69	0.3050	not significant
Pure Error	11.16	4	2.79			
Cor Total	783.19	16				

Table 8. Response 2: Swelling index

Source	Sum of Squares	df	Mean Square	F-value	p-value	
Model	6882.28	6	1147.05	13.55	0.0003	significant
A-Carbopol-934P	225.89	1	225.89	2.67	0.1333	
B-HPMC-K100M	2141.52	1	2141.52	25.31	0.0005	
C-Chitosan	204.22	1	204.22	2.41	0.1514	
AB	1070.93	1	1070.93	12.66	0.0052	
AC	1447.04	1	1447.04	17.10	0.0020	
BC	1792.68	1	1792.68	21.18	0.0010	
Residual	846.22	10	84.62			
Lack of Fit	811.09	6	135.18	15.39	0.0098	significant
Pure Error	35.13	4	8.78			
Cor Total	7728.50	16				

F-value this great may be the result of noise is merely 0.03%. Model terms are considered significant when P-values are less than 0.0500. B, AB, AC, and BC are important model terms in this instance. The model terms are not important if the value is bigger than 0.1000. Model reduction might make your model better if it has a large number of unimportant model terms (apart from those needed to maintain hierarchy). The F-value of 15.39 for the lack of fit indicates that the lack of fit is substantial. A significant Lack of Fit F-value has a 0.98% probability of being caused by noise. We need the model to fit, thus a significant lack of fit is undesirable.

Response 3: Drug release Factor coding is coded, as Table No. 9 demonstrates.Square sums are Type III - Partial The model is deemed significant based on its F-value of 12.50. The likelihood of an F-value this big occurring as a result of noise is just 0.15%. Model terms are considered significant when P-values are less than 0.0500. In this instance, important model terms are BC, A2, and B2. The model terms are not important if the value is bigger than 0.1000. Model reduction might make your model better if it has a large number of unimportant model terms (apart from those needed to maintain hierarchy). Given the pure error, the Lack of Fit F-value of 0.21 suggests that the Lack of Fit is not statistically significant. A significant Lack of Fit F-value has an 88.42% probability of being caused by noise. Good—we want the model to fit—is a non-significant lack of fit.

Fit Statistics for Mucoadhesive strength: Taken from Table No. 10 The discrepancy between the predicted R2 of 0.6882 and the adjusted R2 of 0.9261 is more than 0.2, as one would often anticipate. This might point to a significant block effect or a potential issue with your data or model. Model reduction, response transformation, outliers, and other issues should be taken into account. Confirmation runs ought to be used for testing any empirical model. Adeq Precision calculates the ratio of signal to noise. Ideally, the ratio should be higher than 4. With a ratio of 12.698, you have a sufficient signal. The design area may be navigated with the help of this model.

Fit Statistics for swelling index: The Predicted R^2 of 0.4566 from Table No. 11 differs by more than 0.2 from the Adjusted R^2 of 0.8248, which is not as close as one would typically anticipate. This might point to a significant block effect or a potential issue with your data or model. Model reduction, response transformation, outliers, and other issues should be taken into account. Confirmation runs ought to be used for testing any empirical model. Adeq Precision calculates the ratio of signal to noise. Ideally, the ratio should be higher than 4. With a ratio of 13.658, you have a sufficient signal. The design area may be navigated with the help of this model.

Fit Statistics for drug release: Table 12 shows that the Adjusted R2 of 0.8661 and the Predicted R2 of 0.7931 are reasonably in agreement, meaning that the difference is less than 0.2. Adeq Precision

Table 9. Response 3: Drug release

Source	Sum of Squares	df	Mean Square	F-value	p-value	
Model	1939.13	9	215.46	12.50	0.0015	significant
A-Carbopol-934P	0.0190	1	0.0190	0.0011	0.9744	
B-HPMC-K100M	6.64	1	6.64	0.3855	0.5544	
C-Chitosan	21.71	1	21.71	1.26	0.2987	
AB	1.24	1	1.24	0.0721	0.7960	
AC	7.45	1	7.45	0.4325	0.5318	
BC	135.26	1	135.26	7.85	0.0265	
A^2	1462.36	1	1462.36	84.86	< 0.0001	
B^2	152.91	1	152.91	8.87	0.0206	
C^2	49.21	1	49.21	2.86	0.1349	
Residual	120.63	7	17.23			
Lack of Fit	16.47	3	5.49	0.2108	0.8842	not significant
Pure Error	104.17	4	26.04			
Cor Total	2059.76	16				

calculates the ratio of signal to noise. Ideally, the ratio should be higher than 4. With a ratio of 8.192, your signal strength is sufficient. The design area may be navigated with the help of this model.

Results of optimized formula by QbD for the preparation of the EM BT

Table No. 13 displays the statistical conclusions drawn from the model to demonstrate its best fit, which were based on a modified coded value that was produced when considering the optimal condition. You can see the improved formulation in Table No. 14 &15 show the findings of an additional triplicate experiment that was conducted under different experimental conditions to compare the anticipated results with the actual values of Mucoadhesive strength, swelling index, and drug release.

Results of optimized formulation of EM BT-18

The buccal tablets' ex vivo Mucoadhesive strength was assessed using modified physical balancing. The QbD technique yielded the best predicted formulation with a Mucoadhesive strength of 23.402g, whereas the observed response for EM BT 18 was 24.32±0.30g. Since the polymer slowly absorbs water as a result of its hydrophilicity, the swelling of the EM BT increases with time. As a result, the EM BT 18 formulation was shown to have strong swelling properties, just as predicted by QbD. At 8th hour, the observed swelling index response was 90.743 ± 0.65, whereas the expected response was 87.941%. Moreover, The USP II dissolving device was used to administer optimized EM BT-18 drug release in PB (pH 6.8), and the prediction error was computed. With a percentage error of ±3.28, which was less than 5% and within acceptable limits, the drug release was determined to be 98.60%, which was in good agreement with the predicted response of 90.649% using the QbD technique. Result are shown in table No. 15

Comparison of in vitro drug release of marketed formulation Vs optimized EM-BT-18

The publicly available product was dissolved in vitro and its results were compared with those of the optimized EM BT 18. Table No. 16 and Fig. No. 7 provide the comparative dissolution outcomes. While the EM BT 18 extended the drug release until the tenth hour, the marketed tablet released the medication entirely at the eighth hour.

Ex vivo permeation study of EM BT 18

The Franz diffusion cell was used to study the drug release of EM BT 18, and the results indicate that the drug penetration was gradual and constant. The drug release behavior is sustained by carbopol 934P, the designed buccal tablet's retention duration is affected by chitosan, and polymers like HPMC K100 may produce delayed degradation of drug release. According to Maroni A. et al. (2016), the ex vivo study's backing membrane causes the release to be unidirectional. The data shown in Table No. 17 demonstrate that the drug penetration from buccal tablets through the porcine buccal mucosa was slow and constant, releasing 99.12±0.17% of the medication in 8 hours at a flux of 0.065 ± 0.017 mg h^{-1} cm^{-2}. With a higher concentration of polymer comes a higher viscosity of the gel, which in turn could lead to a longer diffusion route. As a result, the drug's effective diffusion coefficient may drop, which in turn reduces the dissolving medium's ability to penetrate the tablet matrix and slows the drug's release rate.Product, process, and control knowledge are key components of the QbD strategy during optimization and formulation.

Table 10. Fit Statistics for Mucoadhesive strength

Std. Dev.	1.90	**R^2**	0.9677
Mean	11.36	**Adjusted R^2**	0.9261
C.V. %	16.75	**Predicted R^2**	0.6882
		Adeq Precision	12.6982

Table 11. Fit Statistics for swelling index

Std. Dev.	9.20	**R^2**	0.8905
Mean	49.77	**Adjusted R^2**	0.8248
C.V. %	18.48	**Predicted R^2**	0.4566
		Adeq Precision	13.6579

Table 12. Fit Statistics for drug release

Std. Dev.	4.15	**R^2**	0.9414
Mean	86.85	**Adjusted R^2**	0.8661
C.V. %	4.78	**Predicted R^2**	0.7931
		Adeq Precision	8.1919

According to Javed MN et al. (2018), the CMAs (HPMC K100, Carbopol 934p, and Chitosan) that were chosen for the buccal tablet release control are mostly based on quality risk management and solid scientific evidence. Figure No. 8 and Table No. 18 displayed the findings.

CONCLUSION

One strategy for treating hypertension using a sustained-release tablet that included both enalapril maleate was to create buccal tablets of each medicine. When administering medicine orally is not an option,

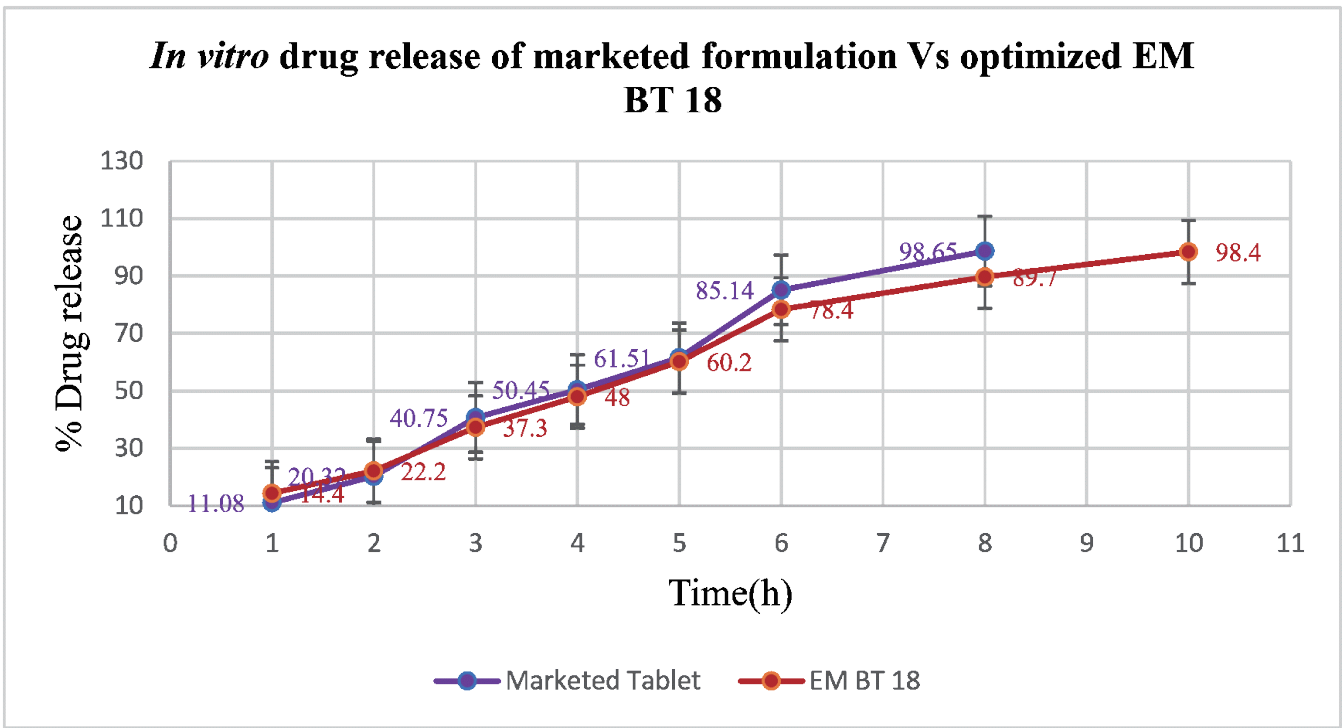

Figure 7. Comparative *in vitro* release of Optimized EM-BT-18 with the Marketed Product

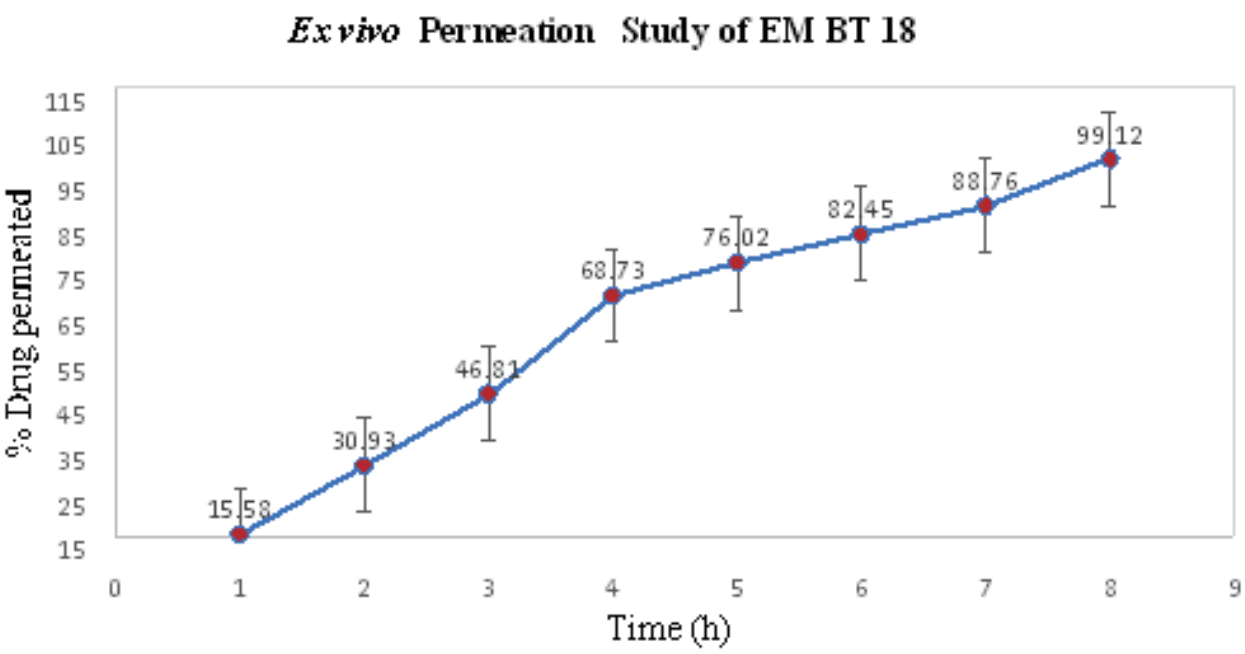

Figure 8. Results of *ex-vivo* permeation study of EM-BT-18

Table 13. Results of optimization parameter constraints for fixing the goal in QbD

Name	Goal	Lower Limit	Upper Limit	Lower Weight	Upper Weight	Importance
A:Carbopol-934P	Range	-1	1	1	1	3
B:HPMC-K100M	Range	-1	1	1	1	3
C:Chitosan	Range	-1	1	1	1	3
Mucoadhesive strength	Maximize	4	24	1	1	3
Swelling index	Maximize	16	88	1	1	3
Drug release	Maximize	95	99.3	1	1	3

Table 14. Result of predicted Composition of optimized formulation by QbD Batch EM BT-18

Carbopol-934P (mg)	HPMC-K100M (mg)	Chitosan (mg)	Mucoadhesive strength (g)	Swelling index (%)	Drug release (%)	Desirability
15	17	10	23.402	87.941	90.649	0.898

Table 15. Results of optimized formulation of EM BT-18

Variables	Predicted response	Observed response	% Predicted error (% PE)	Acceptance criteria for % PE
Mucoadhesive strength (g)	23.402	24.32	±0.30	Less than 5.0 %
Swelling index (%)	87.941	90.743	±0.65	Less than 5.0 %
Drug release (%)	90.649	98.40	±3.28	Less than 5.0 %

Table 16. *in vitro* drug release of marketed formulation Vs optimized EM-BT

Formulation	Time (hr)							
	1	2	3	4	5	6	8	10
Marketed Tablet	11.08± 1.14	20.32± 1.87	40.75± 2.34	50.45± 2.33	61.51± 4.14	85.14± 2.88	98.65± 4.56	---
EM-BT-18	14.4±2.22	22.2±1.89	37.3±2.19	48.0±3.48	60.2±4.17	78.4±2.44	89.7±3.54	98.4±3.62

Table 17. Results of *Ex vivo* permeation study of EMBT-18

Time (hr)	1	2	3	4	5	6	7	8
Drug permeated (%)	15.58± 0.10	30.93± 0.24	46.81± 0.25	68.73± 0.47	76.02± 0.11	82.45± 0.22	88.76± 0.26	99.12± 0.17

Table 18. Results Flux and Permeability coefficient of EM-BT-18

Formulation	Flux (mg/cm^2/hr)	Permeability coefficient
EM BT-18	0.065 ± 0.017	0.02687 ± 0.003

such as in cases of trauma or unconsciousness, the formulation may be used. There is a low probability of success in the lengthy, expensive, and risky process of developing a new pharmaceutical product. Therefore, a new method using QbD tools to precisely target the high-quality product allowed the research to choose the already-marketed medicinal products EM as the buccal drug delivery system.

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Conflict of interest

There are no conflicting interests, as the authors have stated.

Abbreviations

EM =Enalapril maleate
ANOVA= Analysis of variance
BBD= Box-Behnken Design
CMAs =Critical Material Attributes
CPPs= Critical Process Parameters
CV =Coefficient of variation
DDS =Drug Delivery System
DoE= Design of Experiments
FD= Factorial Design
FTIR =Fourier Transform Infrared Spectroscopy
IP =Indian Pharmacopoeia
QbD= Quality by Design
QTPP= Quality Target Product Profile
R2 =correlation coefficient
RSM= Response Surface Methodology
SD= Standard Deviation
USP= United States of Pharmacopeia
UV= Ultraviolet Spectroscopy

References

1. Campisi G, Giannola LI, Florena AM, De Caro V, Schumacher A, Göttsche T, Paderni C, Wolff A. Bioavailability in vivo of naltrexone following transbuccal administration by an electronically-controlled intraoral device: a trial on pigs. Journal of controlled release. 2010 Aug 3;145(3):214-220.
2. Dash SK, Khan AS, Thakur R, Padhan A, Gupta DK, Behera BC. Formulation And In-Vitro Characterization of Floating Tablet of Enalapril Maleate. International Journal of Pharmaceutical Sciences and Research. 2013 Jan 1;4(1):486-496.
3. Javed MN, Kohli K, Amin S. Risk assessment integrated QbD approach for development of optimized bicontinuous mucoadhesive limicubes for oral delivery of rosuvastatin. AAPS PharmSciTech. 2018 Apr;19(3):1377-1391.
4. Kadam PB, Dias RJ, Mali KK, Havaldar VD, Mahajan NS. Formulation and evaluation of buccoadhesive tablets of atenolol. J Pharm Res. 2008 Oct;1(2):193-199.
5. Lodhi M, Dubey A, Narayan R, Prabhu P, Priya S. Formulation and evaluation of buccal film of Ivabradine hydrochloride for the treatment of stable angina pectoris. International journal of pharmaceutical investigation. 2013 Jan;3(1):47-53.
6. Maroni A, Zema L, Cerea M, Foppoli A, Palugan L, Gazzaniga A. Erodible drug delivery systems for time-controlled release into the gastrointestinal tract. Journal of Drug Delivery Science and Technology. 2016 Apr 1;32:229-235.
7. Shaikh R, Raj Singh TR, Garland MJ, Woolfson AD, Donnelly RF. Mucoadhesive drug delivery systems. J Pharm Bioallied Sci. 2011 Jan;3(1):89-100.
8. Shama P, Jai NM, Dhaneshwar KV, Navneet KV. Formulation And Development of Enalapril Maleate Fast Dissolving Tablets by Direct Compression Method and Its Pharmaceutical Evaluation. WJPLS. 2019 5(1),63-70.
9. Sunitha M, Padma A, Balaji B, Krishna VR. Formulation and in vitro evaluation of buccal mucoadhesive tablets of Chloropheneramine Maleate. International Journal of Pharmaceutical, Chemical & Biological Sciences. 2014 Jul 1;4(3):457-462.
10. Swamy GK, Kumar JR, Rao JS. development and validation of stability indicating rp-hplc method for simultaneous determination of enalapril maleate and felodipine in bulk and tablet dosage form,2015, World Journal of Pharmaceutical Research 4,(4): 2087-2100.
11. Thabet Y, Lunter D, Breitkreutz J. Continuous inkjet printing of enalapril maleate onto orodispersible film formulations. International journal of pharmaceutics. 2018 Jul 30;546(1-2):180-187.
12. Tiwari G, Tiwari R, Sriwastawa B, Bhati L, Pandey S, Pandey P, Bannerjee SK. Drug delivery systems: An updated review. International journal of pharmaceutical investigation. 2012 Jan; 2(1):2-11.
13. Velmurugan S, Raghavarapu KK. Formulation and in vitro evaluation of glipizide mucoadhesive buccal tablets. Int J Pharm Bio Sci. 2013;4(2):594-607.
14. Vora R, Shah Y. Investigation of Quality Target Process Parameters (QTPP) And Critical Material Attributes (CMA) of Nanocellulose as A Potential Excipient. International Journal of Applied Pharmaceutics. 2019 Jul 7:386-395.
15. Wable AJ, Thokal SB, Mittal SS, Shirsat AE, Bhingare CL, Ingle PL. Effect of formulation parameters on Sumatriptan succinate Buccal Mucoadhesive Tablet: Quality by Design approach. Journal of Applied Pharmaceutical Science, 2013 May 4(4):103-111.