NOVEL CHRONOTHERAPEUTIC MULTIPARTICULATE DRUG DELIVERY SYSTEM OF FELODIPINE: AN EFFECTIVE TREATMENT FOR CARDIAC ARRHYTHMIA

Sabyasachi Banerjee1*, K. Ravi Shankar2, Y. Rajendra Prasad3

1CPS Formulations, Dr. Reddy’s Laboratories Ltd, Hyderabad, India
2Sri Sai Aditya Institute of Pharmaceutical Sciences and Research, Surampalem, India
3College of Pharmaceutical Sciences, Andhra University, Visakhapatnam, India

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*Corresponding author
Sabyasachi Banerjee
Email: sabyasachiph@gmail.com

ABSTRACT

Arrhythmia follows chronobiology, thus necessitating the development of a time-dependent formulation for its treatment. The aim of the current work was to develop a solubility-enhanced chronotherapeutic system of felodipine, a widely prescribed anti-arrhythmic. Systematically optimize hot-melt extrusion process was employed to formulate solubility-enhanced extrudates. The film casting method was adopted for the selection of polymers. Drug released at 5, 15, 30min was taken as response variables in 32 face-centered cube design. Nearly 10-fold increase was observed in the solubility of the optimized extrudates in comparison to pure drug. Physical characterization of the extrudates depicted complete amorphization of the drug. The sequential coating was performed onto the extrudates to enable a time-dependent release. In-vitro studies clearly demonstrated that 25% of the drug was available rapidly within 10 min of administration. The remaining 75% of the drug was available over a period of 4, 8 and 12h. Stability studies performed for 6 months at accelerated conditions depicted no significant change in the physicochemical characteristics of the optimized formulation. In-vivo pharmacokinetic studies conducted in beagle dogs ratified the results of in-vitro studies where a sequential time-dependent absorption of felodipine was observed over a period of 12h. Concisely, the studies demonstrated successful development of a solubility-enhanced chronotherapeutic system of felodipine.

Keywords: hot-melt, central composite design, pharmacokinetics, arrhythmia, extrudates

INTRODUCTION

Cardiac arrhythmia is a serious health problem and has remained a hot topic of scientific discussions since a long time (Evans, et al., 2000; Takahara, et al., 2000; Grant, 2003). Some of the arrhythmias can be mild but if the blood supply to vital organs gets compromised during an arrhythmia, then it can be fatal (Schmid, et al., 2013). Also, unlike many other diseases, the arrhythmia is not a chronic disorder in the true sense. It is well reported in the literature that cardiac arrhythmia follows a distinct chronobiological pattern with attacks likely to occur in late hours of the morning, afternoon and early evening (Peters, et al., 1996). Thus the concentration of drug meant for the purpose should also be at peak in the said time intervals.

One way to achieve this is to administer drug every four hours or so. However, this approach is tedious and it is very difficult to ensure patient compliance with this type of regimens. Due to this, there is a dire unmet need of a drug delivery system capable of delivering the drug at required timings while maintaining the ease of single administration.

Literature reports some of the approaches that have been followed to formulate chronotherapeutic systems of calcium channel blockers (CCBs), one of the most promising therapeutic options for cardiac arrhythmia (Karavas, et al., 2006; Pagar and
Vavia, 2012). However, these approaches basically focus on combining three to four sequentially coated tablets into one capsule formulation. Moreover, these systems suffer from disadvantages of dose dumping and high inter and intra-subject variability due to their high dependence on gastric emptying. Further, the existing commercial formulation of FLD (Plendil tablets) is based on matrix technology. For matrix type of dosage form, the varying physicochemical characteristics and contractile intensity along the GI tract are the potential sources to alter drug release. Moreover, in the case of matrix type dosage forms the release rate is governed by erosion mechanism which may not be predictable many a times. Due to which, dose dumping and serious adverse events may occur.

Considering these facts, the present study was aimed to formulate multi-particulate chronotherapeutic systems of felodipine. Multiparticulates are less dependent on gastric emptying, resulting in less inter and intra-subject variability in gastrointestinal transit time (Jain, et al., 2008; Roy and Shahiwala, 2009). They are also less prone to local irritation due to their precise distribution of the drug. Also, as the constituent particles pass through GI tract quickly, this result in better pharmacokinetic profiles than single unit dosage forms. Some of the particles, if less than a certain size, can pass even through a closed pylorus. These result in lower intra- and inter-individual variability in plasma levels and bioavailability (Abdul, et al., 2010). The present approach focussed on increasing the solubility of felodipine by amalgamating it with Eudragit EPO employing hot-melt approach. The extrudates were differentially coated in order to provide a sequential time-dependent release: around 25% of the drug to be released immediately and the rest 75% to be released subsequently at around 4th, 8th, and 12th hr. This would provide a complete therapeutic option for patients with hypertension with the intake of just one capsule. Furthermore, the present work includes a detailed pharmacokinetic study in beagle dogs, a closely related mammal to humans, in order to substantiate our hypothesis. Such a study has not been reported with earlier attempts of chronotherapeutic formulations of felodipine.

**MATERIAL AND METHODS**

Felodipine was sourced from Nivedita Chemicals Pvt. Ltd., India. Eudragit EPO was purchased from Evonik Industries, Germany. Soluplus, Povidone, and Copovidone were purchased from BASF, Germany. Ethyl cellulose and dibutylsebacate were obtained from Dow Chemicals, USA and Vertellus Inc., USA. Empty hard gelatin capsule shells were sourced from ACG Capsules, India. All other chemicals employed were of analytical grade.

**Evaluation of film casting**

Various polymers (in drug: polymer ratio ranging between 1:1 and 1:5) were screened by film casting method. Briefly, the polymer and the drug substance were dissolved in a common organic solvent and the solvent was vacuum evaporated (Rotary Flash Evaporator, Medica Instrument Mfg. Co., India). The resultant slurry was casted onto a glass surface which was then dried at a temperature of 60 °C. The ensuing film was observed, both physically and microscopically, for surface clarity (i.e., the absence of any drug or polymer particles). The polymers which were evaluated in the study are Copovidone, Soluplus and Eudragit EPO.

**Preparation of solid dispersion of felodipine**

Briefly, the blend was prepared by blending 10mg of drug substance with Eudragit EPO and Povidone K-12 for 10min at 25rpm. A co-rotating twin screw extruder (Nano-16, Leistriz, Germany) fitted with a round die, operated at a barrel temperature of 100-130°C and screw speed between 100 and 150rpm, was employed to formulate the extrudates of the blend. The resultant extrudates were sized through co-mill to attain a particle size of about 500µm. Only those extrudates which were retained above # 30 ASTM sieve were selected for further process. Table I depicts the detailed composition of the thirteen experimental trials.
formulated as directed by 3\(^2\) face centred cube design (FCCD) (Singh et al., 2013). The formulation at center point was formulated in quintuplicate.

Table I. Levels of Eudragit EPO and Povidone K-12 in the thirteen formulations prepared as per the experimental design;

<table>
<thead>
<tr>
<th>Formulation</th>
<th>Eudragit EPO</th>
<th>Povidone K-12</th>
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</thead>
<tbody>
<tr>
<td>F1</td>
<td>10</td>
<td>5</td>
</tr>
<tr>
<td>F2</td>
<td>10</td>
<td>5</td>
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<tr>
<td>F3</td>
<td>10</td>
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<td>F4</td>
<td>30</td>
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<td>F5a</td>
<td>30</td>
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<td>F5b</td>
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<td>F5c</td>
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<td>F5d</td>
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<td>F5e</td>
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<td>50</td>
<td>45</td>
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<tr>
<td>F9</td>
<td>50</td>
<td>45</td>
</tr>
</tbody>
</table>

In vitro dissolution studies of solid dispersion

The dissolution of pure drug substance and solid dispersions prepared as per DOE trials were conducted in 900mL of pH 6.8 phosphate buffer using USP II apparatus at 50 rpm. Aliquots were collected from the dissolution vessels, filtered through 0.2µm filters and analyzed employing HPLC. The analysis was performed with PDA detector at a λmax of 360nm. X bridge C18, (150 x 4.6mm), the 3.5µm column was employed with a flow rate of 1mL/min. An injection volume was 5µL was used. For quantitative determination, a linear calibration graph was obtained over a concentration range of 2.49-99.6µg/mL with r=0.9996. The Limit of detection and limit of quantification were 0.6µg/mL and 1.6µg/mL, respectively. Drug released at 5min, 15min, and 30min (Q5, Q15 and Q30 respectively) were taken as the response variables for the purpose of DoE optimization.

Analysis of DoE data

Design Expert software (Stat-ease Corporation) was employed to analyze the data obtained from the experimental runs. Response surface graphs and contour plots were generated for better elucidation of the mechanism of action of polymers. An overlay plot was constructed and the formulation which qualified the best to the criteria mentioned below was selected as the optimized formulation. Optimization criteria*: Q5: NLT65%; Q15: NLT 80%; Q30: NLT 90%

*NLT: Not less than

Saturation solubility studies of solid dispersion

Saturation solubility of felodipine, as well as the optimized solid dispersion, was estimated in buffer solution prepared in the physiological pH range of 1.2 to 7.0. Briefly, excess the drug substance or solid dispersion was added into each of the buffers and was subjected to shaking on a mechanical shaker (Remi Laboratory Instruments, India) for 24h. The samples were collected, filtered through a 0.2µm filter (Millipore, USA) and analyzed employing UV spectroscopy at 365nm (UV-2600, Shimadzu Corp., Japan).

Characterization of solid dispersion

XRD studies

Powder X-ray diffraction (PXRD) (D-8 Advance X-ray diffractometer, Bruker, Germany) studies were conducted for drug substance and the optimized solid dispersion. The study was conducted at a tube voltage and current of 40 kV and 40 mA respectively. The values of 20 ranged between 3 and 45°C.

Modulated temperature differential scanning calorimetry (MTDSC)

MTDSC analysis of the optimized solid dispersion was performed using differential scanning calorimeter (DSC Q2000, TA
instruments) for determining the crystals in the solid dispersion. The following parameters were employed: Data storage : off; Equilibrate : At 0.00°C; Modulate: +/- 1.59°C every 60s Isothermal : for 5.00min; Data storage : on; Ramp: 2°C/min to 200°C; N2 flow : 50.0mL/min.

Accurately weighed samples were transferred to the autosampler tray and were subjected to analysis. The thermograms obtained were processed by determining the onset and peak temperature of melting endotherm.

Thermogravimetric analysis (TGA)
TGA was used to determine the degradation temperature of drug and Eudragit EPO at a starting temperature of 0°C, with an increase of 5°C up to an end temperature of 250°C. The nitrogen flow was 60mL per min for the sample and 40mL per min for the balance.

Coating of solid dispersion
Sub-coating
The optimized solid dispersion extrudates were coated to the level of 5% with HPMC 6 cps solution in fluidized bed processor (GPCG 1.1, ACG Pam Technologies, India). The coating was performed at a product temperature of 40-45°C and atomization of 1.5 bar. The C-type base plate was employed in the coating process. The sub-coated extrudates were divided into two parts: 25% and 75%. The 75% fraction was subjected to pulsatile coating.

Pulsatile coating for time-dependent drug release
Seventy-five percent of the sub-coated extrudates (equivalent to 7.5mg of drug) were further coated with a coating mixture comprising of ethyl cellulose, dibutyl sebacate and Povidone K-12. The quantitative composition is depicted (Table II). Different levels of coating were performed in order to achieve a pulsatile release. One-third of the extrudates were coated to the level of around 6% w/w, another one-third of the extrudates were coated to the level of around 10% w/w and the remaining one-third of the extrudates were coated to the level of around 15% w/w. All differentially coated fractions, along with the sub-coated extrudates were filled in Size “3” hard gelatin capsules.

In vitro dissolution studies of capsules
The sequential in vitro dissolution studies of the prepared capsule formulation was conducted in USP III apparatus (reciprocating cylinder) at 5rpm using 250mL of dissolution medium. In USP III apparatus, the internal cylinders remain in the first line of vessels containing 0.1 NHCl, in the reciprocal movement for 1.5h. After the programmed period of 1.5h, the rods rise until the internal cylinders are positioned over the vessels, where they remain for a pre-established timeframe so that the dissolution medium can drain. Then the rods move to the following line containing pH 4.5 acetate buffer, submerging again and the reciprocating actions begin anew for the duration of 30min. Finally, the rods move to the last line containing pH 6.8 phosphate buffer, submerging into the vessel and reciprocating action continued for 10h. The samples were analyzed for drug content as per method described under section “In vitro dissolution studies of solid dispersions”.

Stability studies
The optimized formulation was subjected to 6 months of stability studies at accelerated conditions of 40 °C/ 75% RH. The formulation was packed in HDPE bottles for the purpose of stability studies. The formulation was analyzed initially and at the end of six months for assay, PXRD, related substances and dissolution profile. Assay and related substance analysis were performed using HPLC with PDA detector at a λmax of 360nm. X bridge C18, (150 x 4.6mm), the 3.5µm column was employed with a flow rate of 1mL/min. The injection volume was 5µL.

In vivo pharmacokinetic studies
The optimized formulation was subjected to in vivo evaluation in dogs. Taking cognizance that the research work adheres to the guidelines for care and use of the laboratory
animals, all the animal investigations were performed as per the requisite protocol approved by Dr Reddy’s Laboratories. A singledose study was carried out using 12 unisex beagle dogs, with their body weights ranging between 9.21 and 12.70 Kg.

The dogs were fasted 12h prior to drug administration. All the animals were allowed free access to water throughout the study. Following administration of the optimized encapsulated formulation, dogs were kept in their housing, and access to food and water was allowed ad libitum after 6h. Serial aliquots of blood samples (1mL) were withdrawn from centrifugation tubes. Plasma was harvested by centrifugation (3000rpm, 800g, 5min), and stored at -20°C until analyzed.

The content of felodipine in plasma samples was analyzed by HPLC as per method described under section “In vitro dissolution studies of solid dispersions”. Various pharmacokinetic parameters such as plasma concentration-time curve (AUC), maximum drug concentration (C_max), the time to reach maximum drug concentration (T_max), elimination half-life (T_1/2), and elimination rate constant (K_e) were calculated employing Win Nonlin software (Version 5.0, M/c Pharsight Corporation, CA, USA).

RESULTS AND DISCUSSION
Evaluation of film casting

The transparency of films prepared with various polymers (i.e. Copovidone, Soluplus and Eudragit EPO) at drug:polymer ratio of 1:5 are compared. Drug-polymer films with Copovidone and Soluplus are opaque whereas clear transparent glassy films are obtained with the Eudragit polymer. Therefore, the Eudragit polymer was selected for further optimization study employing Design of Experiment (DoE) trials.

Analysis of DoE data

A good fit was observed for the polynomial equation (Eq. 1) used to fit the data with a high value of R².

\[ Y = \beta_o + \beta_1X_1 + \beta_2X_2 + \beta_3X_1^2 + \beta_4X_2^2 + \beta_5X_1X_2 + \beta_6X_1^2X_2 + \beta_7X_2^2 + \beta_8X_1X_2^2 + \beta_9X_1^2X_2^2 + \cdots \] (4)

where \( \beta_0 - \beta_7 \) depict the various coefficients and X1 and X2 depict the independent variables. Y represents the response variable. The 3-D response surface graphs and overlay plot for the response variables (Figure 1).

Figure 1 (R1) depicts an inverted U-type curve of Q5 with an increase in the levels of Povidone K-12. The values of Q5 first increase, reach a maximum and then decrease with an increase in the levels of Povidone K-12. The curved lines of the corresponding contour plot also depict that higher levels of Eudragit are imperative for high values of Q5.

As observed in the case of Q5, an inverted U-type curve of Q5 with an increase in the levels of Povidone K-12. Further, in the case of Q15 and Q30, (Figure 1 (R2 and R3)) the similar trend is observed with Eudragit also depicting that optimized levels of both the polymers are imperative for getting adequate dissolution parameters. The corresponding contour plots (not shown in the article) also depict the same.

The optimized formulation is selected by trading-off the values of various responses exhibited the desired values of Q5, Q15 and Q30. The optimized formulation, containing 29.38mg of Eudragit and 29.06 mg of Povidone K-12, exhibited a Q5 of 69%, Q15 of 85% and Q30 of 94%. Figure 1 (overlay plot) depicts the formulation of optimized solid dispersion.

In vitro drug release studies of solid dispersion

The in vitro drug release of the optimized solid dispersion and pure drug in pH 6.8 phosphate buffer was observed to be 100% and 5% respectively, after a duration of 60min. A nearly 20-fold increment was observed in the drug release profile of the optimized solid dispersion as compared to the pure drug.

Saturation solubility studies of solid dispersion

The saturation solubility of optimized solid dispersion and pure drug were observed
to be 0.047 – 0.052 mg/mL and 0.51 – 0.54 mg/mL respectively. A 10-fold increase was observed in the solubility of optimized solid dispersion vis-a-vis pure drug in the entire physiological pH range.

**Characterization of solid dispersion**

Figure 2 shows the XRD pattern of pure drug and optimized solid dispersion of FLD and Eudragit in a molar ratio of 1:5. Many high-intensity diffraction peaks were observed in the diffractogram of pure FLD. The numerous distinctive peaks of the pure drug were disappeared in the solid dispersion, indicating conversion of the drug into an amorphous state.

In the DCS thermogram, a single Tg value ranging from 38.68°C to 41.17°C is noted for solid dispersion of FLD and Eudragit. This clearly indicates the miscibility of the drug in the polymeric carrier. A decrease in the Tg value of Eudragit is also observed which notify the plasticizer property of drug in presence of the polymer. TGA study of of drug and Eudragit indicates that the degradation temperatures of both drug and polymer are around 200°C, which justifies their suitability for HME process.

**In vitro drug release studies of capsule**

The in-vitro drug release profile of final capsule formulation of FLD in sequential dissolution media (0.1 N HCl for 1.5h, pH 4.5 buffer for 30min and pH 6.8 buffer for 10h (Figure 3). Figure 3 clearly showed that around 25% drug was released at 1h, around 50% at 4h, around 75% at 8h and 100% at 12h.
Inspection of the reported data reveals that the dissolution rate of the drug is programmed and a constant amount of drug is released initially and at an interval of every 4 hr (i.e. 4h, 8h and 12h).

**Stability studies**

The assay value of the capsule formulation after 6 months of storage at accelerated conditions of 40 °C/75% RH was 99.4% against an initial value of 100.2%. The two identified impurities (Impurity B and C) and all unknown impurities were within the ICH limits at initial and after stability period of 6 months. The PXRD studies of stability samples also showed overlapping spectra with their initial samples. A comparable dissolution profile was observed for the optimized formulation at the initial stage and after 6 months of storage at accelerated conditions.

**In vivo pharmacokinetic studies**

The efficiency of the developed formulation was validated by *in vivo* performance. Table III depicts the values of various pharmacokinetic parameters calculated from the plasma concentration-time profile data. The optimized formulation exhibited a $C_{max}$ of 5625.00 ng/mL and AUC 296.90 ng.hr.mL$^{-1}$. The $T_{1/2}$ value of 28.39 hr and $K_e$
value of 0.024 hr\(^{-1}\) indicate prolonged absorption rate coupled with the slow elimination of the drug from the body. Figure 4 depicts the curve of plasma drug levels as a function of time after 192h.

Table III. Pharmacokinetic parameter calculated from plasma drug level profile

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Primary parameters</strong></td>
<td></td>
</tr>
<tr>
<td>C(_{\text{max}})</td>
<td>5625.00ng/mL</td>
</tr>
<tr>
<td>AUC(_{0-\text{t}})</td>
<td>296.90ng.h.mL(^{-1})</td>
</tr>
<tr>
<td><strong>Secondary parameters</strong></td>
<td></td>
</tr>
<tr>
<td>T(_{\text{max}})</td>
<td>12.21h</td>
</tr>
<tr>
<td>K(_{e})</td>
<td>0.02h(^{-1})</td>
</tr>
<tr>
<td>T(_{1/2})</td>
<td>28.39h</td>
</tr>
</tbody>
</table>

Like a majority of drug substances, Felodipine, a promising CCB, suffers from a major downside of poor solubility (Raina \textit{et al.}, 2014; Sarode \textit{et al.}, 2014a; Sarode \textit{et al.}, 2014b). Since the drug belongs to Class II as per the Biopharmaceutics Classification system (Tapas \textit{et al.}, 2009), permeability is not the rate-limiting step in its absorption. Thus, apart from the first-pass metabolism, one more predicament governing the low bioavailability of felodipine is its very low solubility in the lumen. Considering this fact, reports abound in the literature describing various attempts to increase the solubility and dissolution of the drug substance (Karavas \textit{et al.}, 2005; Alonzo \textit{et al.}, 2011; Basalious \textit{et al.}, 2011). Approaches like nanodispersion (Karavas \textit{et al.}, 2006) and \(\beta\)-cyclodextrin (Pagar and Vavia, 2012) complexation have been employed for the purpose. However, neither of the two approaches are effective enough to render the drug substance completely amorphous. Complete amorphization of drug essential for ensuring complete absorption, particularly, in the case of felodipine since it suffers from very low solubility. The increased solubility also helps in reducing intra and inter-individual variabilities in drug absorption as the only solubility is the rate-limiting step in its absorption.

The film-forming capacity of various polymers depends upon their ability to form a transparent glassy solution with the drug. Copovidone and Soluplus were not able to form a clear glassy film as the quantity of the polymer employed was not sufficient to induce molecular interactions. On the other hand, Eudragit was able to form a clear transparent glassy film in the same drug: polymer ratio (1:5). The same can be attributed to the flexible chains present in the polymer which accommodate themselves to form a clear matrix at a temperature as low as 40-45°C. This property, in particular, has been imparted by the low glass transition temperature (Tg of around 45°C) of Eudragit EPO (Higashi \textit{et al.}, 2014). As only Eudragit EPO was able to form a transparent film, the same was selected for further optimization employing DoE. Also, the extrudates had to withstand the rigorous fluidization during the coating process. Since, many-a-times, a binder is also required in order to provide adequate strength to the extrudates, it was decided to introduce a low-viscosity binder, Povidone K-12, in the optimization trials. The effect of this additional binder on the dissolution characteristics of extruding was envisaged to be studied employing a detailed FCCD.

A 3\(^{\text{nd}}\)FCCD was employed in the current development work because it provides the benefit of exploring quadratic interactions between the polymers (Singh \textit{et al.}, 2009; Dhawan \textit{et al.}, 2011; Singh \textit{et al.}, 2011). Further, the formulation at center point was formulated in quintuplicate and analyzed five times to ensure the robustness of the design. Such is not feasible with standard factorial designs (Kapil \textit{et al.}, 2013). The high values of correlation coefficient vouched for adequate selection and the high prognostic ability of the design.

The increase in the rate of dissolution of the drug with high levels of Eudragit can be attributed to the ability of the polymer to disperse the drug into amorphous particles or molecular dispersion (Feng \textit{et al.}, 2012). Further, Eudragit EPO prevents the recrystallization by stabilizing the drug substance through hydrogen bonding, vander Waal and ionic interactions.
(Higashi et al., 2014). Povidone, on the other hand, provided a synergistic effect in drug release.

A nearly 20-fold increment was observed in the drug release profile of the optimized solid dispersion as compared to the pure drug. This proves the adequate selection of the solid dispersing polymer, the experimental design, and our hypothesis. The same is also corroborated by saturation solubility studies where a drastic increase in the solubility of drug substance is observed upon being formulated as a solid dispersion.

The absence of any crystalline peak in the PXRD spectra clearly corroborates the complete conversion of crystalline felodipine to its amorphous form (Rezaei Mokarram et al., 2010; Pattnaik et al., 2011). This is because Eudragit EPO was able to disperse the drug substance on a molecular level leading to an amorphous product that exhibits a single glass transition temperature.

MTDSC is an important tool to assess the amorphous content of the sample (Gill et al., 2010). Since the sensitivity of MTDSC is more as compared to conventional DSC, it can even estimate the amount of molecularly dispersed drug. A single Tg in DSC thermogram clearly indicates that no phase separation has taken place and the drug is molecularly dispersed in the polymer. TGA studies showed high degradation temperatures of both drug and polymer, signaling that the temperature at which the hot melt process is carried out is not at all detrimental to the stability of the product.

The in vitro dissolution profile clearly demonstrates that a time-dependent sequential drug release is observed when the optimized capsule formulation was subjected to dissolution studies. Thus the concentrations of which the different extrudates were coated were successfully able to control the release of drug in the programmed hours. A nearly equal amount of drug was released initially, and at 4th, 8th and 12th hr. The coating polymer, ethyl cellulose does not have a solubility in aqueous media, thus providing complete protection till the layer is intact. Dibutyl sebacate, in a generally acceptable concentration of 10% w/w (Alavi et al., 2002), was able to provide enough plasticity to the film in addition to providing a synergistic release-controlling effect because of its hydrophobicity. As the thickness of coat increased, the drug release was observed to be decreased proportionally. This can be attributed to the ethyl cellulose layer becoming more impermeable to dissolution media. Povidone K-12 acted as a pore-former in the formulation thus helping in the release of drug (Zhang et al., 2007; Tuntikulwattana et al., 2010; Yang et al., 2014). On coming in contact with the dissolution medium, Povidone gets dissolved thus leaving behind a pore in the coat. This facilitates the drug release without affecting the flexibility of the film.

Stability studies concluded that there was no significant change observed in the assay, dissolution profile, impurities level and polymorphic form of the optimized formulation upon storage at accelerated condition of 40 °C/75% RH for a period of 6 months. This further emphasizes the robustness and ruggedness of developed formulation.

The in vivo pharmacokinetic studies are in agreement with the anticipated results. There was a strong correlation between the results obtained during in vitro dissolution studies and the in vivo pharmacokinetic studies. As can be discernible from Figure 4, a time-dependent sequential absorption of drug was observed over a period of 12h. Since permeability of felodipine was never a point of concern, the two factors crucial in developing a successful chronotherapeutic systems of felodipine were its rate of drug release and the solubility of released drug in intestinal lumen. One of the factors was successfully addressed by adequately optimized hot melt extrusion process while the other was taken care by a precisely designed sequential coating system. Since this is a long half-life drug, the subsequent dose of drug was absorbed much before majority of drug being eliminated. This gives rise to the characteristics peaks and troughs in the plasma drug level profiles, thus maintaining the drug levels at therapeutic...
concentrations throughout the therapy. The value of half-life (around 28 hr) is in close agreement with previously reported literature. Due to this long half-life of the drug, it took 192 hr to achieve near complete elimination. The in vitro drug release is successfully translated in the in vivo conditions, thus establishing a proof of concept formulation for sequential release of felodipine.

CONCLUSION
In a nutshell, the present study demonstrates the successful development of a solubility-enhanced chronotherapeutic system of felodipine capable of providing a time-dependent drug availability in blood. The said system can be used as single therapy for day-long management of arrhythmia.

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