Acta Scientific Pharmaceutical Sciences (ISSN: 2581-5423)

Volume 6 Issue 8 August 2022

Research Article

Formulation and Evaluation of Clarithromycin Enteric Coated Microcapsules Using 2^2 - Full Factorial Designs

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Received: May 30, 2022

Published: July 03, 2022

Abstract

The purpose of the research was to develop and evaluate Clarithromycin loaded, Cellulose acetate phthalate (CAP) enteric coated, controlled release microcapsules where talc is used as an anti aggrading agent. Clarithromycin degrades rapidly at normal gastric pH (1.0-2.0) and remains stable at intestinal pH at 7.4-10 and shows rapid first-pass hepatic metabolism. In order to improve the bioavailability and reduce the gastric degradation, enteric coating microencapsulation was prepared. Clarithromycin microcapsules were formulated by solvent evaporation technique while using 2^2 factorial designs. A 2^2 full factorial designs was used to derive a statistical equation, ANOVA analysis, contour plots, and 3D response surface plots. Different polymer and anti aggregating agent ratios of CAP and talc were used to formulate five formulations (F1 to F5) of microcapsules. The relationship between dependent variables (Percentage drug release, drug entrapment efficiency, angle of repose) and independent variables (CAP and Talc) has been established by regression analysis and ANOVA.

The optimized formulations F2 exhibited high drug entrapment efficiency of 94.90 ± 0.02%, Angle of repose of % drug release of 71.51 ± 0.04% at 10 hrs. Microcapsules showed drug release by diffusion in the hydrated matrix and polymer relaxation as a controlled release mechanism.

Keywords: Clarithromycin; Enteric Coated Microencapsulation; Solvent Evaporation Technique; Cellulose Acetate Phthalate (CAP); Factorial Design

Introduction

Clarithromycin is a macrolide antibiotic used to treat pharyngitis, tonsillitis, acute maxillary sinusitis, acute bacterial exacerbation of chronic bronchitis, pneumonia (especially atypical pneumonias associated with Chlamydia pneumoniae or TWAR), skin and skin structure infections. In addition, it is sometimes used to treat Legionellosis [1], Helicobacter pylori, and lyme disease.

Clarithromycin prevents bacteria from growing by interfering with their protein synthesis. Clarithromycin binds to the subunit 50S of the bacterial ribosome and thus inhibits the translation of peptides. Clarithromycin has similar antimicrobial spectrum as erythromycin but is more effective against certain gram-negative bacteria, particularly [2] Legionella pneumophila.

It is readily absorbed, and diffused into most tissues and phagocytes. Due to the high concentration in phagocytes, clarithromycin is actively transported to the site of infection. During active phagocytosis, large concentrations of clarithromycin are released. Clarithromycin was developed in 1980 and approved for medical use in 1990. It is on the World Health Organization's List of Essential Medicines.

Clarithromycin has a fairly rapid first-pass hepatic metabolism. Clarithromycin and its metabolites main routes of elimination are urinary and biliary excretion [3]. Of all the drugs in its class, clarithromycin has the best bioavailability at 50%, which makes it amenable to oral administration.

Even when the aim of a microencapsulation application is the isolation of the core from its surrounding, the wall must be ruptured at the time of use [4]. Many walls are ruptured easily by pressure or shear stress, as in the case of breaking dye particles during writing to form a copy. Capsule contents may be released by melting the wall, or dissolving it under particular conditions, as in the case of an enteric drug coating. In other systems, the wall is broken by solvent action, enzyme attack, chemical reaction, hydrolysis, or slow disintegration [5].

Materials and Methods

Materials

Clarithromycin IP obtained from ALEMBIC Ltd, Vadodara, Gujrat; India, Cellulose acetate phthalate [FLUKA], Talc fine powder extrapure. [LOBA CHEME], Tetrahydrofuran [MERCK], Polyvinyl alcohol. [LOBA CHEME], Potassium dihydrogen phosphate(purified) [MERCK], Sodium hydroxide pellets [MERCK], Double distilled water [Prepared in the laboratory from deminaralized water].

Instruments

REMI Electrical stirrer, Magnetic stirrer, UV – Visible spectrophotometer [Shimadzu 1700, Japan], Six station Dissolution apparatus [Labindia Disso].

Methods

Preparation of clarithromycin microencapsulation

Clarithromycin microencapsulation by solvent evaporation technique

25 ml tetrahydrofuran was taken in a bicker and cellulose acetate phthalate (measured according to formula) was added to it, within 15mins cellulose acetate phthalate was dissolved in tetrahydrofuran. 1g of clarithromycin, talc (according to formula), was mixed with above solution. 1% polyvinylaicohol solution (1g of polyvinyl alcohol was dissolved in boiled water and vol was made up to 100 ml with water) was made. (CAP + 25 ml THF + talc + drug) solution was dispersed slowly into 100 ml 1%PVA solution. Stirred the above solution by electric stirrer for 2-3 hrs [5,6].

2^2 full factorial designs

A 2^2 full factorial design was employed taking amount of CAP (x1) and amount of talc (x2) as two independent variables with a center point as per table 1. Example was carried out a triplicate manner for both the corner point and center point, total 15 formulation ware prepared as per table 2. Model accuracy was chalked by employing analysis of variance and plotting normal probability plot with residual plot. A regulation model was chosen including liners, quadratic and interaction terms to study the effect of different independent variables on the response.

CAP (X_1)	TALC (X_2)
L (-1)	L (-1)
H (+1)	L (-1)
L (-1)	H (+1)
H (+1)	H (+1)
M (0)	M (0)

Table 1: 2^2 full factorial designs.

Coded variable: L = (-1); H = (+1); M=(0);

Actual variable: [CAP] [-1 (L) = 2g]; [+1 (H) = 4g]; [0 (M)= 3g]
[Talc] [-1 (H) = 0.5g]; [+1(H) = 1.5g]; [0 (M) = 1g]

Replicate	Code	Coded variables	Actual values (mg)		
	No	X_1 (CAP)	X_2 (TALC)	X_1 (CAP)	X_2 (TALC)
1	F1	-1	-1	2	0.5
	F2	+1	+1	4	1.5
	F3	-1	+1	2	0.5
	F4	+1	-1	4	0.5
	F5	0	0	3	1
2	F1	-1	-1	2	0.5
	F2	+1	+1	4	1.5
	F3	-1	+1	2	0.5
	F4	+1	-1	4	0.5
	F5	0	0	3	1

3	F1	-1	-1	2	0.5
	F2	+1	+1	4	1.5
	F3	-1	+1	2	0.5
	F4	+1	-1	4	0.5
	F5	0	0	3	1

Table 2: 2^2 full factorial design in a triplicate manner.

2^2 Full Factorial Design Studies:

Two regression equations were determined including linear, interaction and quadratic terms for 4 responses (dependent variables) such as angle of repose (AR), % clarithromycin entrapment efficiency (% CEE), % drug released at 2 hrs (Q2) and % drug released at 10 hrs (Q_{10}) [7].

The regression equation was as follows

$$RESPONSE = \beta_0 + \beta_1 X_1 + \beta_2 X_2 + \beta_{11} X_1^2 + \beta_{22} X_2^2 + \beta_{12} X_1 X_2$$

Where, β_0 (Average response), $\beta_1, \beta_2, \beta_{11}, \beta_{22}$, and β_{12} are regression coefficients, whose values are to be determined by the following matrix equation.

$$\beta = (X'. X)^{-1}. (X'.Y)$$

X = Design matrix of the order of 15x3

Fourier transforms infrared spectroscopy (FT-IR)

Fourier Transform Infrared Spectroscopy (FT-IR) is a rapid, easy and inexpensive analytical technique that used to predict the drug-excipients interactions.

Flow property study

Formulation whose flow property is to be studied was passes freely into a fixed funnel which hold by a stand and was produced a conical shaped in the white page which placed below the funnel. Measured the radius and height of the cone. And put into the formula.

$\tan\Theta = H/r$

H = Height of the cone.

r = Radius of the base of the cone

Θ = Angle of repose in degrees.

If $\Theta \leq 30$, it indicates good flow property, and if $\Theta \geq 40$, it indicates poor flow property.

Drug entrapment efficiency

100mg of formulation was added to 900ml PBS [pH 7.4] and Stirred for 48 hrs. Filtered and the solution and analyzed by UV – Vis. Spectrophotometer at 265 nm against reagent blank. From the % of the DC, % of Clarithromycin entrapment efficiency (%CEE) was calculated by using the following formula [8].

% CEE = Practical % DC * 100/Theoretical % DC

Whereas, Wt = Equilibrium weight of microcapsules after swelling and Wo = Initial weight of microcapsules.

In vitro release kinetic study

Formulation containing 250 mg of Clarithromycin was weighed. 900 ml HCl was measured and put it into the dissolution chamber. Formulation was put into the HCl, and started the dissolution apparatus, with 50 rpm, in (25 + 0.5)°C, for 2 hrs. After 30 mins 5 ml sample was collected, and 5 ml HCl was replace into dissolution chamber, thus 4 samples was collected in 1 hrs interval. 5 ml sample was taken in 25 ml volumetric flask, and volume was made up to 25 ml by PBS. All the sample ware measured by U.V Spectrophotometer against blank [5 ml 0.1N HCl + 20 ml PBS] [9].

Then formulation was filtered and put it into phosphate buffer in dissolution apparatus, and run for 12 hrs, in 50 rpm and 37°C. After 30 mins 5 ml sample was collected, and 5 ml PBS was replace into dissolution chamber, thus 10 samples was collected with 1 hrs interval. 5 ml sample was taken in 25 ml volumetric flask and 5 ml HCl was added and volume was up to 25 ml by PBS. All the sample ware measured by U.V Spectrophotometer against blank [5 ml 0.1 N HCl + 20 ml PBS].

Drug release kinetic profile

The following mathematical models evaluated to determine the drug release per unit time, namely zero order and first order whereas Higuchi and Korsmeyer-Peppas model was used to evaluate the mechanism of drug release according to table 3.

Model	Equation	Graph plotted
Zero order	$\%DR = K_o t$	% drug release (%DR) versus time in hour
First order	$\%DR = 100\ (1\text{-}e^{-K_1 t})$	log (%ARA) versus time
Higuchi	$\%DR = K_h t^{0.5}$	% DR versus square root of time
Hixson Crowell	$W_0^{1/3}\text{-}W^{1/3} = K_2 t$	$W_0^{1/3}\text{-}W^{1/3}$ versus time

Korsemeyer Peppas	$M_t/M_0 = K_k t^n$	Log (% DR) versus log (time)

Table 3: Mathematical model.

Where Ko, Kt, Kh, K2 and Kk are respective dissolution rate constants.

% DR = Percentage drug release.

W = Amount of the drug remaining to be released at time t = ARR.

Mt = Amount of the drug released at time t.

$M_0 = W_0$ = Original amount of the drug = Drug Content in mg.

Mathematical and statistics analysis of experimental design

A 2^2 design was employed in triplicate manner and total 15 microcapsules of clarithromycin were prepared. Angle of repose (AR), % clarithromycin entrapment efficiency (%CEE), % DR at 2 hrs (Q2) and at 10 hrs (Q10) were considered as four responses for optimization. Factorial analysis and P-values < 0.05 were considered significant [10]. Responses observed for each of the formulations were simultaneously fitted to quadratic model and Analysis of variance (ANOVA) results for all the responses explored to determine the significant effect of independent variables CAP (cellulose acetate phthalate) and TALC). P-value of CAP and TALC (given in ADX report) revealed that both have significant effect on the response variables. ANOVA table for both the responses along with the design details were given below in ADX report generated by statistical software SAS. Normal probability plots were generated to check the model accuracy.

Result and Discussion

λmax is of clarithromycine was determined from U.V. Visible spectrophotometer by scanning through the entire range (190-1100). The λmax was found to be 265 n against the reagent blank.

Amount of drug released at different time intervals three standard curves were calibrated, Equation of the standard curve for R1 was y = 0.0187x - 0.0201and Equation of the standard curve for R2 was y = 0.0182x - 0.0134, and for R3 was y = 0.0186x -0.0197, having the R^2 value 0.9954,0.9904 and 0.9929 respectively, shown in table 4, which is highly statistically significant.

	Replicate 1	Replicate 2	Replicate 3	Average	SD
Slope	0.0187	0.0182	0.0186	0.0185	0.000216025
Y - intercept	-0.0201	-0.0134	-0.0197	-0.01773333	0.003068478
R^2 - value	0.9954	0.9904	0.9929	0.9929	0.002041241

Table 4: Regression parameters of calibration curve.

Compatibility studies using FTIR

FTIR spectroscopy investigations were carried out on clarithromycin drug and optimised NLC formulation. The IR spectrum of the pure clarithromycin showed characteristic peaks at 2889.37 cm^{-1}, 1112.01 cm^{-1}, due to C-H stretching, C-O stretching, and optimised formulation showed the characteristic peaks at 2914.13 cm^{-1} and 1111.02 cm^{-1} similar to the peaks which are generated in the drug. There is no interaction between drug and polymer. So, observation indicates that the drug and lipids are compatible with each other.

Figure 1: FTIR spectra for Clarithromycin.

Drug content study

The % Drug content of Clarithromysin microcapsules were found to be within the range of 17.25568 ± 0.243243 % to 9.601622 ± 0.249645%. % Drug content is more in case of F2 (17.25568 ± 0.243243) where as less in case of F3 (9.601622 ± 0.249645).

Figure 2: FTIR spectra for optimized formulation.

The present study found that the Drug content was increased when the polymer ratio was increased as well. The studies have proved that the Drug content of microcapsules was improved within increasing the concentration of CAP (cellulose acetate phthalate) and Talc shown in table 5.

Drug entrapment efficiency

The entrapment efficiency is a vital parameter that assists in the identification of drug efficacy, and it depends on various concentrations of enteric polymers such as CAP and anti aggrading agent Talc. The average efficiency ranges % CEE is more in case of F2 (94.90622 ± 1.337838) where as less in case of F3 (38.40649 ± 0.998582), as shown in table 5.

Present work found that some formulation of Clarithromycin microcapsules has shown lower entrapment efficiency due to low concentration of polymer for lower range of binding site and binding force. The result shows that the entrapment efficiency of mucoadhesive micro-capsules was increased with increased polymer concentration.

Code	Absorbance at 265 nm	Concentration in µg/ml	Average of % DC	SD of % DC	% TH DC	Average of % CEE
F1	0.272	15.65946	14.06108	0.390201	28.57143	49.21378
F2	0.342	19.44324	17.25568	0.243243	18.18182	94.90622
F3	0.184	10.9027	9.601622	0.249645	25	38.40649
F4	0.231	13.44324	12.35838	0.244859	16.66667	74.15027
F5	0.269	15.4973	14.23946	0.319011	21.05263	67.63743

Table 5: Drug content, Clarithromycin entrapment efficiency study for 100 mg formulations.

Percentage drug release (DR) AT 2 hrs and 10 hrs

All the formulations from F1 to F5 shows a maximum of 9-17% Drug release after 2 hrs followed by a controlled release pattern for more than 10 hrs in PBS medium, because the polymer CAP (cellulose acetate phthalate) was retarding drug release in acidic medium but soluble in buffer medium (pH-7.4) shown in table 6. The drug release increases as the amount of CAP and TALC. % DR shown optimized result at higher concentration of enteric coating polymer. % DR and all other dissolution parameters those are interpreted for fitting dissolution data into different mathematical models were summarized in table 6.

Formulation code	Time in hr.	% DR	Time in hr.	% DR
F1	2	9.6421	10	68.9309
F2	2	12.0917	10	71.5109
F3	2	16.1732	10	52.3935
F4	2	9.7096	10	68.0499
F5	2	17.5886	10	68.4749

Table 6: Average values of drug release F1-F5.

In-vitro dissolution studies of microcapsules

The present study showed that clarithromycin enteric coated microcapsules in most of the formulations were negligible amounts of drug release in simulated gastric fluid (0.1N HCl, pH 1.2); whereas for those formulations were increased amount of drug release in simulated intestinal fluid (pH 7.4-10), as shown in figure 3.

It was found that the percentage of cumulative drug release (CDR %) in the range of 71.5109 ± 0.02% to 52.39 ± 0.01%, respectively.

Figure 3: Drug release profile of all the microcapsule batches.

It was observed that Clarithromycin - CAP in high concentration microcapsules formulation F2 has slower drug release rates. The results showed that the drug release was decreased when the polymer and talc concentration was decrease.

Mechanism of drug release

Clarithromycin microcapsules F2 was selected as the most potential for its drug release kinetics model like zero order, first order, Higuchi and Korsmeyer-Peppas models. The R2 of these models were determined and compared. The result of the curve fitting into various mathematical models was shown in table 7.

From the R^2 value of different mathematical models it is concluded that all the formulation are best fitted to korsemeyer peppas model figure 4. Slope of the Peppas equation indicates their release exponent (n values) from which the mechanism of the drug release is identified. Release exponent (n) of microcapsule F2 shows n value of 0.79, which was define the drug release by diffusion in the hydrated matrix and polymer relaxation method, shown in table 8. This is the perfect characteristic of an enteric coated microcapsule because cap is soluble in PBS (pH = 7.4).

SL. NO.	Formula code	Zero order	First order	Hixson crowell	Higuchi	Korsmeyer peppas
1	F1	0.5239	0.5968	0.5703	0.6601	0.7622
2	F2	0.2378	0.5913	0.5755	0.6582	0.7287
3	F3	0.2404	0.1742	0.1966	0.3659	0.5863
4	F4	0.5916	0.6314	0.6178	0.7065	0.7859
5	F5	0.5309	0.6231	0.5905	0.6616	0.7442

Table 7: R^2 – Values of mathematical models.

$n \leq 0.45$	----Fickian diffusion-controlled drug release
$n \geq 0.89$ --- non-Fickian, zero order release	
$0.45 \leq n \leq 0.89$ ----- by both phenomenon (diffusion in the hydrated matrix and polymer relaxation)	

Table 8: Release exponent values of Korsmeyer peppas.

Figure 4: Dissolution profile of F2 as per Peppars model.

Mathematical and statistics analysis of experimental design

Clarithomycin enteric coated microcapsules were used to derive a statistical equation, ANOVA analysis, contour plots, and 3D response surface plots. Statistical analysis was analyzed according to table 5.

A 2^2 design was employed in triplicate manner and total 15 microcapsules of clarithromycin were prepared. Angle of repose (AR), % Clarithromycin entrapment efficiency (%CEE), % DR at 2hrs (Q2) and at 10hrs (Q10) were considered as four responses for optimization.

Analysis of variance (ANOVA) results for all the responses explored to determine the significant effect of independent variables (CAP and TALC). P-value of CAP and TALC (given in ADX report) revealed that both have significant effect on the response variables. ANOVA table for both the responses along with the design details were given below in ADX report generated by SAS. Normal probability plots were generated to check the model accuracy.

Clarithromycin microcapsules regression equation (1, 2, 3 and 4) showed that positive sign X1 (CAP) illustrates synergistic effect, and indicates that if polymer concentration increases; the value of depended variables (% Drug release, Angle of repose, entrapment efficiency,) is also increases. Negative value of the coefficient of TALC indicates that drug release decreases as the amount of TALC increases. Negative effects of X12 and X22 suggest that as the total amount of polymer increases all depended variables increases slowly. Positive effects of X12 and X22 suggest that as the total amount of polymer increases, all depended variables increases significantly.

Regression equations

Q2 = 17.5886 - CAP (1.0035) +TALC (1.0373) – CAP^2 (5.6844) -(CAP)(TAL)(2.2283)

Q10 = 75.276 + CAP (4.7662) -TALC (2.97) - CAP^2 (7.855) + (CAP)(TALC)(3.6158)

% CEE = 67.6374 + CAP (20.3591) -TALC (7.8908) - CAP^2 (3.4682) -(CAP)(TALC)(2.4872)

AR = 31.974 + CAP (4.9379) -TALC (6.8197) +CAP^2 (4.748) +(CAP)(TALC)(2.9551)

Factorial design of ANOVA analysis, 3d response surface and two-dimensional contour plots for clarithromycin enteric coated microcapsules:

ANOVA analysis was used to response combination formulations and it is also used to identify the formulations significant or insignificant. On other hands, three-dimensional response surface plots were generated for every response to study the performance of the manner and also assisted the main and interaction effects of the independent variables (factors), as well as two-dimensional contour plot provides a visual representation of values of the response.

Table 9 is seen that CAP (cellulose acetate phthalate) and Talc value less than 0.0500 which are achieved statistically significant. Clarithromycin enteric microcapsules were prepared using polymers and Talc as anti aggregating agent was observed significant.

Normal probability plot figure 5-8. of all the four responses depicts that the points are close to a straight line, which supports the accuracy of our chosen model.

Contour plot figure 9 and Response surface plot Figure of all the dependent variable were explored to observed the effect of CAP and TALC on them.

Contour line of Angle of repose (AR) shows that AR decreases as amount of TALC decreases, which justified the antiaggregatory property of TALC. But the reverses observed in contour plot of %CEE. Here %CEE increases with CAP but decreases with TALC. The same as also evident from the corresponding response surface plot.

Q2 and Q10 decreases with increases in TALC. The counter line are curvilinear due to fitter quadrate model.

ANOVA for Drug release at time 2hrs						
Source	**Sum of squares**	**df**	**Mean square**	**F value**	**p-Value Prob > F**	**Significant/Insignificant**
F2	12.08406	1	12.08406	1.637705	0.022697	Significant
F2	12.91025	1	12.91025	1.749676	0.021275	Significant
F2	59.58401	1	59.58401	8.075189	0.016033	Significant
ANOVA for Drug release at time 10hrs						
Source	**Sum of squares**	**df**	**Mean square**	**F value**	**p-Value Prob > F**	**Significant/Insignificant**
F2	272.5971	1	272.5971	13.77902	0.003429	Significant
F2	105.8512	1	105.8512	5.350477	0.041073	Significant
F2	156.8868	1	156.8868	7.930183	0.016786	Significant
ANOVA for Angle of repose						

Source	Sum of squares	df	Mean square	F value	p-Value Prob > F	Significant/Insignificant
F2	292.5947	1	292.5947	26.49901	0.000319	Significant
F2	558.1071	1	558.1071	50.54529	0.0001	Significant
F2	104.7924	1	104.7924	9.490582	0.010459	Significant
ANOVA for DRUG ENTRAPMENT EFFICIENCY						
Source	**Sum of squares**	**df**	**Mean square**	**F value**	**p-Value Prob > F**	**Significant/Insignificant**
F2	12763440	1	12763440	2630.328	0.0001	Significant
F2	2166976	1	2166976	446.5769	0.0001	Significant
F2	606705.5	1	606705.5	125.0317	0.0001	Significant

Table 9: ANOVA Analysis variance for Drug Release At Time 2hr, Time 10hrs, Drug entrapment efficiency and Angle of Repose.

Normal probability plot

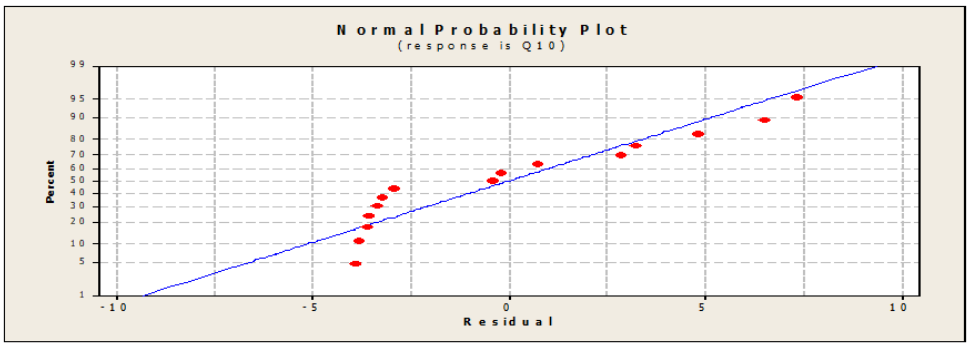

Figure 5: Normal probability plot of Q10.

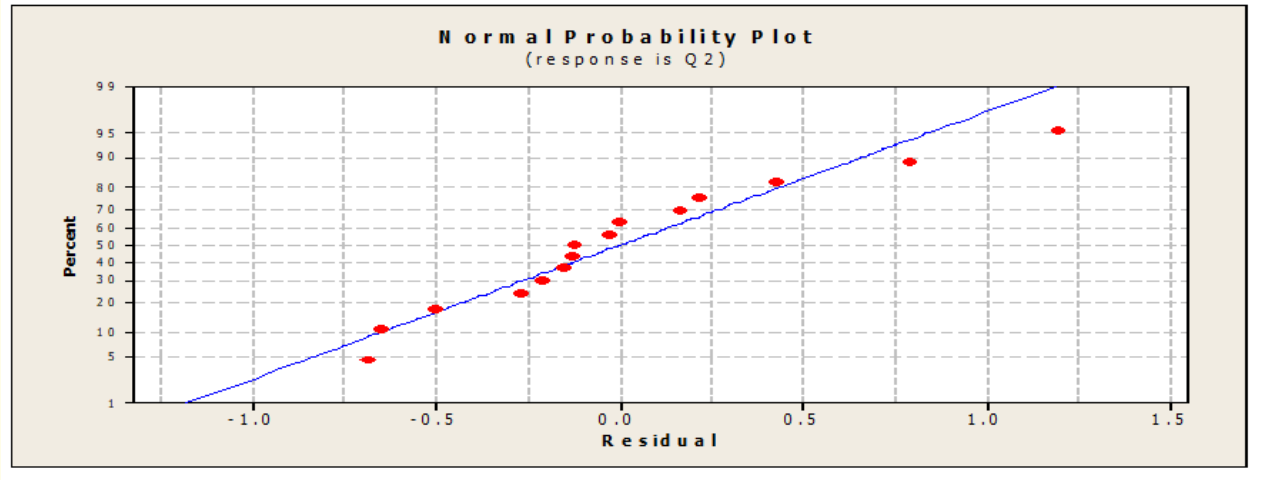

Figure 6: Normal probability plot of Q2.

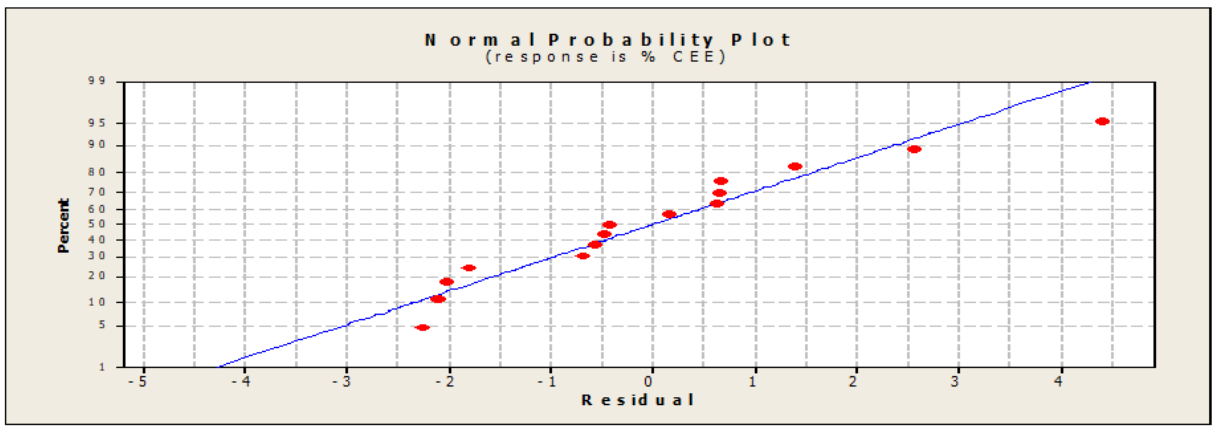

Figure 7: Normal probability plot of %CEE.

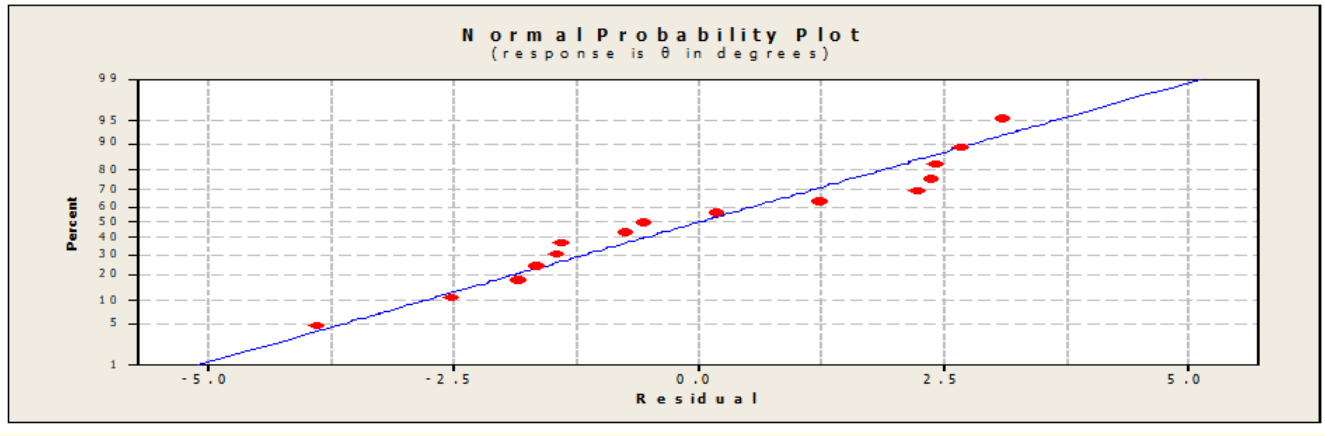

Figure 8: Normal probability plot of AR.

Contour plot

Figure 9: Contour plot for Q2, and Q10.

Figure 10: Contour plot for Q2, and Q10.

Figure 11: Clarithromycin Microencapsules A (Contour Plot) showing the effect of independent variables on independent variables.

Figure 12: Clarithromycin Microencapsules A (Contour Plot) showing the effect of independent variables on independent Variables.

Figure 13: Response plot showing the effect of independent variables on the particle size of mucoadhesive microcapsules.

Figure 14: Response plot showing the effect of independent variables on the particle size of mucoadhesive microcapsules.

Figure 15: Response plot showing the effect of independent variables on the particle size of mucoadhesive microcapsules.

Figure 16: Response plot showing the effect of independent variables on the particle size of mucoadhesive microcapsules.

Optimisation

The amount of Cap and Talc were optimized to get a Q2 value of 10%, since the drug is unstable in acidic medium and to get a hiher % CEE value of 75%. The optimization results were given in table 10.

CAP (Coded value)	TALC (Coded value)	CAP (Actual value)	TALC (Actual value)	AR	Q2	Q10	CEE
1	1	4 gm	1.5 gm	36.845	10.81	74.38	74.25
1	0.5	4 gm	1.25 gm	38.78	11.78	74.06	79.84

Table 10: Table for Optimized data.

Conclusion

The observations made during study and results obtained showed the suitability of the investigated polymers for microencapsulation of Clarithromycin for its sustained release. The Solvent evaporation method was easy to adopt and also to achieve high drug entrapment efficacy. The result observed that Clarithromycin enteric coated microcapsules of Drug release with 2 hrs and 10 hrs, entrapment efficiency, particle flow property, varies with increased drug-polymer concentration and talc as ant aggregative agent. Additionally, the microencapsulated forms of Clarithromycin are also anticipated to have enhanced oral bioavailability, minimized harmful side effects and reduced dosing frequency which would be further helpful to improve patient compliance.

The *in-vitro* drug release studies demonstrated that the drug release was sustained about 14 h, by diffusion in the hydrated matrix and polymer relaxation method which is suitable for enteric coated dosage form of Clarithromycin microcapsules. The results of 2^2 factorial designs revealed that drug and polymer concentration significantly affected dependent variables.

The Clarithromycin microcapsules drug content amount drug release after 10 hrs shows satisfactory results. Microencapsules were optimized for % drug releases after 2 hrs with amount of CAP 4 gm amount of talc 1.25gm (formulation F2) exhibited high entrapment efficiency, the percentage of drug release in intestine shows adequate results. Therefore, one can assume that Clarithromycin enteric coated micro-capsules are promising pharmaceutical forms by providing controlled-release drug delivery systems.

Acknowledgement

We are grateful to Bharat Institute of Technology, Telangana for their support to this research. Dr. Semmachalam Rath reviewed the work and also contributed in the writing of the final version of the manuscript.

Conflict of Interest

The authors declare no conflict of interest.

Bibliography

1. Malhotra-Kumar S., *et al*. "Effect of azithromycin and clarithromycin therapy on pharyngeal carriage of macrolide-resistant streptococci in healthy volunteers: A randomised, double-blind, placebo-controlled study". *Lancet* 369.9560(2007): 482-490.

2. *Pharmaceutical Development and Technology* 6.4(2001): 607-6149.

3. CJ Malm., *et al*. "Preparation of Phthalic Acid Esters of Cellulose". *Journal of Industrial and Engineering Chemistry* (2005): 658.

4. *Iranian Polymer Journal* 14.12 (2005): 1058-1065.

5. Spitael J., *et al*. "Dissolution rate of cellulose acetate phthalate and Brönsted catalysis law". *Pharmazeutische Industrie* 42 (1980): 846-849.

6. Dubey R., *et al*. "Microencapsulation Technology and Applications". *Defence Science Journal* 59.1 (2009): 82-95.

7. Ardenia A and Gupta AK. "Development and optimization of gastroretentive mucoadhesive microspheres using 33 factorial design". *International Journal of Pharmaceutical Sciences and Research* 7.5 (2016): 2020-2030.

8. Abraham S., *et al*. "Preparation, evaluation and *in-vitro* characterization of biopolymer derived hybrid microcapsules for extended release of cefaclor". *International Journal of Chemical and Pharmaceutical Sciences* 5.2 (2014): 145-155.

9. Takenaka H., *et al*. "Preparation of enteric-coated microcapsules for tableting by spray-drying technique and *in vitro* simulation of drug release from the tablet in GI tract". *Journal of Pharmaceutical Sciences* 69 (1980): 1388-1392.

10. Paul B., *et al*. "Formulation and in-vitro evaluation of metronidazole loaded HPMC K15M mucoadhesive microcapsules for H. pylori infection using 32- full factorial designs". *International Journal of Pharmaceutical Sciences and Research* 10.2(2019): 555-567.