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Design, development and *in vitro* evaluation of sertaconazole mucoadhesive vaginal tablet

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ABSTRACT

In present study, mucoadhesive vaginal tablet of sertaconazole was designed using a combination of mucoadhesive polymers like Carbopol 934P, chitosan, carboxymethylcellulose sodium, sodium alginate, methyl cellulose, hydroxypropyl methylcellulose and hydroxypropyl cellulose. Effervescent was incorporated into formulations to enhance swellabilty of mucoadhesive tablet. For various drug-free formulations, the effect of effervescent on polymers swelling characteristics was investigated. Swelling, mucoadhesive property and drug release study of the tablets with different proportions of mucoadhesive polymer and effervescent in formulations were conducted. A good sustained effect and moderate bioadhesion were obtained with the tablets containing 100 mg of effervescent, with Carbopol 934P: HPMC K4M ratio of 1:1 seemed to be the optimum one for the tablet. From the ex vivo retention study it was found that the mucoadhesive polymers hold the tablet for more than 24 hours inside the vaginal tube. Our study may provide a potential vaginal tablet formulation of sertaconazole against Candida albicans.

Keywords: Mucoadhesion, Mucoadhesive polymers, Effervescent mixture, Vaginal candidiasis.

INTRODUCTION

Vaginal candidiasis is a common condition and up to 75% of all women suffer at least one episode of this infection during their lifetime. *Candida albicans* is the most important cause of vaginal candidiasis, accounting for over 80% of the infection. Most patients with *Candida vaginitis* respond to topical treatment with nystatin or imidazole [1]. Sertaconazole is an imidazole derivative antifungal agent developed for treatment of human mycotic infections and plays an essential role in antifungal chemotherapy [2]. It is lipophilic with limited water solubility except at low pH [3]. For treatment of vulvovaginal candidiasis local antifungal has been favored due to numerous side effects, toxicity and teratogenic potential of systemically applied drug. Sertaconazole generally given by oral route but one of limitation of conventional dosage form in vaginal therapy is the relatively short residence time of drug at site of application. To achieve desirable therapeutic effect vaginal delivery system for sertaconazole need to reside at the site of infection for prolong period. Hence there is need to develop effective drug delivery system that should prolong the contact of drug with vaginal mucosal surface.

Traditional vaginal drug delivery systems include solutions, suspensions, gels, foams and tablets [4]. Vaginal creams and gels provide lubrication, but tend to be messy, and are easily removed if they are water soluble. Suspensions and solutions tend to spread unevenly in the vagina. Foam producing dosage forms are preferred as excessive lubrication and leakage from the vagina are minimal and the foam adheres to the vaginal walls. Thus vaginal tablets appear to be useful dosage forms as they are easy to apply, portable and the user knows how many units remain [5, 6].

Therefore present work was aimed to develop effervescent mucoadhesive tablet of sertaconazole capable to efficiently deliver drug during extended period of time against *C. albicans*. Effervescent added into the formulations

to enhance swellabilty mucoadhesive tablet. During the in vitro study, Carbopol 934P, chitosan, carboxymethylcellulose sodium (sodium CMC), sodium alginate, methyl cellulose (MC), hydroxypropyl methylcellulose (HPMC) and hydroxypropyl cellulose (HPC) were chosen as mucoadhesive polymers. The performances of these mucoadhesive polymers were evaluated by two parameters, the swelling behavior and the mucoadhesive strength. For the various drug-free formulations, the effect of effervescent on polymers' mucoadhesive characteristics was investigated. On the basis of these data, suitable polymers were selected to prepare the mucoadhesive effervescent vaginal tablets of sertaconazole. Swellings, mucoadhesive properties and drug release of the tablets with different proportions of mucoadhesive polymer and effervescent in formulations were conducted. One ideal formulation was selected for the subsequent ex vivo mucoadhesion time of the tablet and in vitro antifungal study.

MATERIALS AND METHODS

Sertaconazole was kindly gifted by Cipla Ltd. (Mumbai, India) Carbopol 934P was kindly gifted by Corel pharmaceutical Ltd. (Ahmedabad, India), Carbopol 934P was obtained as gift sample from Corel Pharmaceuticals Ltd. (Ahmedabad, India) , carboxymethylcellulose sodium (sodium CMC), hydroxypropyl methylcellulose (HPMC) and hydroxypropyl cellulose (HPC) were received a gift sample from Zydus-Cadila Healthcare Ltd. (Ahmedabad, India), were used as received. Sodium alginate and methyl cellulose (MC) were purchased from S.D. Fine Chemicals Ltd. (Mumbai, India); all other chemicals were of analytical reagent grade. Candid®-V3 tablet and Candid®-V gel were purchased from local market.

Preliminary Screening of polymer

Preliminary trial batches of sertaconazole mucoadhesive vaginal tablet with or without effervescent mixture were prepared using different polymers like Carbopol 934P, chitosan, carboxymethylcellulose sodium, sodium alginate, methyl cellulose, hydroxypropyl methylcellulose and hydroxypropyl cellulose by direct compression technique. The required quantity of polymers, sertaconazole, effervescent (consisting of sodium bicarbonate and citric acid at the mole ratio of 3:1), MCC, 1% magnesium stearate and 2% talc and finally the mixture was compressed to tablets using Cadmack single punch tablet compression machine. Talc and magnesium stearate were added as glidant and lubricant respectively. Each tablet contained 500mg of sertaconazole and has an approximate weight of 1000mg. Swelling study, mucoadhesion study and dissolution study of preliminary trial batches has been performed and suitable polymer combination HPMC K4M and Carbopol 934P has been selected for formulation of factorial design batches from the results of swelling and mucoadhesion study.

Optimization using 3^2 full factorial design [7]

A statistical model incorporating interactive and polynomial term was used to evaluate the response:

$$Y = b_0 + b_1X_1 + b_2X_2 + b_{12}X_1X_2 + b_{11}X_1^2 + b_{22}X_2 \qquad (1)$$

Where, Y is the dependent variables, b_0 is the arithmetic mean response of the nine runs, and b_1 is the estimated coefficient for the factor X_1. 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 simultaneously changed. The polynomial terms (X_1^2 and X_2^2) are included to investigate non-linearity.

A 3^2 full factorial design was adapted to optimize the variables. In this design 2 factors were evaluated, each at 3 levels, and experimental trials were performed at all 9 possible combinations. Polymer ratio (X_1) and amount of effervescent (X_2) were selected as independent variables. Mucoadhesive strength, % swelling, and Q_8 were selected as dependent variables (response; Y). The preparation and evaluation method for tablets and amount of sertaconazole were kept constant for all the trials. The full factorial design lay out , coded values for polymer ratio(X_1) and amount of effervescent mixture(X_2), and composition of factorial batches A1 to A9 is shown in Table: 1 and Table: 2 respectively.

Table: 1 Full factorial design layout

Coded value	HPMC K4M: Carbopol 934P(X_1)	Amount of effervescent(X_2)
-1	1:0.5	50
0	1:1	75
1	1:1.5	100
Check point	1:0.7	92.5

Table: 2 Composition of 3^2 full factorial design batches

Ingredients(mg)	A1	A2	A3	A4	A5	A6	A7	A8	A9
Sertaconazole	500	500	500	500	500	500	500	500	500
HPMC K4M	75	75	75	75	75	75	75	75	75
Carbopol 934P	37.5	37.5	37.5	75	75	75	112.5	112.5	112.5
MCC	315	280	265	277.5	242.5	227.5	240	205	190
Sod. Bicarbonate	37.5	66.3	75	37.5	66.3	75	37.5	66.3	75
Citric acid	12.5	18.7	25	12.5	18.7	25	12.5	18.7	25
Mg. stearate	7.5	7.5	7.5	7.5	7.5	7.5	7.5	7.5	7.5
Talc	15	15	15	15	15	15	15	15	15

Physicochemical characterization of sertaconazole mucoadhesive vaginal tablet

The Uniformity of weights of all vaginal tablets determined by using Electronic balance Sartorius (Model BT- 224 S) [8]. The hardness of the vaginal tablets was determined by hardness tester (Model no 1101, Shivani Scientific Ind). Vaginal tablet thickness was measured by placing tablet between two arms of the Vanier calipers [9].

Swelling study

The swelling behavior of tablet described as the water absorbing capacity. Prepared vaginal tablets were weighed individually (W_0) and placed separately in 2% agar gel plates and incubated at 37 ± 1°C. At regular 0.5 hour time intervals unto 4 hours, the tablet was removed from the Petri dish and excess surface water was removed carefully using filter paper. The swollen tablet was then reweighed (Wt) and the % swelling were calculated using the following formula:

$$\% \text{ Swelling} = \{(Wt - W_0)/ W_0\} \times 100 \qquad (2)$$

Where *Wt* is the weight of the tablet at time *t* and W_0 is the initial weight of tablet. The swelling was calculated and then plotted as a function of time. The slope of the linear plots was taken as the swelling rate [5, 10].

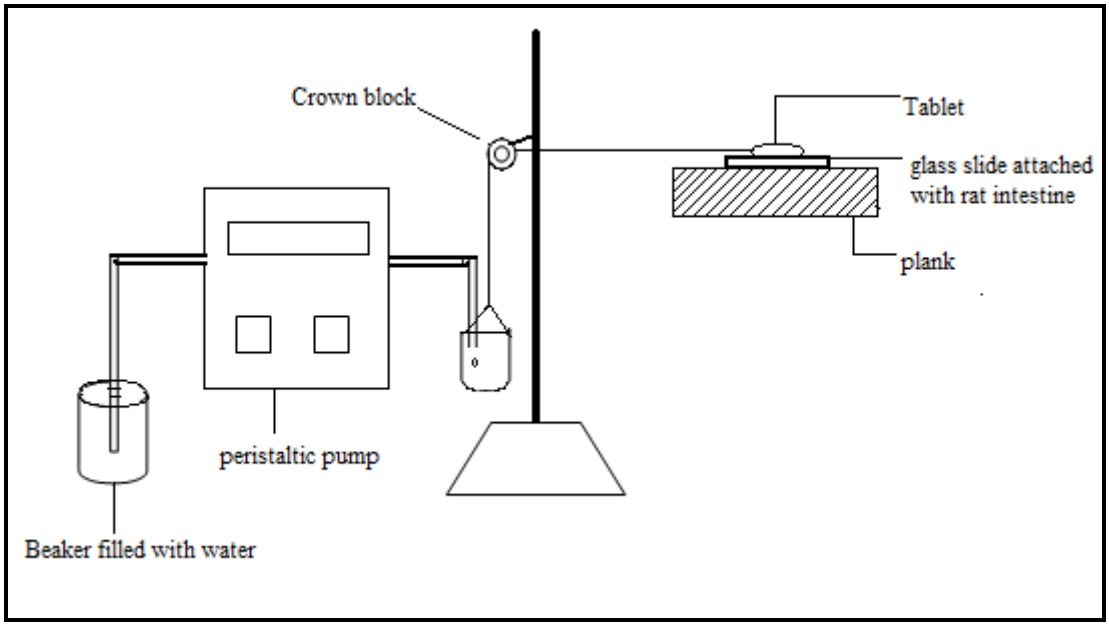

Fig: 1 The scheme of the device used in the mucoadhesion studies

In vitro mucoadhesion study

Several types of mucosa, including rat intestine, pig oral, bovine sublingual, cow vaginal mucosa [11, 12], have been used as model biological tissues for the evaluation of bioadhesion, which. In this study, rat intestine was preferred. A simple apparatus was devised to measure the minimum detachment force (Fig. 1). A piece of rat intestine (2.0 cm×1.0 cm) removed from newly sacrificed rat was adhered to a piece of glass, which was fixed on a plank and the plank was assembled with a little crown block. After hydrating the rat intestine with distilled water, the tablet was brought into contact with the rat intestine by applying little force for minute. After the initial contact, the tablet was encircled by a thread which fastened a light plastic beaker through the crown block. Next, water was dropped into the beaker at a speed of 3.0 ml·min-1 using peristaltic pump until the tablet and rat intestine were pulled apart by the gravity of water. The beaker containing water was weighed and the minimum detachment force was calculated

accordingly. The experiments were performed in triplicate and average values with standard deviation (SD) were reported. The study was approved by Nootan Pharmacy College, Institutional Animal ethics Committee.

Ex-vivo residence time

The ex-vivo residence was determined using a locally modified USP paddle apparatus (dissolution test apparatus type I). Residence time was examined after application of the vaginal tablet on freshly cut rat intestine. The fresh rat intestine was tied on the glass slide, and a mucoadhesive tablet was wetted with 1 drop of phosphate buffer and pasted to the rat intestine by applying a light force with a fingertip for 30 seconds. The glass slide was then tied on paddle of dissolution apparatus, put in dissolution bowl, which was filled with 250 mL of the phosphate buffer and kept at 37 ± 1°C. After 2 minutes, a slow stirring rate was applied to simulate the vaginal cavity environment, and tablet adhesion was monitored for 24 hours. The time for the tablet to detach from the rat intestine was recorded as the mucoadhesion time [13].

In Vitro sertaconazole release study

The release rate of sertaconazole mucoadhesive vaginal tablet (n=3) was determined using *The United States Pharmacopoeia* (USP) XXIV dissolution testing apparatus I (basket method) in 500ml of phosphate buffer pH 4.0 as the dissolution medium.. The tablet was placed in a settling basket to prevent the tablet from floating [14]. The rate of stirring was 30 rpm. And the medium temperature was maintained at 37 ± 0.5 ºC. A sample (10 ml) of the solution was withdrawn from the dissolution apparatus hourly and the samples were replaced with fresh dissolution medium. The samples were filtered through a filter and diluted to a suitable concentration with phosphate buffer pH 4.0. Absorbance of these solutions was measured at 260 nm using a Shimadzu UV-1800 UV/Vis double beam spectrophotometer. Cumulative percentage of drug release was calculated using the equation obtained from a standard curve.

Statistical analysis

Tests for significant differences between means were performed by Student's *t*-test or one-way ANOVA by using the software. Differences were considered significant at $P < 0.05$ level.

Kinetic modeling of drug release

To analyze the mechanism of drug release from the vaginal tablet, the dissolution data were fitted to the following equations. Kinetic modeling was preformed using Microsoft excel 2007 version. The dissolution data were fitted to the following equation.

$$Mt/M\infty = Kt^n \qquad (3)$$

Where $Mt/M\infty$ is the fraction of drug released at time *t*, *k* is the kinetic constant of the system, and *n* is the release exponent indicating the type of drug release mechanism. The release exponent takes various values depending upon different geometries. For the drug release from a cylindrical or a flat swellable polymer, if *n* approaches to 0.89, the release mechanism could be Case-II transport and if *n* is close to 0.45, the release mechanism can be Fickian. On the other hand if $0.45 < n < 0.89$, non-Fickian transport could be obtained [15, 16, 17].

In vitro antifungal studies

In vitro antifungal studies were performed against *Candida albicans* in Sabouraud's agar medium by the cup plate method. The cups cut in the inoculated solidified media were filled with different formulations using sterilized syringes. The marketed tablet (Candid®-V_3 tablet) was crushed into a powder and dissolved in 2 mL of sterilized water in a sterilized syringe. The marketed gel (Candid®-V gel) was applied using the sterilized syringe. The developed sertaconazole mucoadhesive vaginal tablet was swelled in 2 mL of sterile water applied into the cups. The covered Petri plates were incubated at 22°C in the BOD incubator for 48 hours. The zone of inhibition was measured at the end of 48 hours [18].

RESULTS AND DISCUSSION

Differential scanning calorimetry enables the quantitative detection of all processes in which energy is required or produced. In the physical mixture of drug with excipients of vaginal formulation the endothermic peak of sertaconazole was appeared at 154.21 ºC (Fig: 2) almost very near to that of pure drug 157.67 ºC (Fig: 3). This confirmed the physicochemical stability of drug with the formulation excipients used in the study.

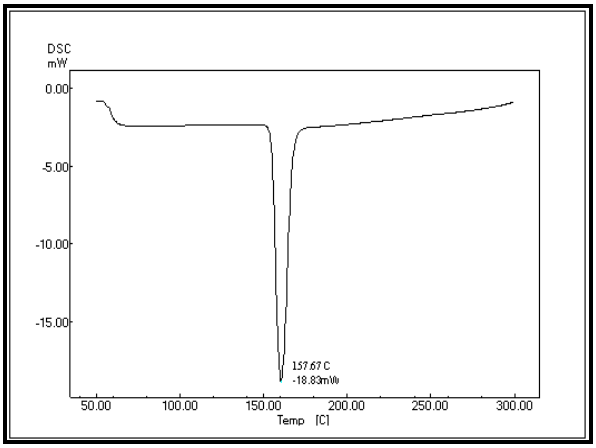

Fig: 2 DSC study of sertaconazole

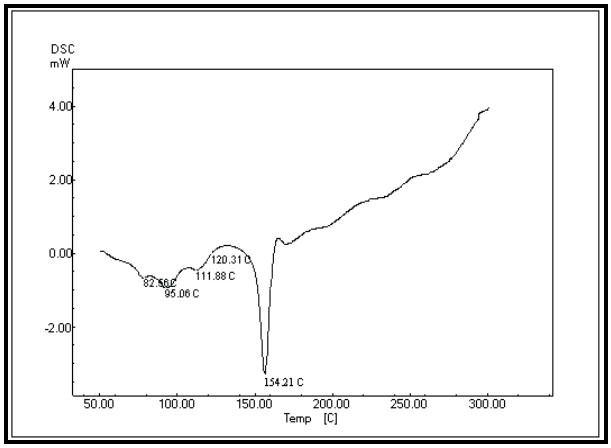

Fig: 3 DSC study of physical mixture of sertaconazole with formulation excipients

Preliminary screening of polymer

Swelling study of preliminary trial batches

Swelling is important for the assessment of adhesion. Shortly after swelling, adhesion does occur, but with a weak bond formed. To develop maximum adhesion strength, an optimum water concentration was needed for polymer particles [14]. It was observed that the order of swelling rate were Carbopol 934P > Chitosan > Sod.CMC > HPMC K4M in preliminary trial batches. According to the comparison of the corresponding swelling profiles of formulations with or without effervescent, it could be seen that the effervescent resulted in a marked increase in swelling rate. Furthermore, most tablets with 100 mg effervescent showed a higher swelling capacity than tablets without effervescent. The phenomenon of swelling increasing could be explained by the good disintegration effect of effervescent, which made tablets increase in volume and construct porous channels on surface and inside of tablets. The porous channels increased the area of contacting between polymer particles and water so that the polymers could be hydrated more easily.

In vitro mucoadhesion study of preliminary trial batches

Mucoadhesive strength of preliminary trial batches was determined using self developed force detachment method and observed within the range of 0.160 to 0.385 N. Based on results obtained from swelling study, mucoadhesion study and dissolution study formulation containing Carbopol 934P and HPMC K4M was considered a good candidate for development of a sertaconazole mucoadhesive vaginal tablet because of its good drug release and moderate mucoadhesion. As observed Carbopol 934P having better swelling property than HPMC K4M so Carbopol 934P was used to achieve suitable mucoadhesion and drug release because more mucoadhesion strength leads to local irritation in vagina.

Optimization of formulations using 3^2 full factorial design

The number of experiments required for these studies is dependent on the number of independent variables selected. The response (Yi) is measured for each trial.

In order to investigate factors systematically, a factorial design was employed in the present investigation. On the basis of the preliminary trials a 3^2 full factorial design was employed to study the effect of independent variables i.e. Polymer ratio (X_1) and amount of effervescent (X_2) on dependent variables mucoadhesive strength, % swelling, and Q_8.

Physicochemical characterization of factorial design batches

The tablet mould was especially designed (capsule shape) using stainless steel, and the formulated tablets had an average weight of 998.67±7.02mg, 20.8mm length, 8.5mm width and 6.17±0.057mm thickness. Hardness of the prepared tablets was observed within the range of 4.5-6 kg/cm^2. Off-white, capsule shaped mucoadhesive tablets were compressed using Cadmack single punch tablet compression machine.

Swelling study of factorial design batches

Comparing with drug-free tablets employing the same amount of polymer and effervescent, all sertaconazole mucoadhesive vaginal tablets showed lower swelling rates, which is related with the poor solubility of sertaconazole. % swelling of all the nine batches shown in Table: 3.

In vitro mucoadhesion study

In general, the swelling state of polymer contributes to its mucoadhesive behavior. It was observed that the swelling rate was developed as effervescent applied to formulation, increased with increasing amount of effervescent; however, the effervescent led to a significant drop in adhesive strength. The influences of effervescent on swelling and mucoadhesion were opposite, mainly due to the tiny bubbles created by effervescent. These tiny bubbles depressed the mucosa-polymer interaction, resulting in a decrease in the mucoadhesive strength [19].

The minimum adhesion strength (0.111N) was observed in formulation A3, which could be due to the lower ratio of HPMC K4M: Carbopol 934P and the higher content of effervescent. On the contrary, with an increase in HPMC K4M: Carbopol 934P ratio and reduction of effervescent, the maximum adhesion strength (0.275N) was obtained for formulation A7. (Table: 3)

Ex-vivo mucoadhesion time

The time for the tablet to detach from the rat intestine was recorded as the mucoadhesion time. The increase in concentration of polymer blend in series from formulation A1 to A9, showed a gradual rise in mucoadhesion time. The factorial batches A1 to A9 showed mucoadhesion time more than 10 hours.

In vitro drug release study of factorial design batches

The release rate of sertaconazole from effervescent mucoadhesive vaginal tablet was described as a function of time as shown in Fig: 4.

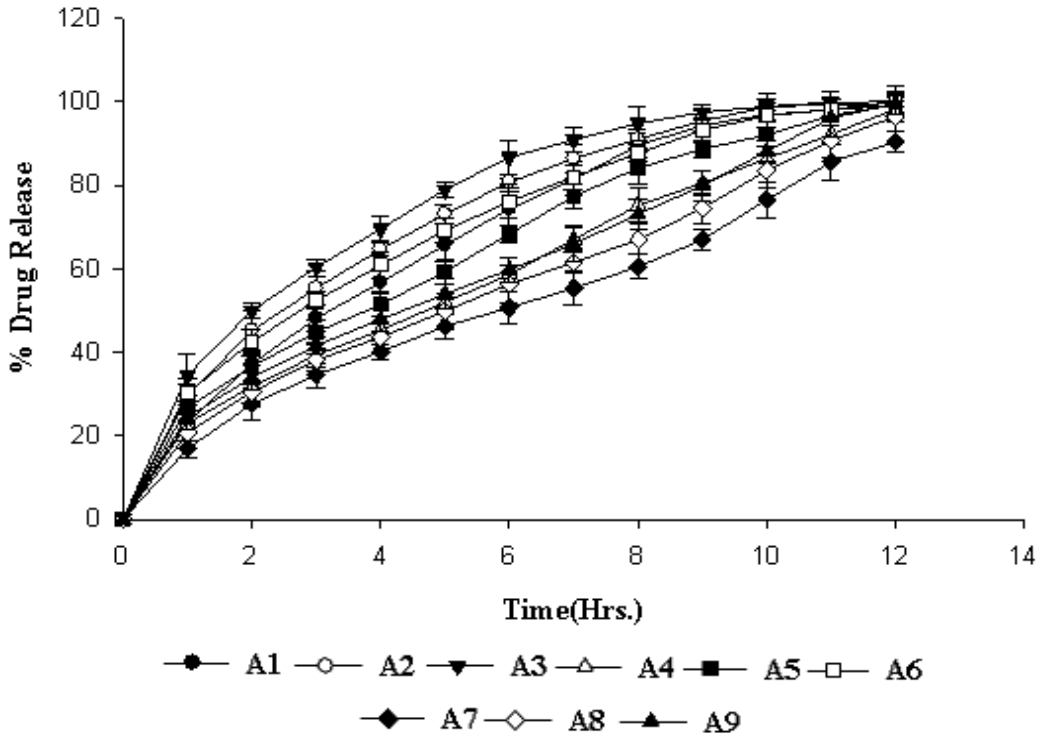

Fig: 4 Release profile of sertaconazole from factorial design batches (A1 to A9)

In all the formulations, the burst release of sertaconazole was observed within first 2 hrs, and then gradually increased up to 8-12 hrs. For the polymer mixture of HPMC K4M and Carbopol 934P, more drug release could be seen as decreasing HPMC K4M and Carbopol 934P and increasing amount of effervescent mixture.

Results of full factorial design batches

The mucoadhesive strength, % swelling, and Q_8 for the nine batches showed wide variation. The results depicted in Table: 3 clearly indicate that all the dependent variables are strongly dependent on the selected independent variables as they show a wide variation among the nine batches (A1 to A9).

Drug content for sertaconazole was carried out by measuring the absorbance of samples at 260 nm using Shimadzu UV/Visible double beam spectrophotometer. And comparing the content from a calibration curve prepared with

standard sertaconazole in the same medium. The total amount of sertaconazole present in each tablet was found to be in the range of 97.5 to 99%.

Table: 3 Characterization of factorial design batches (Mean ± SD: n=3)

Batch Code	Mucoadhesive strength(N)	% Swelling	Q_8
A1	0.18±0.014	15.30±1.3	89.51±3.03
A2	0.156±0.021	18.83±3.41	91.01±2.289
A3	0.111±0.029	21.13±3.11	94.79±3.98
A4	0.267±0.009	20.02±5.08	75.17±4.025
A5	0.236±0.025	22.74±1.87	84.15±3.995
A6	0.149±0.032	22.98±3.53	87.69±2.15
A7	0.275±0.032	22.32±3.04	60.48±2.99
A8	0.254±0.033	23.58±2.72	66.97±3.583
A9	0.162±0.016	25.51±1.41	73.03±3.456
Check point	0.121±0.018	21.65±2.09	86.03±2.025

Statistical analysis of factorial design batches

The statistical analysis of the factorial design batches was performed by multiple linear regression analysis carried out in Microsoft Excel 2007. The results are shown in Table: 4. The data clearly indicate that the values of mucoadhesive strength, % swelling, and Q_8 are strongly dependent on the independent variables. The fitted equations (full and reduced) relating the responses mucoadhesive strength, % swelling and Q_8 to the transformed factors are shown in Table: 4. The polynomial equations can be used to draw conclusions after considering the magnitude of coefficient and mathematical sign it carries (i.e. positive or negative). Table: 5 shows the results of the analysis of variance (ANOVA), which was performed to identify insignificant factors [20].

Table: 4 Summary of results of regression analysis

Model	Coefficients for mucoadhesive strength						
	b_0	b_1	b_2	b_{12}	b_{11}	b_{22}	R^2
FM	0.24	0.041	-0.05	-0.011	-0.028	-0.025	0.9838
RM	0.20	0.041	-0.05	-	-	-	0.8709
	Coefficients for % swelling (4hrs.)						
	b_0	b_1	b_2	b_{12}	b_{11}	b_{22}	R^2
FM	22.25	2.69	1.99	-0.66	-0.80	-0.51	0.9774
RM	21.37	2.69	1.99	-	-	-	0.9286
	Coefficients for Q_8						
	b_0	b_1	b_2	b_{12}	b_{11}	b_{22}	R^2
FM	82.73	-12.47	5.06	1.83	-3.03	0.59	0.9917
RM	80.31	-12.47	5.06	-	-	-	0.9629

FM indicates full model and RM indicates reduced model

Table: 5 Calculation for testing the model in portion (ANOVA)

For mucoadhesive strength						
Regression	DF	SS	MS	F	R^2	
FM	5	0.0282	0.0056	36.53	0.9838	*F*cal=5.5
RM	2	0.0249	0.0125	20.24	0.8709	*F*tab=9.28
Error						*DF*=(3,3)
FM	3	0.0005	0.002	-	-	
RM	6	0.0037	0.0006	-	-	
For % swelling						
Regression	DF	SS	MS	F	R^2	
FM	5	70.93	14.18	25.96	0.9774	*F*cal=2.145
RM	2	67.39	33.70	39.02	0.9286	*F*tab=9.28
Error						*DF*=(3,3)
FM	3	1.64	0.55	-	-	
RM	6	5.18	0.86	-	-	
For Q_8						
Regression	DF	SS	MS	F	R^2	
FM	5	1118.76	223.75	71.28	0.9917	*F*cal=3.443
RM	2	1086.33	543.17	77.88	0.9629	*F*tab=9.28
Error						*DF*=(3,3)
FM	3	9.417	3.14	-	-	
RM	6	41.85	6.97	-	-	

The high value of correlation coefficient for mucoadhesive strength, % swelling and Q_8 indicates good fit i.e., good agreement between dependent and independent variables. The equations may be used to obtain estimates of the responses as small error of variance was noticed in the replicates. The significant test for regression coefficients was performed by applying student *F* test. A coefficient is significant if the calculated *F* value is greater than the critical value of *F*. The fitted equation for full (YF) and reduced model (YR) relating the response for mucoadhesive strength(MS) , % swelling and Q_8 are shown in following equations respectively.

$$YF\,(MS) = 0.24 + 0.041X_1 - 0.05X_2 - 0.011X_1X_2 - 0.028X_1^2 - 0.025X_2^2 \quad (4)$$
$$YR\,(MS) = 0.2 + 0.041X_1 - 0.05X_2 \quad (5)$$
$$YF\,(\%\ swelling) = 22.25 + 2.69X_1 + 1.99X_2 - 0.66X_1X_2 - 0.8X_1^2 - 0.51X_2^2 \quad (6)$$
$$YR\,(\%\ swelling) = 21.37 + 2.69X_1 + 1.99X_2 \quad (7)$$
$$YF\,(Q_8) = 82.73 - 12.47X_1 + 5.06X_2 + 1.83X_1X_2 - 3.03X_1^2 + 0.59X_2^2 \quad (8)$$
$$YR\,(Q_8) = 80.31 - 12.47X_1 + 5.06X_2 \quad (9)$$

The significant level of coefficient b_{12} for mucoadhesive strength was found to be $p = 0.17$ hence it was omitted from full model to generate reduced model. The critical value of *F* for $\alpha = 0.05$ is equal to 9.28 (*DF* = 3, 3). Since the calculated value (*F* = 5.5) is less than critical value, it may be concluded that the interaction term b_{12} do not contribute significantly to the prediction of mucoadhesive strength.

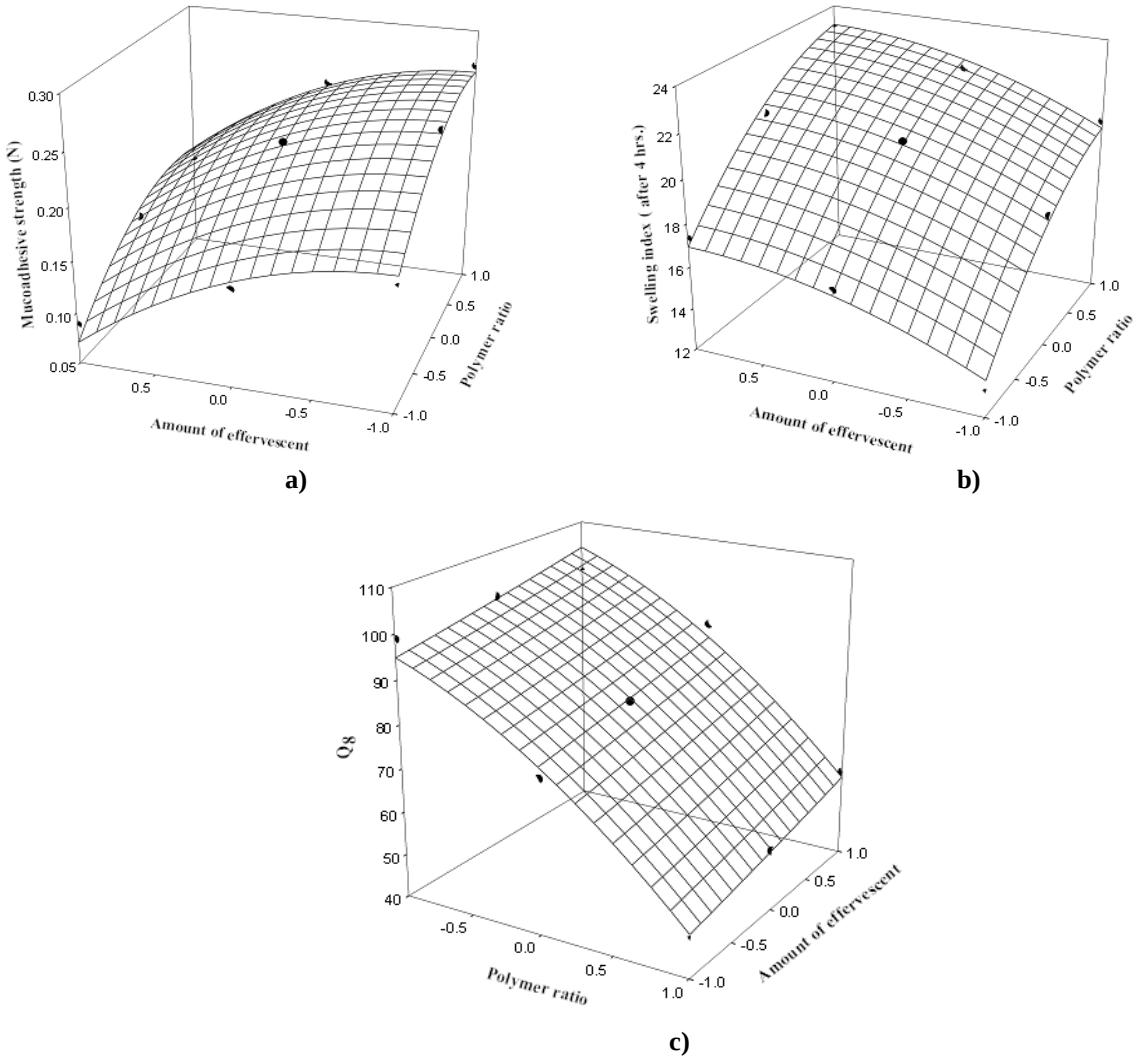

Fig: 5 3D mesh plot for a) mucoadhesive strength, b) % swelling, c) Q8

The significance level of coefficients b_{12}, b_{11} and b_{22} for % swelling are found to be $p = 0.17$, p=0.22 and $p = 0.40$ greater than $p = 0.05$, hence they were omitted from the full model to generate the reduced model. The critical value of *F* for $\alpha = 0.05$ is equal to 9.28 (*DF* = 3, 3). Since the calculated value (*F* = 2.145) is less than the critical value, it

may be concluded that the interaction term and polynomial terms do not contribute significantly to the prediction of % swelling.

The significant level of coefficient $b_{12,}$ b_{11} and b_{22} for Q_8 are found to be $p = 0.13$, $p = 0.09$ and $p = 0.67$ respectively, hence it was omitted from full model to generate reduced model. The critical value of F for $\alpha = 0.05$ is equal to 9.28 (DF = 3, 3). Since the calculated value (F = 3.443) is less than critical value, it may be concluded that the interaction term and polynomial terms $b_{12,}$ b_{11} and b_{22} do not contribute significantly to the prediction of Q_8 and therefore can be omitted from the full model.

For drawing the conclusions, 3D mesh plot was used. Fig: 5 a-c shows the plot of polymer ratio (X_1) and amount of effervescent (X_2) versus mucoadhesive strength, % swelling and Q_8 respectively. The plots were drawn using Sigma plot software Version 11 (Systat software, USA).

It was arbitrarily decided to select a batch of tablets that gives good mucoadhesive strength. The final selection is done after considering some aspects such as drug release profile, mucoadhesive strength and ex-vivo retention time. On the basis of mucoadhesive strength and dissolution release studies A7 comprising HPMC K4M: Carbopol 934P (1:1.5 ratio) and amount of effervescent (50mg) was considered a good candidate and also fall within acceptable criteria. The aim of study was, tablet should release more than 90% drug within 8-10 hrs and tablet should have satisfactory adhesive strength. Batch A7 shows good release profile which exactly fit in our objective and also shows good adhesive strength (0.275 N) which was sufficient to retain the tablet in vagina for more than 10 hrs. A checkpoint batch was prepared at X_1= -0.3 and X_2=0.7. From the reduced model, it is expected that the value of mucoadhesive strength of the checkpoint batch should be 0.121N; the value of % swelling should be 21.65 and the value of Q_8 of checkpoint batch should be 86.03%. Table: 3 indicate that the results are as expected. Thus, we can conclude that the statistical model is mathematically valid.

Kinetic analysis of dissolution data

The *in vitro* release data obtained were fitted in korsmeyer peppas kinetic model. In the entire batches exponent 'n' was in between 0.45 and 0.89, so predominant drug release mechanism is non-Fickian (anomalous) transport.

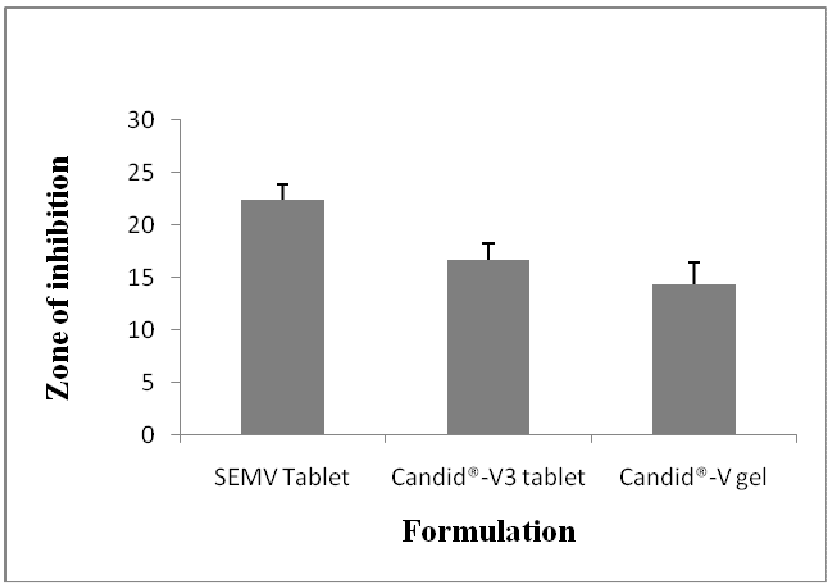

Fig: 6 Zone of inhibition (mm) for SEMV vaginal tablet, marketed vaginal Candid®-V3 tablet and Candid®-V gel. Error bar represent SD (n=3)

Anti fungal study

The sertaconazole mucoadhesive vaginal tablet A7 had better antimicrobial activity as compared with the marketed formulations (Candid V_3®- tablet and Candid V®-gel). Mucoadhesive polymers of the tablet had prolonged drug release and provided better contact with the wells cut in the plate, while the Candid V_3 suspension dried up as water was not available in the wells for prolonged time to allow diffusion of drug molecule(s). The zone of inhibition was measured at the end of 48 hours. The results of antifungal studies are reported in Fig: 6.

CONCLUSION

The results of this study reveal that incorporation of effervescent into the mucoadhesive tablets leads to the increase in the swellings and the rate of drug release and conversely the adhesion could be decreased. It was observed that with the developed formulations, the sertaconazole release and mucoadhesion properties of mucoadhesive vaginal tablets can be controlled by changing the polymer type, polymer concentration and effervescent content. The use of sertaconazole against vaginal candidiasis allowed preparing the tablets by a simple direct compression. The tablet with formulation A7 was optimized. Ex-vivo retention studies justified the prolong retention of the tablet inside the vaginal tract.

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