Spray-Dried Floating Microparticles of Ranitidine HCl by Factorial Design for Oral Delivery

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Abstract-In the present study, gastro retentive microparticulate systems of ranitidine HCl were prepared by spray drying. Biocompatible polymers like Eudragit RS100, Eudragit RL100 were used in the formulation. Factorial design (2^2) was employed by taking drug to polymer ratio and polymer to polymer ratio as factors. FTIR and DSC studies showed that Ranitidine HCl and excipients are compatible. Scanning electron microscopic studies shows spherical particles in the size range of 150 to 320 µm. Ranitidine HCl microparticles showed excellent buoyancies and release pattern upto 12 h in a controlled manner. The n value of Korsmeyer Peppas equation for the optimized formulation was found to be 0.366 which indicates the Fickian diffusion mechanism. Encapsulation efficiency was achieved up to 83.67%. The predicted values of drug release of optimized formulation at 1 h, 8 h, 12 h and entrapment efficiency were 9.88%, 72.99%, 89.86%, and 83.67% and actual values were 9.73%, 71.95%, 88.89% and 82.79% respectively. XRD studies indicate that the crystallinity of the drug is reduced in the microparticles. Photostability studies indicate that the polymer coat on microparticles protect the drug from light. The data obtained, thus suggests that a microparticulate, novel drug delivery system can be successfully designed to give oral controlled drug delivery with many desirable characteristics.
Key words: Ranitidine HCl; Floating Microparticles; Eudragit RS100; Eudragit RL100; spray drying technique.

INTRODUCTION

Current state of art is witnessing a revolution in new techniques of drug delivery. These techniques are capable of controlling the rate of drug delivery, sustaining the duration of therapeutic activity and/or targeting the drug to specific tissue (1). An appropriately designed sustained or controlled release drug delivery system can be a major advance towards solving the problems associated with the existing drug delivery system (2).

Oral drug delivery has been known for decades as the most widely utilized route of administration among all the routes that have been explored for the systemic delivery of drugs via various pharmaceutical products of different dosage forms. (3). Drugs that are easily absorbed from the gastrointestinal tract and those that have a short half life are eliminated quickly from the blood circulation. To avoid these problems, oral controlled release formulations have been developed, as they release the drug slowly into the gastrointestinal tract and maintain a constant drug concentration in the serum for a longer period of time (4, 5). Such system uses macromolecules as carriers for drugs.

The process of targeting and controlled drug delivery can be achieved by using liposomes, bioerodible polymers, implants, monoclonal antibodies and various particulate carriers like micro and nanoparticles. Microparticles can be defined as solid particles ranging in size from 1 to 1000 µm (6). Microparticles are small and have large surface to volume ratio.

It was reported that microparticles prepared with histamine H_2 receptor antagonist is effective in reducing gastric acid level and allowing acid related disease to heal (7).

Ranitidine HCl (RHCl) is an important histamine H_2 receptor antagonist whose mechanism of action is to reduce the increased gastric acid level by acting on H^+K^+ATP'ase, an enzyme present in the gastric parietal cell. This drug is effective in the treatment of gastric and duodenal ulcer, in the treatment of heartburn disease and other symptoms associated with gastro-oesophageal reflux disease. Its plasma elimination half-life is 2-3 hours.

The idea behind a controlled release microparticulate drug delivery system is to incorporate the drug within a polymeric carrier that controls the release rate of the drug (8). Eudragit microparticles are used to provide controlled release of many drugs (hydrophilic). Eudragit has also been used as a potential carrier for prolonged delivery of drugs (9), and for targeted drug delivery. In the literature, controlled release microparticulate drug delivery system for drugs like stavudine (10), zidovudine (11) and pantoprazole (12) is reported. Eudragit RS100 and Eudragit RL100 are biocompatible, hydrophobic polymers which prolong the release of water soluble and water insoluble drugs from their matrices. Acetone has lower toxicity potential compared to many other solvents and does not show any hazardous effects on the body. The polymer precipitates as the solvent evaporates during the formulation process to form porous microparticles.

Control of placement of a drug delivery system in a specific region of the GIT offers numerous advantages especially for drugs exhibiting an absorption window in the GIT. Absorption window in the proximal gut can limit the bioavailability of orally administered compounds (13, 14).

Ranitidine hydrochloride, a H_2 receptor antagonist can inhibit gastric acid secretion up to 5 hours but not up to 10 hours with the conventional dose of 150 mg. An alternative dose of 300 mg leads to plasma fluctuations; it has a short half life of 2-3 h. It is absorbed only in the initial part of the small intestine so that it can be employed for ulcers in the stomach and in the upper part of GIT. It has an absolute oral bioavailability of only 50%. Colonic metabolism of ranitidine hydrochloride is partly responsible for this low bioavailability (15, 16).

The present study is to formulate controlled gastroretentive microparticles for water soluble RHCl by using water insoluble polymers, Eudragit RS 100 and Eudragit RL 100.

MATERIALS AND METHODS

Materials
RHCl was obtained as a gift sample from BAL pharmaceutical LTD (Bangalore, India). Eudragit RS-100 and Eudragit RL-100 were received as gift samples from Evonik Degussa (Mumbai, India). Acetone and liquid paraffin light were purchased from SD fine Chem. LTD (Mumbai, India). Other chemicals were of HPLC and analytical grade.

Experimental design
2^2 factorial design is used in experiments where the effects of different factors or conditions on experimental results are to be elucidated. It is the design of choice for simultaneous determination of the effect of several factors. The simplest factorial design is the two-factorial design where two factors are considered each at two levels, leading to four experiments, which are situated in 2-dimensional factor space at the corners of a rectangle (17). The number of experiments is given by 2^n, where 'n' is the number of factors.

Factorial Design
The drug to polymer ratio was taken as factor 1 (independent variable) with its low value as 0.20 (-1) and high value as 0.50 (+1), Eudragit RS 100 to Eudragit RL 100 ratio was taken as factor 2 (independent variable) with its low value as 0.33 (-1) and high value as 0.50 (+1). Drug release at 1 hour, drug release at 8th hour, drug release at 12th hour and drug entrapment efficiency were selected as response variables. Computer-aided optimization technique using 2^2 factorial design was employed to investigate the effect of two independent variables (factors) on drug release (7). All response variables were fitted to linear equation and regression analysis was carried out to get the quantitative relationship between the dependent and analyzed independent variables.

Preformulation studies
FTIR and DSC were performed for pure drug sample. FTIR was performed at 40 ± 2^0C / 75 % RH for 2 months using Shimadzu FTIR model 8700. In order to perform DSC, pure drug sealed in 40ml aluminum crucible was analyzed under at a standard heating rate of 15ºC/minute over a temperature range of 30ºC - 300ºC using Mettler-Toledo STAR system.

Preparation of microparticles
The floating microspheres were prepared using a spray drier (Labultima LU-222, India). The experimental parameters of the process were set as follows: inlet temperature: 75ºC, feed pump speed: 120 ml/hr, aspirator setting: 30. A nozzle 0.7μm was used throughout the experiment.

RHCl containing microparticles were prepared as follows: the dichloromethane solution of RHCl, Eudragit RL100 and Eudragit RS100 was prepared in various drug: polymer and polymer: polymer ratios, fed at a concentration of 25% at a rate of 120 mL/hr (inlet temperature 75ºC) by means of a peristaltic pump and sprayed through a 0.7 μm nozzle in the drying chamber of the instrument by means of hot air flow aspirated by a pump. The obtained particles were separated in a cyclone separator and settled down in a collector (18).

Evaluation of microparticles

Drug content
The HPLC system (Agilent 1120 LC Compact, Germany) with UV detector set at 228 nm and manual injector was used. A 3 μm spherisorb cyano-bonded phase column was used at ambient temperature with a mobile phase flow rate of 1.0 mL/min. The mobile phase consists of ammonium phosphate buffer and it was prepared by adding 0.025M ammonium hydroxide to 0.025M ammonium dihydrogen phosphate to obtain a final solution of pH 5. A 300 mL aliquot of acetonitrile was placed in a suitable vessel and 0.025M ammonium phosphate buffer (pH 5.0) was added to obtain a final volume of 1 L. The mixture was well stirred, filtered, and deaerated under reduced pressure (19). Floating microparticles equivalent to 5 mg of drug were dissolved in 5 mL methanol and diluted suitably with the above buffer. Sample of 20 μL was injected into the column and analyzed.

In vitro buoyancy
Floating microparticles (equivalent to 100 mg of RHCl) were dispersed in 900 mL of 0.1N hydrochloric acid solution (pH 1.2) containing tween 80 (0.01 W/V %) / tween 20 (0.02 W/V %) to simulate gastric fluid at 37°C. The mixture was stirred with a paddle at 100 rpm and after 12 h, the layer of floating microparticles (W_f) was separated by filtration. Simultaneously, sinking microparticles (W_s) were also separated. Both floating and sinking microparticles were dried at 40°C overnight. Each weight was measured and buoyancy was determined by the weight ratio of the floating microparticles to the sum of floating and sinking microparticles.

$$\text{Buoyancy (\%)} = W_f/W_{f+} W_s \times 100$$

Entrapment efficiency
RHCl microparticles equivalent to 5mg of drug were taken into pH 1.2 buffer and placed on a magnetic stirrer. Sample was withdrawn at 15 min. and the microparticles were filtered by using 0.3 μm membrane filter. The filtered microparticles were dissolved in 5ml of methanol and assayed for drug content by HPLC method (19).

Scanning Electron Microscopy (SEM)

Scanning Electron Microscopy **was** carried out for the optimized formulation. Dry microparticles were placed on an electron microscope, fixed on a brass stub using double-sided tape and then gold coated in vacuum by a sputter coater. The picture of microparticles was taken by random scanning of the stub. The SEM analysis of the microparticles was carried out by QUNTA-200 FEI (Netherland) using analytical Scanning Electron Microscope. The microparticles were viewed at an accelerating voltage of 20 KV.

Particle size

The particle size was measured by optical microscopic technique. In this method suspension of microparticles was prepared using castor oil. A drop of suspension was mounted on a slide and observed using Jenoptik[TM] camera (3.1MP) attached to optical microscope (LX 400; x400 magnification). About 100-200 particles were measured with the help of the measuring scale provided in the software (Capture Pro[TM]) which was previously calibrated using a stage micrometer. All the microparticles in field were counted (20).

In vitro dissolution studies

In vitro release profile of the RHCl microparticles was evaluated using USP type-2 paddle dissolution apparatus (Electrolab dissolution tester TDT08L, India). Simulated gastric fluid (0.1N hydrochloric acid buffer pH-1.2 without enzymes, 900 mL) was used as dissolution medium maintained at 37 ± 0.5^0C, and the paddle was rotated at a constant speed of 100 rpm. The dissolution apparatus was enclosed in a black paper as the drug is photosensitive. Accurately weighed amount of RHCl microparticles equivalent to 300 mg of drug were tied in muslin cloth and placed in the jar. Aliquots o f 5 m L were withdrawn at an interval of 1 h for 12 h and replaced with 5mL of dissolution medium. The samples withdrawn were filtered by using nylon filters of 0.45 μm pore size, diluted suitably and analyzed by HPLC for drug release at 228 nm (19).

Regression analysis

The response parameters were statistically analyzed by applying one way ANOVA at 0.05 level using commercially available software Design-Expert software (Stat-Ease Inc, Minneapolis, USA). The individual parameters were evaluated using F-test and linear models were generated for each response parameter using Multiple Linear Regression Analysis (MLRA) equation:

$$R = b_0 + b_1 A + b_2 B + b_3 AB + b_4 A^2 + b_5 B^2 + b_6 AB^2 + b_7 A^2 B \qquad (1)$$

Where, R is the level of measured response, b_0 is the intercept of the arithmetic mean response of 4 runs, A and B are the coded levels of the independent variables, AB is the interaction term that shows how response changes when two factors are simultaneously used, A^2 and B^2 are quadratic terms of the independent variables to evaluate the non-linearity.

Fourier Transform Infrared Spectroscopy (FTIR)

IR spectroscopy (Shimadzu FTIR model 8700) was carried out for Ranitidine HCl(Pure drug) , Drug and Eudragit RS100, Drug and Eudragit RL100 by keeping at 40 ± 2^0C / 75 % RH for 2 months using KBr disc method and scanned for absorbance.

Differential Scanning Calorimetry (DSC)

The sample of RHCl and binary mixtures of drug and polymer were weighed and sealed in 40ml aluminum crucibles with a pierced aluminum lid. The analyses were performed under nitrogen (nitrogen flow rate 50 mL/min) in order to eliminate oxidative and pyrolytic effects at a standard heating rate of 15ºC/minute over a temperature range of 30ºC - 300ºC using a Mettler-Toledo STAR system.

X-Ray Diffraction (XRD)

The XRD patterns of samples were recorded using Philips PW 1830 X-ray diffractometer to find out any change in the crystallinity of drug during microencapsulation.

Photostability studies

Stability studies were carried out according to ICH Q1B guidelines in Newtronic chamber. Studies were carried out in UV light, tube light 1.2 million lux hours and an integrated near ultraviolet energy of not less than 200 watt hours per square meter to allow direct comparison to be made between the drug substance and the drug product.

RESULTS

Percentage Drug Content

The percentage drug content of all the formulations was found to be in the range of 92.01 to 96.34%, shown in table no. II.

In vitro buoyancy

The values for *in vitro* buoyancy were shown in Table no. II, they were in the range of 89% to 92%.

Entrapment Efficiency (EE)

The values for entrapment efficiency were shown in Table no. II. The entrapment efficiency of Eudragit based microparticles were in the range of 77% to 83%.

Scanning Electron Microscopy

The SEM of microparticles was shown in fig. no. I.

Particle Size

The particle size of all the microparticles (F1-F4) ranged from 150 to 320 μm.

In vitro Release

In vitro drug release was shown in fig. no. II.

Effect of formulation variables on in vitro drug release after 1st h

Total amount of ranitidine released from all formulations ranged from 5.83% to 12.87 % in 1 h (Table no: III) as shown in the fig. no. III.

Effect of formulation variables on in vitro drug release after 8th h

Amount of RHCL released from all formulations at 8th h ranged from 68.34% to 79.36% (Table no. III). Decreased rate of drug release was observed with increased ratio of polymers as shown in the fig. no. IV.

Effect of formulation variables on in vitro drug release at 12th h

Amount of ranitidine released from all formulations at 12th h. ranged from 81.96 % to 97.13% (Table no: III) as shown in fig. no. V.

ANOVA

The results of ANOVA demonstrate that the model was significant. The summary of ANOVA was given in the table no. IV.

Optimization

The predicted values of drug release of optimized formulation at 1 h, 8 h, 12 h and entrapment efficiency were found to be 9.88%, 72.99%, 89.86%, and 83.67% and actual values were 9.73%, 71.95%, 88.89% and 82.79% respectively.

FTIR studies

The peaks of drug, binary mixture of drug and eudragit RS 100, drug and eudragit RL 100 are shown in the Table No.VI. In the drug-excipient interaction study, it was found that RHCl was compatible with all the excipients used in the formulation (Table no. VI).

DSC

In DSC studies melting peak appeared at 76.43° C for ranitidine HCl. The peaks were shown in the fig. no. VII.

XRD

The XRD of drug and microparticles was depicted in fig. no. VIII.

Table No. I: Factorial Design Formulation chart

Formulation code	Ranitidine (mg)	Eudragit RS 100 (mg)	Eudragit RL 100 (mg)
F1	300	400	800
F2	300	150	150
F3	300	600	600
F4	300	100	200

Table No. II: Percentage Yield, Drug Content and Entrapment Efficiency of Ranitidine Microparticles

Formulation code	Percentage yield	Drug content (%)	Entrapment Efficiency (%)	Invitro buoyancy (%)
F1	89.02	94.4	80	89.44
F2	80.16	95.1	82.10	92.73
F3	86.9	92.6	83	90.21
F4	85.20	96.34	77.19	92.69
Optimized formula	86.96	98.82	83.67	92.81

Table No. III: Design and Summary of Responses Data

Run	Formulation code	D:P (mg)	P:P (mg)	In-vitro release 1hr	In-vitro release 8hr	In-vitro release 12hr	% EE
1	F1	0.20	0.33	7.65	70.13	84.37	88
2	F2	0.50	0.50	12.87	73.23	94.39	79
3	F3	0.20	0.50	5.83	68.34	81.96	89
4	F4	0.50	0.33	12.41	79.36	97.13	77.19

Table No. IV: Summary of ANOVA table for dependable variables

Source	d.f.	Sum of squares	Mean square	F value	Probability
Drug release at 1 hour					
A	1	34.81	34.81	39.51	0.0244*
Drug release at 8 hour					
A	1	49.84	49.84	4.89	0.1576
Drug release at 12 hour					
A	1	158.63	158.63	47.65	0.0203*
Entrapment efficiency					
A	1	108.26	108.26	660.05	0.0248*
B	1	1.97	1.97	12.03	0.1787

*Significant

**Table No. V: Summary of ANOVA results model, residual and corrected total**

Source	Sum of squares	d.f.	Mean square	F value	Probability > F
Drug release at 1 hr					
Model	1	34.81	34.81	39.51	0.0244*
Residual	2	1.76	0.88	-	-
Corrected total	3	36.57	-	-	-
Drug release at 8 hr					
Model	1	49.84	49.84	4.89	0.1576
Residual	2	20.39	10.20	-	-
Corrected total	3	70.23	-	-	-
Drug release at 12hr					
Model	1	158.63	158.63	47.65	0.0203*
Residual	2	6.66	3.33	-	-
Corrected total	3	165.29	-	-	-
Entrapment efficiency					
Model	2	110.24	55.12	336.04	0.0385*
Residual	1	0.16	0.16	-	-
Corrected total	3	110.40	-	-	-

d.f. degree of freedom
* Significant

Table No.VI: FT-IR data of Ranitidine alone and with excipients

Bond	Wave no. cm^{-1} (Pure drug)	Wave no. cm^{-1} (Eudragit RS 100 + drug)	Wave no. cm^{-1} (Eudragit RL 100 + drug)
N –H stretch	2500	2509.64	2504.91
C=N stretch	1610	1603.18	1611.84
-NO2	1460	1467.93	1463.73
-NO2	1252	1246.38	1248.46
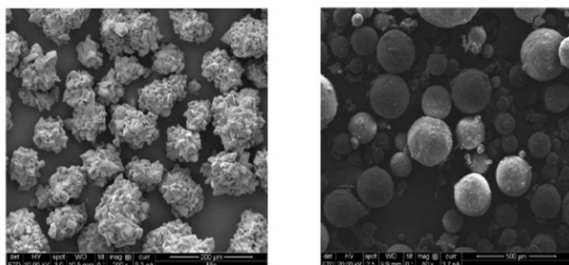
Fig. No. I: SEM photographs of pure Ranitidine HCl drug and optimized formulation

% CDR = Percentage Cumulative drug Release
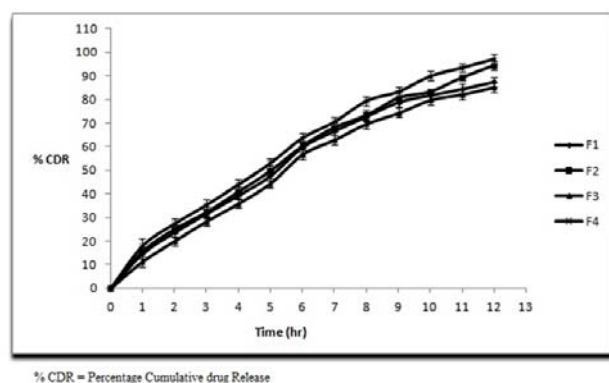
Fig. No. II: In-vitro drug release profiles of formulations F1-F4

DISCUSSION

FTIR studies
Fourier-transform infrared (FTIR) spectroscopy was performed on each of the samples to identify the presence of specific functional groups within a sample. Furthermore, drug-polymer interactions were examined using the resulting spectra. Spectra are obtained by passing infrared radiation through a sample and determining what fraction of incident radiation is absorbed at a particular energy. In the drug-excipient interaction study, it was found that RHCl was compatible with all the excipients used in the formulation. As there are no extra peaks and no shifting of peaks of the functional groups of RHCl (all peaks are within the ± 5 cm^{-1}) in the spectra of binary mixtures of drug and excipients.

Scanning Electron Microscopy
The scanning electron microscopic studies were carried out to study the shape of the microparticles formed and surface morphology of the microparticles. From the SEM photographs it can be concluded that microparticles are spherical in shape with smooth surface. SEM is depicted in fig. no. I.

In vitro buoyancy
More than 87% of the microspheres remained buoyant for 12 hrs in all the formulations due to hollow nature of the microspheres (Table No. II).

In vitro Release

The *in vitro* release of Ranitidine from the prepared microparticle formulation was studied in pH 1.2 acid buffer for 12 h. As the ratio of polymer increased, the drug release decreased proportionally. The release rate of ranitidine from the Eudragit microparticles increased in the following order: Eudragit RS< Eudragit RS/RL<Eudragit RL. The release rate of microspheres prepared from Eudragit RL was remarkably faster than that from Eudragit RS microspheres. This may be due to the structural differences between polymer types i.e. the difference in the content of the quaternary ammonium group, high content of the ammonium group facilitates the diffusion of the entrapped drug to the surrounding medium at a faster rate due to its high permeability. It could be explained considering the chemical structure of Eudragit. Eudragit RL and RS are synthesized from acrylic and methacrylic esters with high and low content of quaternary ammonium groups that result in microspheres with different water permeabilities (21). *In vitro* drug release is represented in fig. no. II.

Effect of formulation variables on in vitro drug release after 1st h

Total amount of ranitidine released from all formulations ranges from 5.83% to 12.87 % in 1 h (Table No. III). Decreased rate of drug release was observed with increase in the ratio of polymers.

Drug release at 1st h = + 9.69 + 2.95 A (Coded values)

Drug release at 1st h = + 2.81 + 19.67 A (Actual values)

The linear model selected for these responses with Model F-value 39.51 and p value 0.0244 less than 0.0500 indicate that the model is significant. From the above equation, the coefficient of factor A is positive which indicates that the drug release increases as factor A (D: P) increases. The effect of A can be further elucidated with the help of response surface plot (Fig. No. III). At low level and high level of B and as factor A increases the drug release increases significantly. Factor B was not found to be significant. The PRESS value was found to be 22.30 which is less. From the graph of actual and predicted values, it can be understood that the there is experimental agreement with the model.

Effect of formulation variables on in vitro drug release after 8th h

Amount of RHCL released from all formulations at 8th h ranges from 68.34% to 79.36% (Table No. III). Decreased rate of drug release was observed with increased ratio of polymers which indicate that Eudragit RS 100 retards release to more extent than that of Eudragit RL 100 (Fig. No. IV). In this case, effect of drug to polymer ratio can be explained by mathematical equation in terms of actual factors:

Drug release at 8th h = + 72.77 + 3.53 A (Coded values)

Drug release at 8th h = + 64.53 + 23.53 A (Actual values)

The linear model selected for these responses with Model F-value 4.89 and p value 0.1576 more than 0.0500 indicate

that the model is not significant. From the above equation, the factor A (D: P) increases the drug release as indicated by the positive coefficient of factor A. The effect of A can be further elucidated with the help of response surface plot (Fig. No. IV). At low and high level of B and as factor A increased the drug release increased significantly. Factor B was not found to be significant. The PRESS value was found to be 7.08 which is very less. From the actual and predicted values graph, it can be understood that the there is experimental agreement with the model.

Effect of formulation variables on in vitro drug release at 12th h

Amount of RHCl released from all formulations at 12th h ranges from 81.96 % to 97.13% (Table No. III). In this case, effect of drug to polymer ratio can be explained by mathematical equation in terms of actual factors:

Drug release at 12th h = + 89.46 + 6.30 A - 1.29 B (Coded values)

Drug release at 12th h = + 81.05 + 41.98 A - 15.15 B (Actual values)

The linear model selected for this response with Model F-value 47.8 and p value 0.0203 indicates that the model is significant. From the above equation, the coefficient of factor A is positive which indicates the increase in drug release with increase of factor A (D: P). The effect of factor A can be further elucidated with the help of response surface plot (Fig. No. V). At low and high level of B and as factor A is increased the drug release increased significantly. Factor B was not found to be significant. The PRESS value was found to be 34.20 which is less. From the graph of actual and predicted values, it can be understood that there is experimental agreement with the model.

Entrapment Efficiency (EE)

The values for entrapment efficiency were shown in table no. III. For Eudragit based microparticles, the entrapment efficiency was in the range of 77.19% to 89%. The results of entrapment efficiency and drug content indicated that EE increased with increase in drug to polymer ratio. The relative entrapment efficiency and drug content of all the formulations is depicted in fig no.VI.

Entrapment efficiency = + 83.3 - 5.2 A + 0.7 B (coded values)

Entrapment efficiency = + 92 - 34.68 A + 8.26 B (Actual values)

The linear model was found to be significant for entrapment efficiency. The Model F-value of 660.05 and p value of 0.0248 which is less than 0.0500 indicate that the model is significant. In the above equation the coefficient of the factor A is negative value which indicate that entrapment efficency decreases as the factor A increases. The coefficient of factor B is positive which indicate that the entrapment efficiency increases as the factor B increases. Hence, the entrapment efficiency is high with more Eudragit RS 100. The PRESS value was found to be 0.00. From the actual and predicted values graph, it can be understood that there is experimental agreement with the model as it is linear.
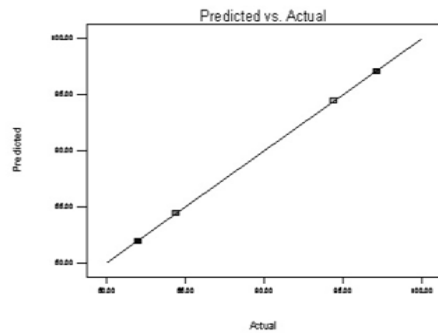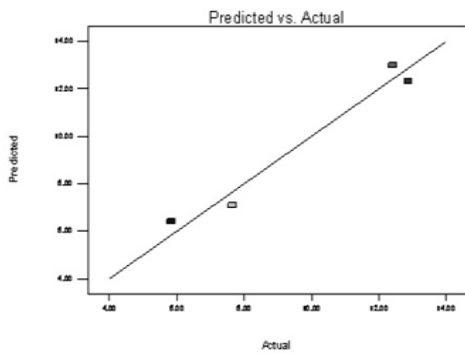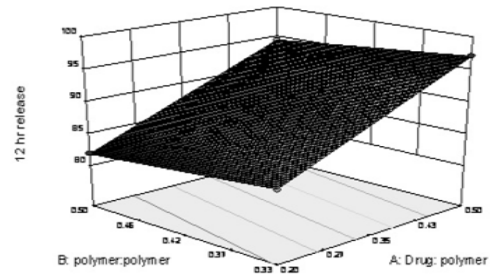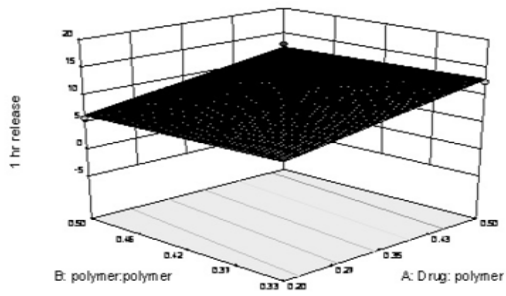
Fig No. III. Response surface plot showing the effect of formulation variables on percentage drug release at 1 h and comparison graph of actual vs. predicted values of the response

Fig No. V. Response surface plot showing the effect of formulation variables on percentage drug release at 12th h and comparison graph of actual vs. predicted values of the response
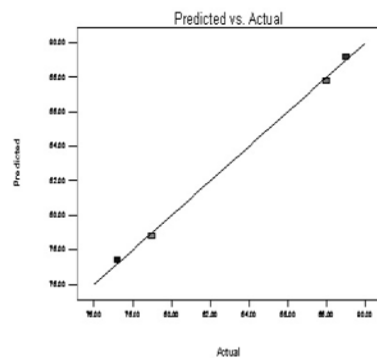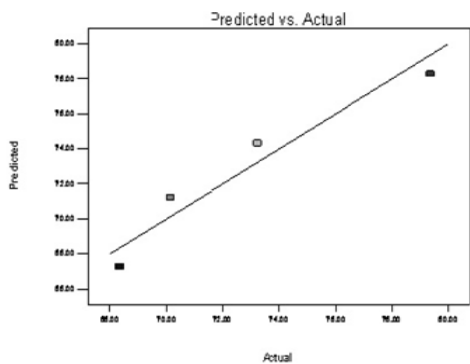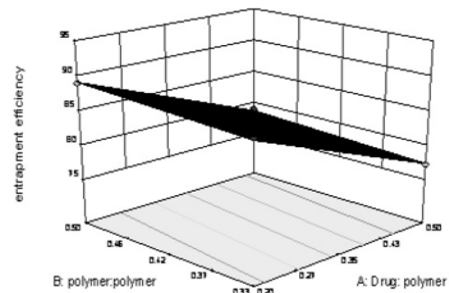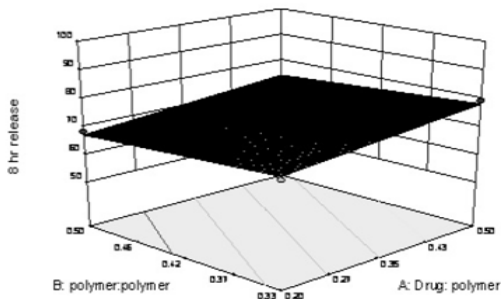
Fig No. IV. Response surface plot showing the effect of formulation variables on percentage drug release at 8th h and comparison graph of actual vs. predicted values of the response

Fig No. VI. Response surface plot showing the effect of formulation variables on entrapment efficiency and comparison graph of actual vs. predicted values of the response
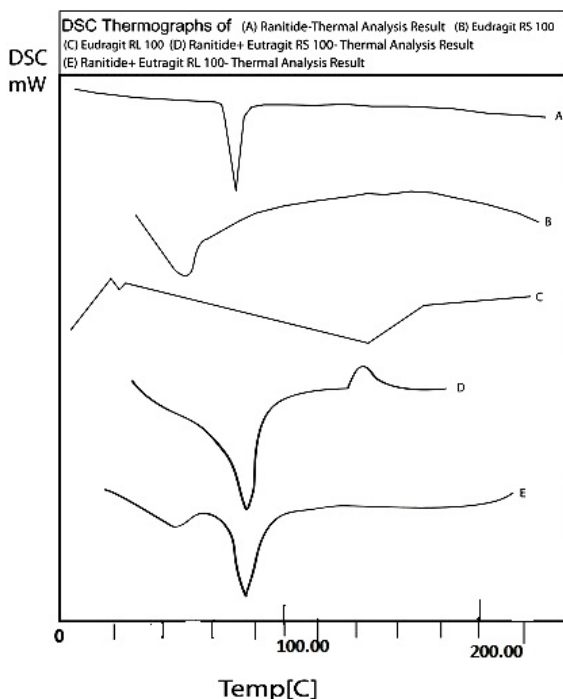
Fig. No. VII: DSC graphs of Ranitidine HCl with Eudragit RS-100 and Eudragit RL-100
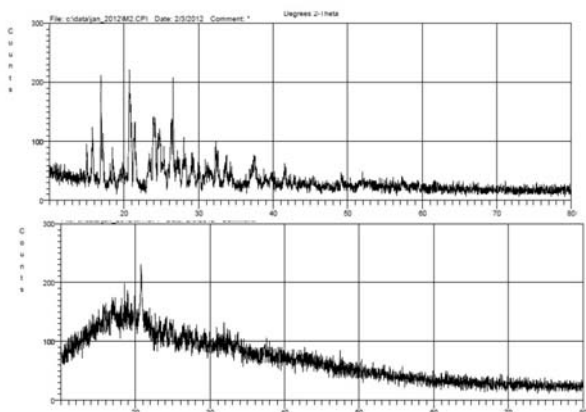

Fig. No. VIII: XRD of Ranitidine HCl and optimized formulation

ANOVA
The results of ANOVA demonstrate that the model was significant. Regression analysis was carried out to determine the regression coefficients. The factor A was found to be significant for response variables. The linear model was found to be significant for responses. So, above results indicate that the factor A plays an important role in the formulation of microparticles containing RHCl. The summary of ANOVA can be observed from the table no. IV.

Optimization
In the numerical optimization technique, the desirability approach was used to generate the optimum settings for the formulation. For the optimized formulation, drug entrapment efficiency was kept at maximize, the drug release at 1st h was kept at maximize, the drug release at 8th h was kept in range, the drug release at 12th h was kept at minimize. The composition of optimized formula is Ranitidine HCl (675 mg), Eudragit RS 100 (600 mg), Eudragit RL 100 (600 mg). The optimized formulation was prepared according to the predicted model and evaluated for responses. The optimized formula showed controlled drug release of 84.68% and high encapsulation efficiency of 83% for F3 (Table No. II) . The predicted values of drug release of optimized formulation at 1 h, 8 h, 12 h and entrapment efficiency are 9.45%, 70.78%, 86.61%, and 85.39% while the actual values are 9.33%, 69.95%, 85.89% and 84.79% respectively.

FTIR studies
Fourier Transform Infra Red (FTIR) spectroscopy was performed on each of the samples to determine the structure of the organic compounds and to identify the presence of specific functional groups within a sample. Furthermore, drug-polymer interactions were examined using the resulting spectra. Spectra were obtained by passing infrared radiation through a sample and determining what fraction of incident radiation was absorbed at a particular energy. In the drug-excipient interaction study, it was found that ranitidine was compatible with all the excipients used in the formulation (Table No. VI) as there are no extra peaks and no shifting of peaks of the functional groups of ranitidine (all peaks are within the $\pm 5cm^{-1}$) in the spectra of binary mixtures of drug and excipients.

DSC:
In DSC studies melting peak appeared at 76.43°C for ranitidine HCl. There was no change in the melting point of binary mixture of Ranitidine HCL + Eudragit RS-100 and Ranitidine HCl + Eudragit RL-100 which indicate that there is presence of drug in the microparticles. No interaction was observed between the drug and polymers. The peaks were shown in the fig. no. VII.

XRD
In powder x-ray diffraction pattern of ranitidine HCl, peaks were observed which indicates the crystalline nature of the drug. The number of peaks reduced in the formulation of microparticles compared to pure ranitidine HCl which indicate that the crystallinity is reduced in the formulation. Powder XRD is depicted in the fig. no. VIII.

Photostability studies
The photostability studies show that the percentage drug content of microparticles (96.81%) was found to be more than that of pure drug (93.90%) [without light protection] which indicate that the microparticles protect the drug from light due to the polymer coating present around the drug. The drug content for reference [with light protection] microparticles (98.82%) was found to be more than that of reference pure drug (96.69%).

CONCLUSION

The factorial design was used to find out the effect of independent variables on the dependent variables. The results of factorial design revealed that the drug to polymer ratio and polymer to polymer ratio had significant effect on the drug release at 1 h, drug release at 8th h, drug release at 12th h and entrapment effiency. The drug to polymer ratio has greater significant effect than polymer to polymer ratio and the observed independent variables were found to be very close to predicted values of optimized formulation which demonstrates the feasibility of the optimization procedure in successful development of microparticles containing Ranitidine HCl by using Eudragit copolymers RS-100 and RL-100.

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