www.ThePharmaJournal.com

The Pharma Innovation

ISSN (E): 2277- 7695
ISSN (P): 2349-8242
NAAS Rating: 5.03
TPI 2020; 9(3): 408-420

www.thepharmajournal.com
Received: 10-01-2020
Accepted: 14-02-2020

BVP Deepthi
Assistant Professor, Joginpally B.R. Pharmacy College, Yenkapally, Moinabad, Hyderabad, Telangana, India

N Rachana Kumari
Joginapally B.R. Pharmacy College, Yenkapally, Moinabad, Hyderabad, Telangana, India

V Manisree
Joginapally B.R. Pharmacy College, Yenkapally, Moinabad, Hyderabad, Telangana, India

K Sandhya Rani
Joginapally B.R. Pharmacy College, Yenkapally, Moinabad, Hyderabad, Telangana, India

P Krishna
Joginapally B.R. Pharmacy College, Yenkapally, Moinabad, Hyderabad, Telangana, India

Dr. JVC Sharma
Joginapally B.R. Pharmacy College, Yenkapally, Moinabad, Hyderabad, Telangana, India

Corresponding Author:
BVP Deepthi
Assistant Professor, Joginpally B.R. Pharmacy College, Yenkapally, Moinabad, Hyderabad, Telangana, India

Pulsincap Designing of Rivaroxaban

BVP Deepthi, N Rachana Kumari, V Manisree, K Sandhya Rani, P Krishna and Dr. JVC Sharma

Abstract
The purpose of the present study was to design and evaluate an oral, site specific, pulsatile drug delivery system containing Rivaroxaban as a model drug, which can be time dependent manner, to modulate the drug level in synchrony is a member of the drug class known as statins. It is used for lowering cholesterol based on chrono pharmaceutical considerations. The basic design consists of an insoluble hard gelatin capsule body, filled with powder blend and sealed with a hydrogel plug. The powder blend containing Rivaroxaban, sodium starch glycolate, cross carmellose sodium, microcrystalline cellulose and talc was prepared and evaluated for flow properties and FTIR studies. The prepared formulations were evaluated for drug content, weight variation and *in vitro* release studies. FTIR studies confirmed that there was no interaction between drug and polymers and *in vitro* release studies of pulsatile device revealed that increasing hydrophillic polymer content resulted in delayed release of Rivaroxaban from the pulsincap after a predetermined lag time of 6hrs. Based on *in vitro* studies performed, F8 was found to be optimized formulation.

Keywords: Pulsatile system, time dependent delivery, Rivaroxaban, Chronopharmaceutics, *In vitro* release studies

Introduction
Oral controlled drug delivery systems represent the most popular form of controlled drug delivery systems for the obvious advantages of oral route of drug administration [1]. A pulsatile release profile is characterized by a lag time followed by rapid and complete drug release [2] related to the circadian rhythm of the body [3]. Pulsatile release systems release the drug with constant or variable release rates as per the need. These dosage forms offer many advantages, such as nearly constant drug level at the site of action, prevention of peak-valley fluctuations, reduction in dose of drug, reduced dosage frequency, avoidance of side effects, and improved patient compliance, however, there are certain conditions for which such a release pattern is not desirable. These conditions demand release of drug after a lag time. In other words, it is required that the drug should not be released during the initial phase of dosage form administration. Such a release pattern is known as time controlled or pulsatile release [4]. Pulsatile drug delivery systems are generally classified into time-controlled and site-specific delivery systems. The release from the first group is primarily controlled by the system, while the release from the second group is primarily controlled by the biological environment in the gastro-intestinal tract such as pH or enzymes [5]. Pulsatile release systems can be classified into multiple-pulse and single-pulse systems [6]. Pulsatile drug release system, which allows the release of active pharmaceutical material in single or successive pulses at precise and well-controlled time periods, is a recently developed drug delivery system [7]. Drugs are usually encapsulated in one way or another within a barrier material, which is composed of an erodible or biodegradable polymer [8]. Depending on the barrier material structure and thickness, different release lag times can be achieved [9]. After the barrier material is dissolved, eroded or degraded, drugs are rapidly released from the inner reservoir core [10].
Rivaroxaban (Xarelto®) is an oral oxazolidinone-based anticoagulant agent. It inhibits not only free factor Xa with high selectivity but also prothrombinase bound and clot-associated factor Xa in a concentration-dependent manner. It is a potent, selective direct inhibitor of factor Xa that is used in the prevention of venous thromboembolism (VTE) in adult patients after total hip replacement (THR) or total knee replacement (TKR) surgery. The recommended dose of Xarelto is 10 mg taken orally once daily. For a 10 mg dose, the oral bioavailability of Rivaroxaban is high (80–100%) and is not affected by food intake. These pharmacological properties underpin the use of Rivaroxaban in fixed dosing regimens, with no need for dose adjustment or routine coagulation monitoring [11].

The purpose of the present study was to design and evaluate an oral, site specific, pulsatile drug delivery system containing Rivaroxaban as a model drug, which can be time dependent manner, to modulate the drug level in synchrony is a member of the drug class known as statins. It is used for lowering cholesterol based on chronopharmaceutical considerations.

Materials and methods

Rivaroxaban pharma grade was obtained from spectrum labs, Hyderabad. Sodium starch glycolate and crosscarmellose sodium, hydrochloric acid, methanol was obtained from S.d fine chemicals limited, Mumbai. Microcrystalline cellulose, magnesium stearate, talc was obtained from loba chemie private limited. Ethyl cellulose and HPMC was obtained from Otto chemicals, Mumbai. Formladehyde, potassium permanganate, potassium dihydrogen phosphate, sodium hydroxide pellets was obtained from Qualigens fine chemicals, Mumbai. All other chemicals, reagents and solvents were used are of analytical grade.

A. Drug-excipient compatibility studies

To know the chemical compatibility of the drug spectroscopic technique that is FTIR studies were used. The FTIR spectra were recorded using an IR spectrophotometer (IR-Affinity-1, Shimadzu, Japan). The IR spectra for the samples were obtained by KBr disk method. The samples were prepared by grinding the pure drug, polymer and physical mixture with KBr separately. The pellets of drug and potassium bromide were prepared by compressing the powders at 20 psi for 10 min on KBr-press and the spectra were scanned in the wave number range of 4000- 600 cm^{-1}.FTIR study was carried on Rivaroxaban, physical mixture of Rivaroxaban and for the best formulation.

B. Flow properties of API

i. **Bulk Density (DB):** It is the ratio of total mass of powder to the bulk volume of powder. It was measured by pouring the weighed powder (passed through standard sieve#20) into a measuring cylinder and the initial volume was noted. This initial volume is called the bulk volume. From this, the bulk density is calculated according to the formula mentioned below. It is expressed in g/cc and is given by:

Db = m/Vo

Where,
m = mass of the powder
Vo = bulk volume of powder

ii. **Tapped density (Dt):** It is the ratio of total mass of powder to the tapped volume of powder. The volume was measured by tapping the powder for 500 times. Then the tapping was done for 750 times and the tapped volume was noted (the difference between the two tapped volumes should be less than 2%). If it is more than 2%, tapping is continued for 1250 times and tapped volume was noted. It is expressed in g/cc and is given by:

Dt = m/Vt

Where,
m = mass of the powder
Vt = tapped volume of powder

iii. **Angle of Repose (θ):** This is the maximum angle possible between the surface of a pile of powder or granules and the horizontal plane. The powders were allowed to flow through the funnel fixed to a stand at definite height (h). The angle of repose was then calculated by measuring the height and radius of the heap of granules formed.

Tan θ= h/r

(Or)
$\theta = \tan^{-1}(h/r)$
Where,
θ = angle of repose
h = height of the heap
r = radius of the heap

iv. **Compressibility Index:** The flowability of powder can be evaluated by comparing the bulk density (Db) and tapped density (Dt) of powder and the rate at which it packed down. Compressibility index is calculated by:

Compressibility index (%) = Dt – Db/Dt x 100

Where,
Db = Bulk density
Dt = Tapped density

v. **Hausner's Ratio:** It is the ratio of tapped density to the bulk density. It is given by:

Hausner's ratio = Dt / Db

Where,
Dt = Tapped density
Db = Bulk density

The flow properties of API alone and along with excipients i.e., powder blend was calculated by using the above formulae and the type of flow can be compared by using the following standard specifications in Table-1.

Table 1: Standard specifications for comparison of flow properties

S. No	Flow property	Angle of repose	Carr's Index	Hausner's ratio
1.	Excellent	25-30	<10	1.00-1.11
2.	Good	31-35	11-15	1.12-1.18
3.	Fair	36-40	16-20	1.19-1.25
4.	Passable	41-45	21-25	1.26-1.36
5.	Poor	46-55	26-31	1.35-1.45
6.	Very poor	56-65	32-37	1.46-1.59
7.	Very very poor	>66	>38	>1.6

Pulsincap designing

Designing or preparation of pulsincap capsules involves 3 steps:

A. Preparation of cross-linked gelatin capsule.
B. Preparation of powder blends for filling into capsules.
C. Formulation of pulsincap of Rivaroxaban.

A. Preparation of cross-linked gelatin capsule

Formaldehyde treatment

About 100 hard gelatin capsules size '0' were taken. Their bodies were separated from the caps and placed on a wire

mesh. The bodies which were placed on a wire mesh were spread as a single layer. 25 ml of 15% v/v of formaldehyde solution was prepared and placed in a desiccators. To this 5 g of potassium permanganate was added. The wire mesh containing the bodies of the capsules was kept on the top of desiccators' containing formaldehyde liquid at the bottom in equilibrium with its vapor and immediately the desiccators' was tightly closed and sealed. The bodies of capsules were made to react with formaldehyde vapors by exposing them for varying periods of time *viz*., 2, 4, 6, 8, 10hrs. Then they were removed and kept on a filter paper and dried for 24 hrs to ensure completion of reaction between gelatin and formaldehyde vapors, afterwards the capsules were kept in an open atmosphere, to facilitate removal of residual formaldehyde. These capsule bodies were capped with untreated cap and stored in a polythene bag [12, 13].

Use of Formaldehyde treatment

The main aim of formaldehyde treatment was to modify the solubility of hard gelatin capsules. Cross-linking of gelatin molecules was achieved by exposing to formalin vapors. Cross-linking involves the reaction of amino groups in gelatin molecular chain with aldehyde groups of formaldehyde by a "Schiff's base condensation" so that the gelatin becomes water insoluble. Formaldehyde reacts with gelatin forming an irreversible complex. The primary amine group present in gelatin reacts with formaldehyde making it irreversibly bound. Potassium permanganate was added to formaldehyde solution so that formalin vapors were produced. When bodies of hard gelatin capsule were exposed to formaldehyde vapors for different periods of time in a closed dessicator, vapor gets equilibrated with formaldehyde liquid and therefore makes the gelatin water insoluble [14, 15].

Evaluation of formaldehyde treated capsules

Physical Tests

- **Identification attributes:** Suitable size capsules which are lockable were selected. Generally the gelatin capsules when touched with wet hand they become sticky but upon formaldehyde treatment the capsules are observed for the stickiness.
- **Visual defects:** Selected 100 treated capsules and observed for visual defects by physical observation and not more than 15-20 capsules must be distorted.
- **Dimensions:** Variations in the dimensions between the formaldehyde treated and untreated capsules were studied. The length and diameter of the capsules were measured before and after formaldehyde treatment by using vernier calipers [16, 17].

Optimization of formaldehyde treated capsule bodies exposed at various time intervals *viz*., 2, 4, 6, 8, 10hrs

Formaldehyde treated capsule bodies which were exposed at various time intervals *viz*., 2, 4, 6, 8, 10hrs were optimized by conducting disintegration test. The test was performed on both untreated and treated capsules. Formaldehyde treated bodies joined with untreated caps and was tested for disintegration. Disintegration test was carried out by using Hiccon disintegration test apparatus. pH 1.2, pH 6.8, buffers were used as medium and maintained at 37°C throughout the experiment. The time at which the capsules disintegrate are noted [18].

B. Preparation of rivaroxaban tablet for filling into capsules

All the ingredients were passed through # 60 mesh sieve separately. The drug & MCC were mixed by adding small portion of each at a time and blending it to get a uniform mixture and kept aside.

Then the other ingredients were mixed in geometrical order and passed through coarse sieve (#44 mesh) and the tablets were compressed using hydraulic press. Compression force of the machine was adjusted to obtain the hardness in the range of 3-4 kg/cm2 for all batches. The weight of the tablets was kept constant for all formulations F1 to F6 (100 mg).

Table 2: Formulae for preparation of blend for filling of Rivaroxaban pulsincap

Ingredients (mg)	F1	F2	F3	F4	F5	F6
Drug	10	10	10	10	10	10
SSG	2	4	6	--	--	--
CCS	--	--	--	2	4	6
MCC	84	82	80	84	82	80
Mg. sterate	2	2	2	2	2	2
Talc	2	2	2	2	2	2
Total	100	100	100	100	100	100

Formulation of pulsincap of rivaroxaban

The modified release pulsincaps containing 10mg of Rivaroxaban were prepared by using different excipients and polymers in varying ratios. The formaldehyde treated capsule bodies which were exposed to 6 hrs was optimized and chosen for the pulsincap formulation based on disintegration time. Optimized formulation of rivaroxaban tablet was filled into the capsule body. For hydrogel plug formulation, the plug was prepared by using the combination of Ethyl cellulose: HPMC in varying ratios. Initially the total weight of the plug was taken as 100 mg and the ratio of hydrophobic & hydrophilic polymer as 1:1, 1:2, and 2:1.

Method of preparation of Pulsincap dosage form

I. Preparation of powder blend

- Hard gelatin capsules of 'size 0' which were hardened with formaldehyde treatment for 6hrs were chosen for the formulation. The bodies and caps separated manually. Optimized formulation F3 was fitted at the bottom of the capsule body.

II. Preparation of Hydrogel plug

- Plug was prepared as a compressed tablet and placed at the opening of capsule body. The capsule body was closed by a cap.
- Hydrogel plug was prepared by using different polymers like Ethyl cellulose, HPMC at different concentrations.
- A combination of hydrophobic and hydrophilic polymers were used *viz*.,
- Ethyl cellulose: HPMC, in different ratios like 1:1, 1:2, and 2:1.
- A tight fit between the plug and impermeable capsule shell is essential to regulate water penetration into the capsule content and the drug release prior to complete erosion of plug material. Ideally plug should erode only from the surface exposed to the release medium.
- Plug ejection can be done by swelling on contact with aqueous fluids (or) pushing out by increased internal pressure (or) erosion (or) by enzyme degradation.

I.

III. Capsule filling

- Homogeneous mixture of drug and excipients were filled into the 6th hr formaldehyde treated capsule body manually by filling method.
- Then, hydrogel plug in the form of a tablet is placed above the mixture i.e., at the opening of capsule body
- The capsule body was closed by a cap.

IV. Capsule sealing

- The joint of the treated capsule body and untreated cap of the capsules was sealed with a small amount of 5% ethyl cellulose ethanolic solution.

Evaluation of tablets

Tablet Dimensions

Thickness and diameter were measured using a calibrated vernier caliper. Three tablets of each formulation were picked randomly and thickness was measured individually.

Hardness

Hardness indicates the ability of a tablet to withstand mechanical shocks while handling. The hardness of the tablets was determined using Monsanto hardness tester. It is expressed in kg/cm^2. Three tablets were randomly picked and hardness of the tablets was determined [19].

Friability test

The friability of tablets was determined by using electrolab friabilator. It is expressed in percentage (%). Ten tablets were initially weighed (WI) and transferred into friabilator. The friabilator was operated at 25 rpm for 4 minutes or run up to 100 revolutions. The tablets were weighed again (WF). The % friability was then calculated by –

%F = 100 (1-WI/WF)

% Friability of tablets less than 1% was considered acceptable.

Weight Variation Test

Ten tablets were selected randomly from each batch and weighed individually to check for weight variation. A little variation was allowed in the weight of a tablet according to U.S. Pharmacopoeia. The following percentage deviation in weight variation was allowed [20].

Average weight of a tablet	Percentage deviation
130 mg or less	±10
>130mg and <324mg	±7.5
324 mg or more	±5

In all formulations, the tablet weight is 100 mg, hence 10% maximum difference allowed.

Test for Content Uniformity

Tablet containing 10mg of drug was dissolved in 50ml of 6.8 pH buffer in volumetric flask. The drug was allowed to dissolve in the solvent. The solution was filtered, 2ml of filtrate was taken in 10ml of volumetric flask and diluted up to mark with distilled water and analyzed spectrophotometrically at 247nm. The concentration of Rivaroxaban was obtained by using standard calibration curve of the drug. Drug content studies were carried out in triplicate for each formulation batch.

***In vitro* disintegration time**

Tablet was added to 900ml of distilled water at 37±0.5ºC. Time required for complete dispersion of a tablet was measured [21].

***In vitro* dissolution study**

In vitro dissolution of Rivaroxaban tablets was studied in USP XXIV dissolution test apparatus. 900ml Phosphate buffer 6.8 (simulated fluid) was used as dissolution medium. The stirrer was adjusted to rotate at 50RPM. The temperature of dissolution medium was maintained at 37±0.5ºC throughout the experiment. One tablet was used in each test. Samples of dissolution medium (5ml) were withdrawn by means of syringe fitted with pre-filter at known intervals of time and analyzed for drug release by measuring the absorbance at 247nm. The volume withdrawn at each time interval was replaced with fresh quantity of dissolution medium. Cumulative percent rivaroxaban released was calculated and plotted against time [22].

Evaluation of pulsincap dosage form

***In vitro* release studies**

Dissolution study was carried out to measure the release rate of the drug from the pulsincap formulation. Invitro dissolution profile of each formulation was determined by employing USP I apparatus by rotating basket method. In order to stimulate the pH changes along GI tract 2 different dissolution media with pH 1.2, 6.8, 2 buffers were sequentially used, and therefore referred to as “Sequential pH change method”. The dissolution media were maintained at a temperature of 37 ± 0.5ºC throughout the experiment and the speed of rotation of basket maintained at 50 rpm. 900ml of dissolution medium was used at each time. Rivaroxaban pulsincaps was placed in basket in each dissolution vessel to prevent floating. While performing experiments, stimulated gastric fluid (SGF) pH 1.2 buffer was first used for 2 hrs (since the average gastric emptying time is 2hrs) and then removed and the fresh stimulated intestinal fluid (SIF) pH 6.8 buffer was added and used for remaining hours. 5 ml samples of dissolution fluid were withdrawn at predetermined time intervals with the help of a syringe. The volume withdrawn at each time interval was replaced with 5ml of fresh dissolution medium maintained at same temperature. The filtered samples were suitably diluted whenever necessary and assayed for Rivaroxaban by measuring absorbance at 247 nm, by UV absorption spectroscopy. %CDR was calculated over the sampling times [23, 24, 25].

Release kinetics

In the present study, data of the *in vitro* release were fitted to different equations and kinetic models to explain the release kinetics of Rivaroxaban from the pulsincap system. The kinetic models used were Zero order equation, First order, Higuchi release and Korsmeyer-Peppas models [26].

The results of *in vitro* release profiles obtained for the pulsincap system were fitted into four models of data treatment as follows [27].

1. Cumulative percent drug released versus time (zero order kinetic model).
2. Log cumulative percent drug remaining versus time (first- order kinetic model).
3. Cumulative percent drug released versus square root of time (higuchi’s model).

4. Log cumulative percent drug released versus log time (korsmeyer – Peppas) equation.

Results and Discussion

Preformulation studies

Rivaroxaban was found to be soluble in 6.8pH buffer and soluble in methanol.

Drug-Excipient compatibility studies: The IR spectrum of pure drug was found to be similar to the standard spectrum of Rivaroxaban. The spectrum of Rivaroxaban shows the following functional groups at their frequencies shown in Figure-1.

From the spectra of Rivaroxaban, combination of Rivaroxaban with polymers, it was observed that all characteristic peaks of Rivaroxaban were not altered and present without alteration in the combination spectrum, thus indicating compatibility of the drug and polymers. FTIR spectra of Rivaroxaban, and optimized formulation are shown in Figure -1 and 2 respectively.

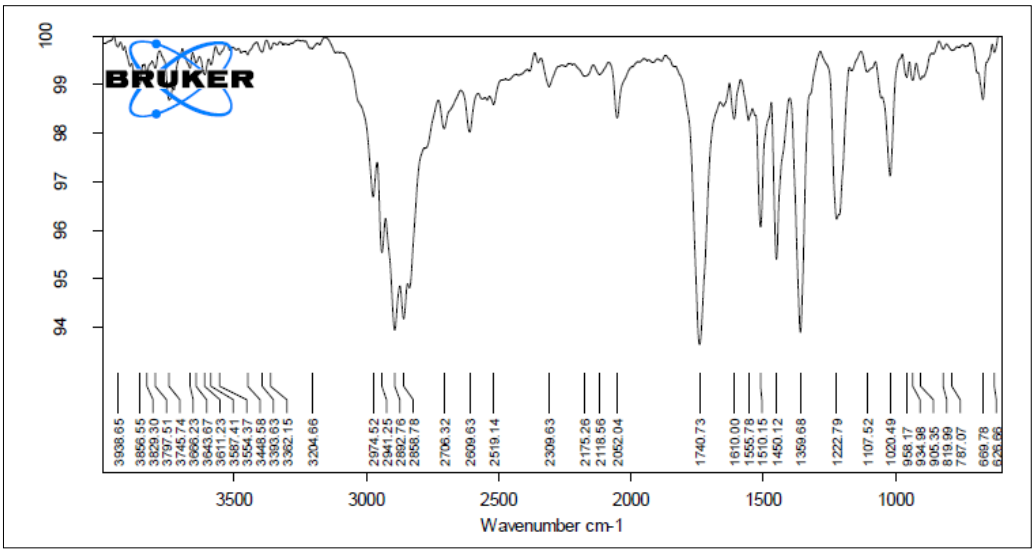

Fig 1: FTIR spectrum of Rivaroxaban

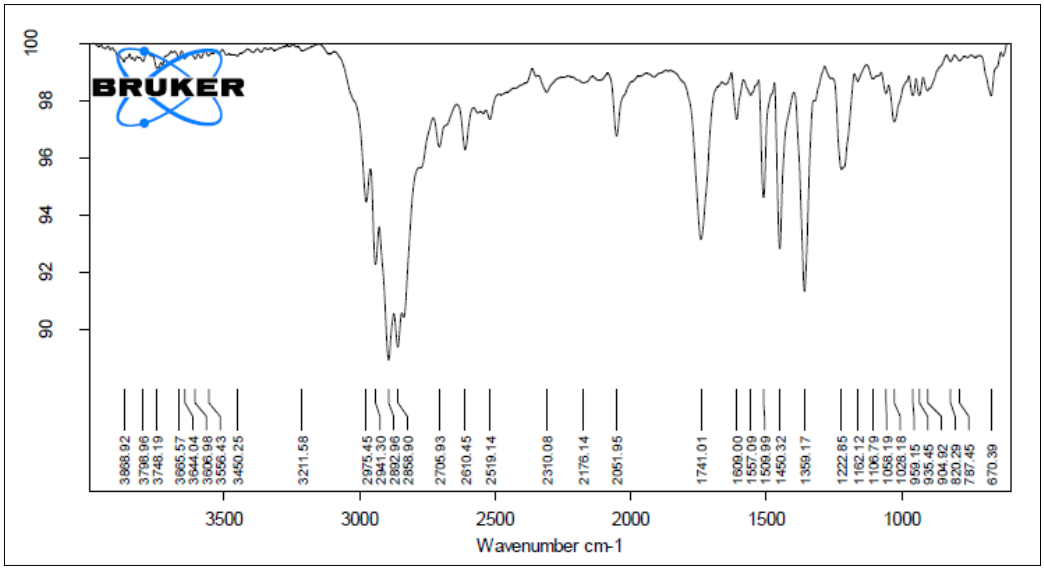

Fig 2: FTIR Spectrum of optimised formulation

Chemical interaction between drug and the polymeric material was studied by using FTIR. There was no difference between the IR patterns of Rivaroxaban, physical mixture of Rivaroxaban and Rivaroxaban optimized formulation.

λ_{max} Determination of Rivaroxaban

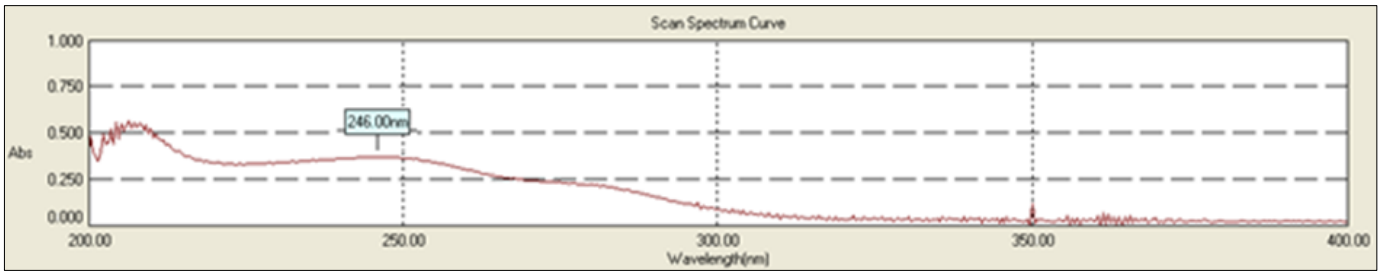

Fig 3: λ_{max} Determination of Rivaroxaban

Standard Calibration Curve

The standard calibration curve of Rivaroxaban was developed in different pH media such as pH 1.2, and pH 6.8 phosphate buffer. Two buffers were selected in order to mimic the *in-vivo* conditions of the GIT.

a. Standard Calibration Curve in 1.2 pH

Standard graph of Rivaroxaban showed linearity at the concentration range of 5-30µg with correlation coefficient of 0.999. Table-3 gives the data of the standard graph and Figure-4 shows the standard graph in pH 1.2.

Table 3: Data for calibration curve of Rivaroxaban in pH 1.2 at 247nm

Concentration (µg/mL)	Absorbance
0	0
5	0.117
10	0.245
15	0.371
20	0.482
25	0.608
30	0.712

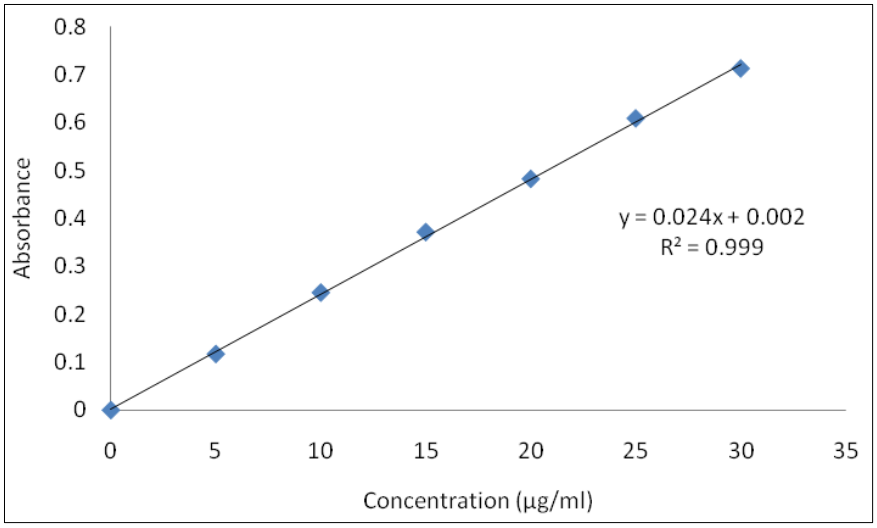

Fig 4: Standard Calibration Curve of Rivaroxaban in pH 1.2 at 247 nm

b. Standard Calibration Curve in 6.8 pH phosphate buffer

Standard graph of Rivaroxaban in pH 6.8 phosphate buffer shows linearity in the concentration range of 5-30 µg with correlation coefficient of 0.999. Table-4 gives the data of the standard graph and Figure-5 shows the standard graph in pH 6.8 phosphate buffer.

Table 4: Data for calibration curve of Rivaroxaban in pH 6.8 at 247nm

Concentration (µg/mL)	Absorbance
0	0
5	0.153
10	0.329
15	0.477
20	0.624
25	0.789
30	0.928

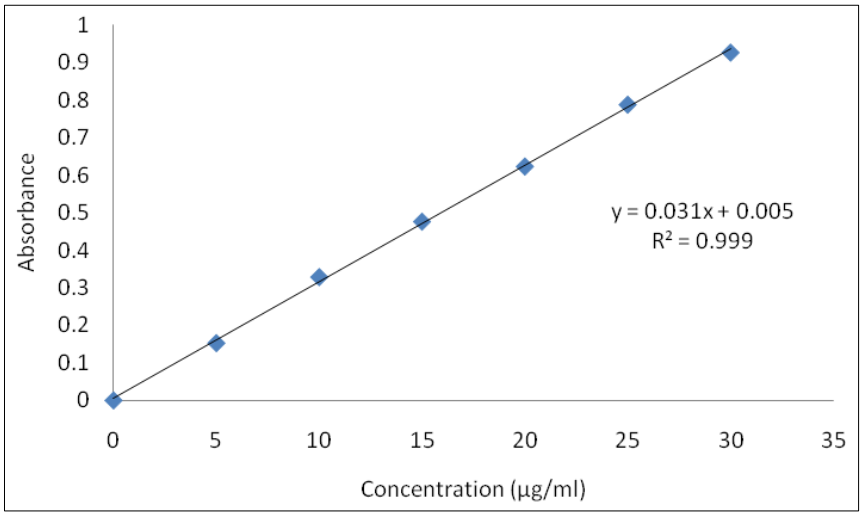

Fig 5: Standard Calibration Curve of Rivaroxaban in pH 6.8at 247 nm

Flow properties of powder blend

Table 5: Flow properties of powder blend

Formulation Code	Angle of Repose±SD	Bulk Density (g/ml)±SD	Tapped Density (g/ml)±SD	Carr's Index. (%)±SD	Hausner's ratio±SD
F1	29.84±0.16	0.395±0.15	0.468±0.86	15.60±0.02	1.18±0.62
F2	26.49±0.24	0.387±0.23	0.475±0.24	18.53±0.52	1.23±0.59
F3	29.43±0.85	0.395±0.64	0.467±0.15	15.42±0.98	1.18±0.18
F4	28.51±0.63	0.375±0.78	0.451±0.39	16.85±0.36	1.20±0.63
F5	29.12±0.21	0.389±0.26	0.459±0.50	15.25±0.42	1.18±0.42
F6	27.46±0.14	0.384±0.94	0.462±0.16	16.88±0.15	1.20±0.15

The angle of repose of different formulations was ≤ 29.84 which indicates that material had good flow property. So it was confirmed that the flow property of blends were free flowing. The bulk density of blend was found between 0.380g/cm^3 to 0.395g/cm^3.Tapped density was found between 0.451g/cm^3 to 0.475g/cm^3.These values indicate that the blends had good flow property. Carr's index for all the formulations was found to be between 15.25-18.53 and Hausner's ratio from 1.18-1.23 which reveals that the blends have good flow character.

Characterization of tablets

Post Compression parameters

All the batches of tablet formulations were characterized for official evaluation parameters like weight variation, hardness, friability, tablet thickness and drug content and results are shown in the table.

Table 6: Characterization Rivaroxaban Tablets

Formulation	%Weight variation	Thickness	Diameter	Hardnes	Friability	Disintegrating	Drug content
F1	0.365	2.13±0.	8.06±0.	3.56±0.	0.054±0.	52.02±0.	96.45±0.
F2	0.298	2.09±0.	8.04±0.	3.49±0.	0.063±0.	39.48±0.	98.05±0.
F3	0.145	2.14±0.	8.06±0.	4.02±0.	0.058±0.	32.15±0.	98.17±0.
F4	0.265	2.06±0.	8.09±0.	4.32±0.	0.012±0.	53.46±0.	99.42±0.
F5	0.948	2.05±0.	8.05±0.	3.89±0.	0.023±0.	49.23±0.	98.06±0.
F6	0.657	2.04±0.	8.01±0.	4.03±0.	0.078±0.	38.12±0.	97.43±0.

Hardness of the tablet was acceptable and uniform from batch to batch variation, which was found to be 3-4 kg/cm^2.All the formulations passed the weight variation test as the % weight variation was within the pharmacopoeial limits of the tablet weight. Friability values were found to be less than 1% in all the formulations F1 –F6 and considered to be satisfactory ensuring that all the formulations are mechanically stable. The drug content values for all the formulations (F1-F6) was found to be in the range of 96.45 – 99.42%.

Dissolution studies of the tablets

The prepared tablets were subjected to dissolution studies in order to know the amount drug release.

Table 7: % Cumulative drug release of formulations F1-F6

Time (min)	F1	F2	F3	F4	F5	F6
0	0	0	0	0	0	0
5	17.45	27.82	36.55	11.18	21.27	28.64
10	27.55	36.82	45.82	26.45	33.82	40.09
15	36.55	48.55	50.45	28.64	43.09	48.27
20	44.73	61.09	69.55	34.09	59.45	64.09

30	52.09	72.82	79.09	45.55	71.18	73.91
45	67.64	83.18	88.36	53.45	79.36	81.82
60	82.09	89.45	98.45	63.82	86.45	91.91

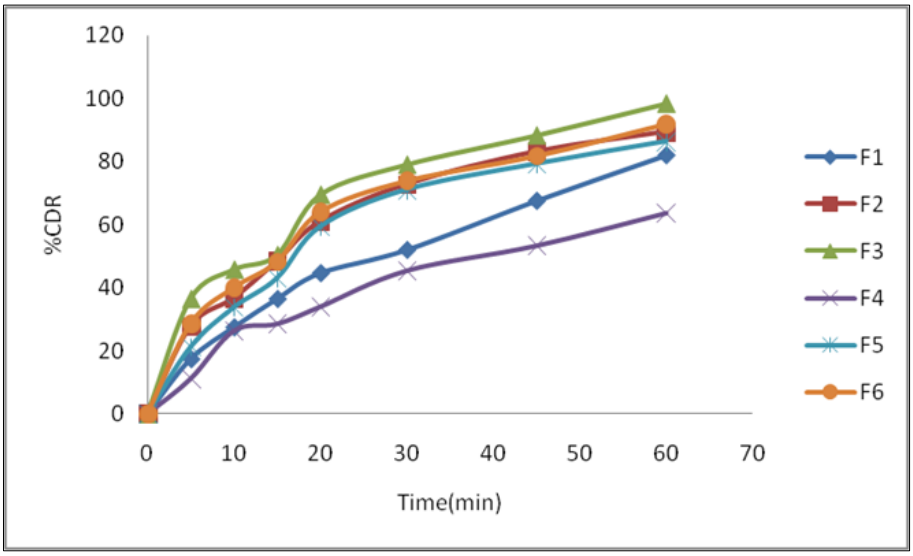

Fig 6: *In vitro* drug release of formulations F1-F6

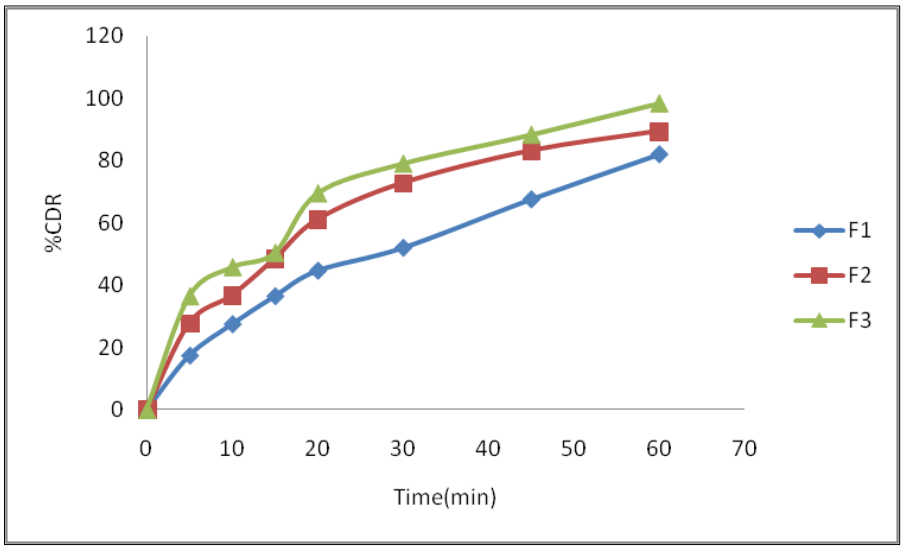

Fig 7: *In vitro* drug release of formulations F1-F3

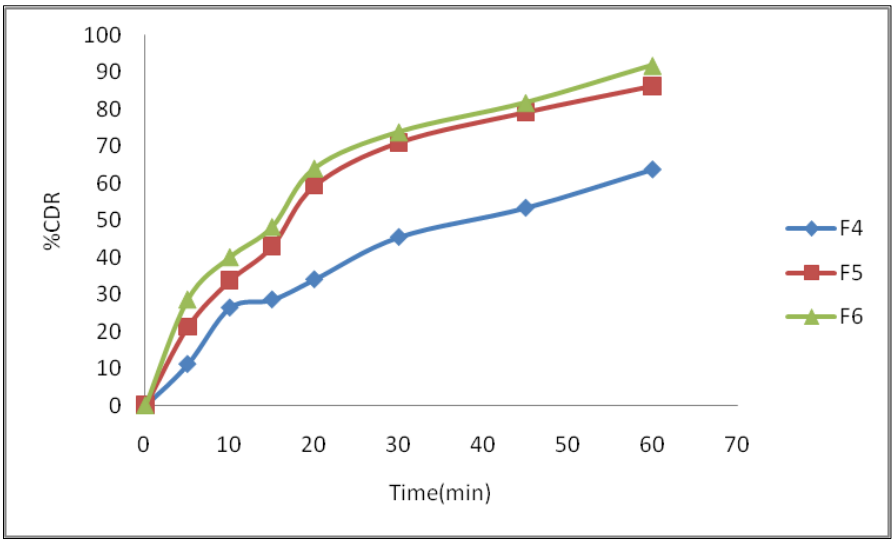

Fig 8: *In vitro* drug release of formulations F4-F6

From the *in vitro* drug release in studies it was observed that the formulations containing SSG as a super disintegrant in different concentrations like 2,4, and 6%, reveals that the increased in the super disintegrant concentration decreases the drug release time and the F3 formulation containing SSG 6% concentration shows maximum amount of drug release (98.45%) at the end of 60mins.

Whereas formulations containing CCS as a super disintegrant in different concentrations like 2,4, and 6%, reveals that the increased in the super disintegrant concentration decreases the drug release time and the F6 formulation containing CCS with 6% concentration shows maximum amount of drug release (91.91%) at the end of 60mins.

So, F3 formulation containing 6% concentration of SSG shows maximum release within 60mins so that it is choosen as optimized formulation.

Evaluation of formaldehyde treated capsules

Physical tests

- **Identification attributes:** The size '0' capsules chosen were opaque, with white colored body and red cap. The normal capsule bodies were soft and sticky when touched with wet hand. After treating with formaldehyde, there were no significant changes in the physical appearance of the capsules except for the stickiness. The body of capsule was hard and non-sticking even when touched with wet hand due to treatment with the formaldehyde.
- **Visual defects:** Among 100 capsules body which were treated with formaldehyde, about 15 to 20 capsule bodies showed visual defects. They were found to be shrunk and distortion into different shapes due to the complete loss of moisture.

Dimensions: Dimensional examination was done by using vernier calipers.

Average capsule length

- Before formaldehyde treatment (untreated cap and body): 20.7mm
- After formaldehyde treatment(treated body and untreated cap): 19.8mm

Average diameter of capsule body

- Before formaldehyde treatment: 7.3 mm
- After formaldehyde treatment: 6.9 mm

Average length of capsule body

- Before formaldehyde treatment: 17.9 mm
- After formaldehyde treatment: 17.2 mm

On formaldehyde treatment, the "0" size capsules bodies showed a significant decreases in length and diameter and attained hardness.

Chemical test

- **Qualitative test for free formaldehyde:** The formaldehyde treated capsules were tested for the presence of free formaldehyde by comparing color of sample solution with standard solution. It was found that the sample solution was not more intensity colored than the standard solution inferring that less than 20µg/ml of free formaldehyde was present in 25 capsule bodies.
- Limit test for the presence of residual formaldehyde, indicated that the amount of formaldehyde present in treated capsules was well within limits.

Optimization of formaldehyde treated capsule bodies exposed at various time intervals *viz*., 2, 4, 6, 8, 10hrs

Table 8: Disintegration test for Treated Capsules

Code	Disintegration Time (hrs)	
	1.2 pH (2hrs)	6.8 pH (upto 24hrs)
C1 (2rd hr)	2	_
C2 (4th hr)	2	1
C3 (6th hr)	2	5
C4 (8th hr)	2	7
C5 (10th hr)	2	12

Basing on the disintegration studies, it was observed that the 6th hr treated capsule (C4) remained intact for 7 hrs so lag time was maintained. C4, C5 remain intact for 9, 12 hrs respectively and therefore they were not selected for the formulation because the required lag time was 6hrs. As the required lag time is 6hrs, C4 (6th hr treated capsule) was selected as optimized time for formaldehyde treatment for further studies.

1. *In vitro* release studies

Dissolution study was carried out to measure the release rate of drug from prepared pulsincap formulation using USP I dissolution apparatus at 37^0C using 2 different dissolution media of pH 1.2, pH 6.8 phosphate buffers in order to mimic *in vivo* GIT conditions. Initially first 2hrs of dissolution was conducted in pH 1.2 buffer, followed by 10hrs of dissolution study in pH 6.8 phosphate buffer.

Table 9: *In vitro* dissolution data of formulations F7 to F9

Time (hrs)	F7	F8	F9
0	0	0	0
1	2.59	0.28	0.82
2	8.46	0.94	3.49
3	12.49	1.52	9.27
4	19.86	3.79	16.48
5	38.46	5.08	29.53
6	67.23	9.21	49.83
7	89.62	82.69	82.46
8	96.48	98.46	97.42
9	99.01		
10			

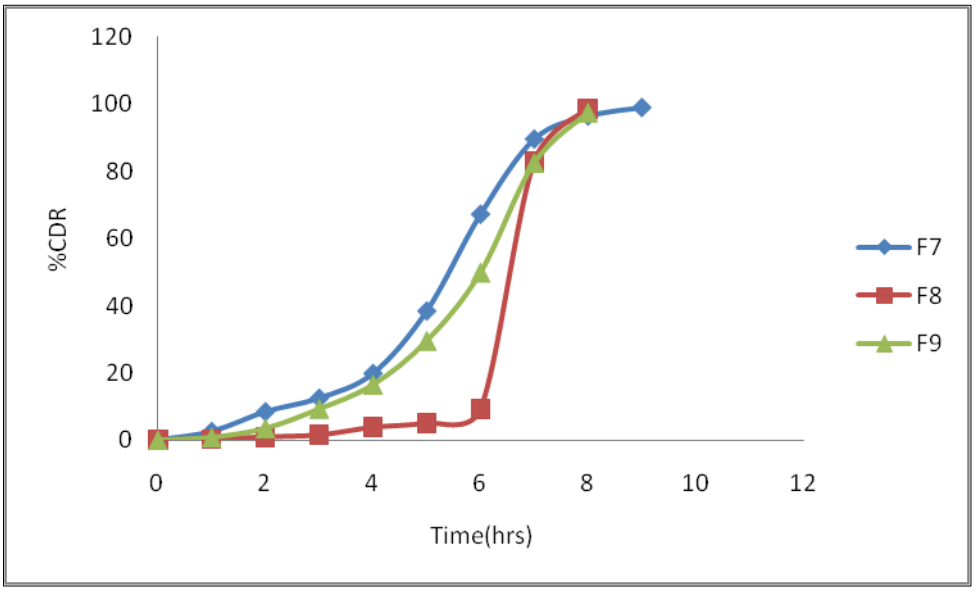

Fig 9: Dissolution plots for formulations F7 to F9

All the 3 formulations of Rivaroxaban pulsincaps were subjected to dissolution studies. Formulations F7, F8, F9 contain the hydrogel plug with combination of hydrophobic polymer and hydrophilic polymer i.e., ethyl cellulose: HPMC in the ratio of 1:1, 2:1 & 1:2 of total 100mg weight of the plug.

It was observed that a proper lag time of 6 hours was maintained with minimal upper GIT drug release for the combination of Ethyl cellulose and HPMC K15M hydrogel plug in the 1:2. It was observed that as the concentration of hydrophilic polymer was increased the release rate of drug was delayed and finally burst release of drug from the formulation occurred after lag time. So basing on these observations, of all the 3 pulsincap formulations, F8 formulation containing hydrogel plug of ethyl cellulose & HPMC K15M in 1:2 ratio was selected as optimized pulsincap formulation.

Release kinetics

Dissolution data was fitted in Zero order, First order, Higuchi's and koresmayer peppas equations. The regression coefficient "R" values for zero order, first order, higuchi's and peppas for formulation F8 was found to be 0.600, 0.501, 0.400 and 0.799 respectively.

Table 10: Correlation coefficient "R" values of F8 optimized formulation

Models	R values
Zero order	0.600
First order	0.501
Higuchi	0.400
Koresmayer peppas	0.799

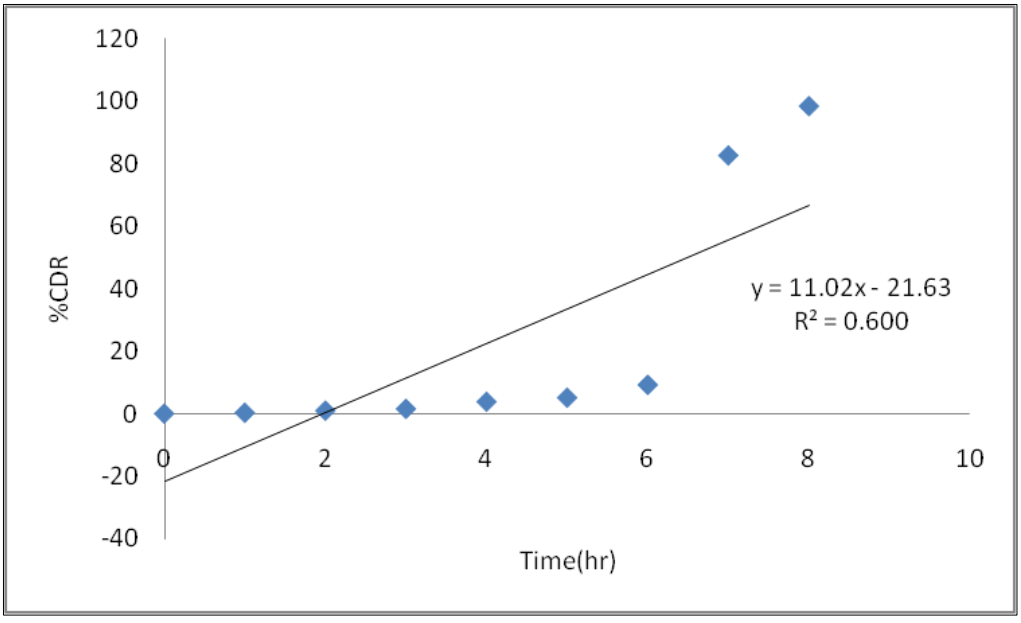

Fig 10: Zero order plot for optimized formulation F8

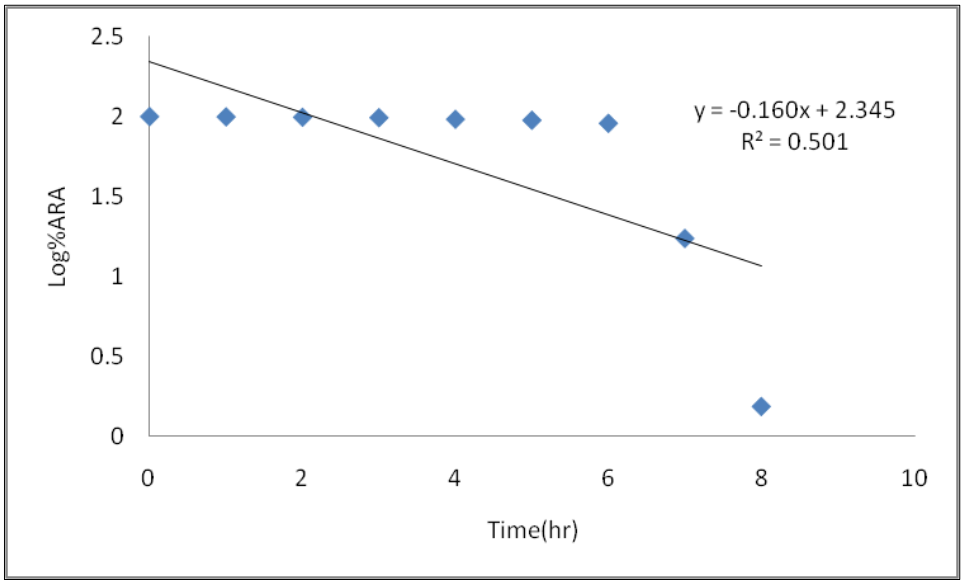

Fig 11: First order plot for optimized formulation F8

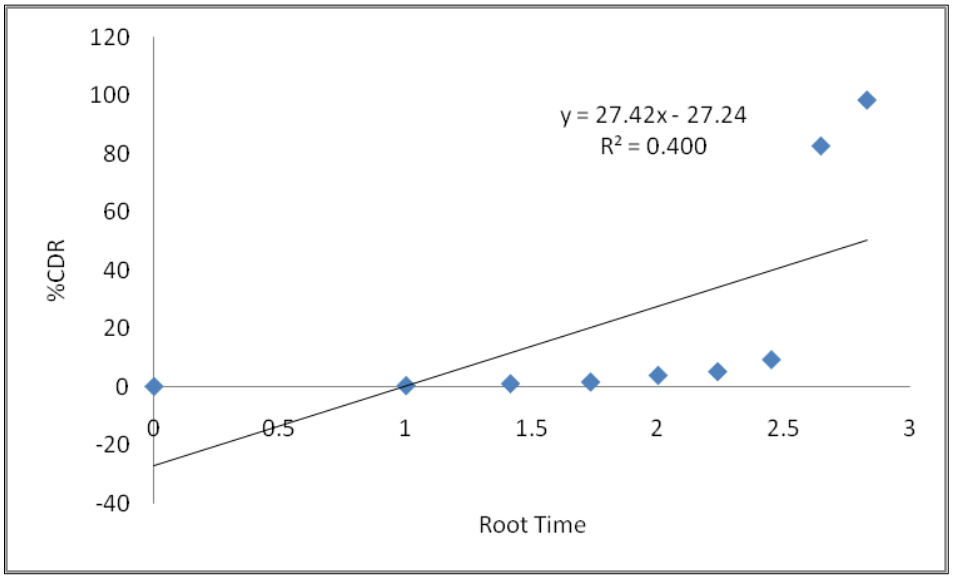

Fig 12: Higuchi's order plot for optimized formulation F8

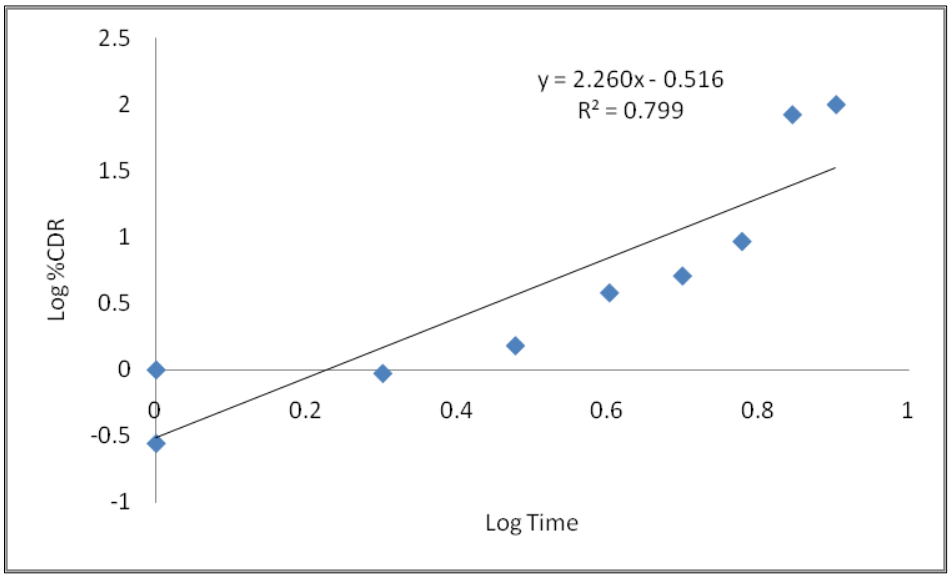

Fig 13: Koresmayer peppas order plot for optimized formulation F8

To analyze the mechanism of drug release from optimized F8 pulsincap formulation, data obtained from the drug release studies was subjected to different kinetic treatments. The correlation coefficient (R) was used as indicator of the best fitting for each of the models considered. The drug release kinetics for the optimized formulation F8 followed the zero order kinetics and follows super case II transport mechanism.

Conclusion

The aim of this study was to explore the feasibility of time specific pulsatile drug delivery system of Rivaroxaban to treat blood clot, and to lower the risk of stroke, heart attack.
From the results obtained from executed experiments it can be concluded that:

- The preformulation studies like pH, solubility and UV-analysis of Rivaroxaban were compiling with BP standards.
- The FTIR Spectra revealed that, there was no interaction between polymer and drug.
- The solubility studies of empty gelatin capsule bodies, which were cross linked with formaldehyde treatment, revealed that they are intact for 24hrs, and hence suitable for colon targeting.
- The polymers like HPMC K4M, and Ethylcellulose can be used as hydrogel plugs to delay the release of Rivaroxaban.
- The result of micromeritic properties showed good flow property of the powder blend indicating uniform distribution of drug within the various batches of capsule with negligible loss during the formulation stage.
- In conclusion, this system can be considered as one of the promising formulation technique for preparing time specific drug delivery systems and in chronotherapeutic management of blood clot. From the preliminary trials it was concluded that it is possible to formulate the pulsatile drug delivery system by the design of time modified chronopharmaceutical formulation.

References

1. Garg BK, Gnanarajan G, Kothiyal P. Formulation and Evaluation of Pulsatile Drug Delivery System of Rosuvastatin Calcium Using Different Swelling Polymers; the pharma innovation. 2012; 1:7.
2. Brahmaiah Bonthagarala, Sarvani Vadrevu, Sreekanth Nama, Donthiboina Sudarshan, Suresh Nuthakki. formulation and evaluation of pulsatile drug delivery system of atenolol. American Journal of Biological and Pharmaceutical Research. 2014; 1(1):28-33.
3. Ashish Kumar Garg, Aakrshan Kumar, Sunita Rani, Mandeep Singh, Amit Sharma, Rajesh Kumar. Formulation and Evaluation of Chronotherapeutic Pulsatile Drug Delivery System Containing Rabeprazole Sodium; Journal of Applied Pharmaceutical Science. 2017; 7(02):093-100.
4. Brahmaiah Bonthagarala, Sarvani Vadrevu, Sreekanth Nama, Donthiboina Sudarshan, Suresh Nuthakki. Formulation and evaluation of pulsatile drug delivery system of atenolol. American Journal of Biological and Pharmaceutical Research. 2014; 1(1):28-33.
5. Garg1 BK, Gnanarajan G, Kothiyal P. Formulation and Evaluation of Pulsatile Drug Delivery System of Rosuvastatin Calcium Using Different Swelling Polymers; the pharma innovation. 2012; 1:7.
6. Manish Kumar Gupta, Swarnlata Saraf. Formulation and Evaluation of Pulsatile Drug Delivery System of Ramipril for Controlling Morning Spate of B.P. Journal of Pharmaceutical Research. 2018; 17(1):1-12.
7. Rohini RS. Formulation and Evaluation of Pulsatile Drug Delivery System of Pregabalin. Pharm Anal Acta. 2016; 7:508. doi: 10.4172/2153-2435.1000508.
8. Shajan A, Banu S, Peter V, Raju S, Das C. Formulation and evaluation of pulsatile drug delivery system containing domperidone and paracetamol. Int J Pharm Sci Res. 2017; 8(3):1407-12. doi: 10.13040/IJPSR.0975-8232.8(3).1407-12.
9. Ashish Kumar Garg, Aakrshan Kumar, Sunita Rani, Mandeep Singh, Amit Sharma, Rajesh Kumar. Formulation and Evaluation of Chronotherapeutic Pulsatile Drug Delivery System Containing Rabeprazole Sodium; Journal of Applied Pharmaceutical Science. 2017; 7(02):093-100.
10. Sadaphal KP, Thakare VM, Gandhi BR, Tekade BW. Formulation and Evaluation of Pulsatile Drug Delivery System for Chronobiological Disorder: Asthma; International Journal of Drug Delivery. 2011; 3:348-356.
11. Rajesh N, Vidya JS, Manjunath MS, Bhavya KN. Formulation and evaluation of pulsatile drug delivery system; International Journal of Pharmaceutical Science and Research. 2016; 1(5):21-29.
12. Halsas M, Ervasti P, Veski P, Jürjenson H, Marvola M. Biopharmaceutical evaluation of timecontrolled press-coated tablets containing polymers to adjust drug release. Eur J Drug Metabol Pharmacokinet. 1998; 23:190-196.
13. Kro gel, Bodmeier R. Pulsatile drug release from an insoluble capsule body controlled by an erodible plug, Pharm. Res. 1998; 15(3):474-481.
14. Sandeep M, Sai Kishore V, Sudheer B, Ershad S, Adithya K, Phanil kumar DS. Design and development of chronopharmaceutical drug delivery of lansoprazole Asian Journal of Pharmaceutical Research and Development. 2013; 1(5):1-9.
15. Ramesh B, Saikishore V, Lakshmana Rao R. Design and Development of Pulsincap for Chronopharmaceutical Drug Delivery Losartan Potassium. Asian Journal of Pharmaceutical Research and Development. 2014; 2(3):78-86.
16. Dasharath Patel M, Rushiraj Jani H, Chhagan Patel N. Design and evaluation of colon targeted modified pulsincap delivery of 5-fluorouracil according to circadian rhythm, Int J Pharm Investig. 2011; 1(3):172-181.
17. Leon Lachman, Herbert Lieberman A. The theory and practice of industrial pharmacy, 3rd edition, Varghese publishing house, 315-317.
18. Pranavi P, Gulshan MD, Eswar Gupta M, Rama Rao N. Formulation and evaluation of immediate release irbesartan pellets and tablets. Indo American journal of pharmaceutical research. 2014; 4(3):1617-1624.
19. Dr. Venkateswara Reddy B, Navaneetha K, Venkata Ramana Reddy K. Formulation and evaluation of fast dissolving tablets of losartan potassium. Indo American journal of pharmaceutical research. 2014; 4(5):2573-2584.
20. Rasmitha Reddy B, Venkateswara Reddy B, Navaneetha K. Formulation and Evaluation of Dasatinib Immediate Release Tablets. World journal of Pharmacy and Pharmaceutical sciences. 2014; 3(3):1113-1123.
21. Ujwala Reddy P, Venkateswara Reddy B, Navaneetha K.

Formulation and evaluation of candesartan immediate release tablets by using liquisolid technique. World journal of pharmacy and pharmaceutical sciences. 2014; 3(2):2270-2282.
22. Deepak G, Raut Rahul, Senthil A, Shantesh Uday M. Formulation and evaluation of irbesartan immediate release tablets. International research journal of pharmacy. 2012; 3(4):410-415.
23. Sharma G, Srikanth M, Sunil S, Ramana Murthy K Application of modified pulsincap technique for oral controlled drug delivery of gliclazide. Int J Pharm Pharm Sci. 2012; 4(3):1-7.
24. Kumar ASN, Pavanveena C, Kavitha K. Colonic drug delivery system of Trimetazidine Hydrochloride for angina pectoris. Int J Pharm Pharm Sci. 2011; 3(2):22-26.
25. Patel M, Jani H, Patel N. Design and Evaluation of Colon Targeted Pulsincap Delivery of 5 Flurouracil According to Circadian Rhythm. Int J Pharm invest. 2011; 1(3):172-183.
26. Peppas NA. Analysis of Fickian and non-fickian drug release from polymer. Pharm Acta Helv. 1985; 60:110-11.18.
27. Edith Mathiowaz, Donald Eichickering, Claus-Michael Lehr. Bioadhesive Drug Delivery Systems: Fundamentals, noval approaches. Marcel Dekkar Inc. New York, Basal. 1999; 98:670.