Evaluation of sustained release matrix tablets of cilostazol

Development and in Vitro-in Vivo Evaluation of Sustained Release Matrix Tablets of Cilostazol

Keywords: Cilostazol; Pharmacokinetics; ER Matrix Tablet; In Vitro Kinetics;

ABSTRACT

The objective of this research had to manufacture extended release matrix tablet of Cilostazol and to evaluate its *in vitro* drug release and *in vivo* absorption. The dosage form was designed by selection of various polymers such asHypromellose, Kollidon SR, Xanthan gum, Ethyl cellulose, Eudragit Polymers. Microcrystalline cellulose and lactose as dilutents to build matrix tablets and povidone as granulating binders. The tablets were prepared by Direct compression, wet granulation and Melt extrusion techniques. Optimized formulation of Cilostazol matrix tablets was prepared by using 7% HPMC K100M polymer, 39 % MCC, 3% of povidone as binder. Matrix tablets were compressed with optimized free flowing granules of uniform drug content. This *in vitro* drug release showed the extended the release period up to as per desired specifications. The matrix formed by HPMC, MCC and Povidone had been showed satisfactorily with the controlled resistance. Bioavilability study of this wet granulation dosage formulation in rabbit model showed 24 h sustained drug release in vivo. A correlation ($R^2 = 0.9833$) was founded between the in vitro drug release and the in vivo drug absorption. The results suggested that wet granulation with is a satisfactory method to develop a sustained release Cilostazol and it can be Performed therapeutically better than conventional IR dosage form.

1. Introduction

In this study the Cilostazol sustained release matrix tablet was developed with various polymers. Since the IR dosage form produces and side effect head of ache due to drug oscillation in plasma. The challenge become to develop a matrix tablets are due to drug morphology and highly insoluble in nature . In the present study, a sustained release dosage form of Cilostazol has been developed that enables less frequent administering of drug . Matrix tablets of Cilostazol were formed by appropriate combination of HPMC and Povidone and lactose monohydrate, MCC and Kollidon K30 was chosen for matrix tablet to extend duration of drug release.

Cilostazol and its metabolites are inhibit the platelet aggregation and exert vasodilatory action by inhibiting phosphodiesterase activity and cAMP degradation with a resultant increase in cAMP in platelets and blood vessels

The objectives of research were: 1) To analyze the physical and chemical characters of prepared Tablets 2) To elucidate the effect of polymers and to study the release kinetics, 3) in-vivo study for the stable formula.

2. Materials and Methods

2. 1. Materials

Cilostazol was obtained from IPCA lab, Mumbai, India. Hypermellose (Methocel K100M CR), Povidone K30 received as a gift sample from Colorcon Pvt Ltd. Kollidon K30 was obtained from BASF. All other Reagents were purchased from local suppliers, India and were of analytical grade.

2. 2. Drug and Excipient Interactions

Drug Excipient interaction study was investigated by DSC (differential scanning calorimeter). The DSC Thermo gram of only drug and Drug+Excipient mixtures were noted. The samples were separately packed in aluminum cells and kept a set in Metler TA 4000 Thermal analyzer.

2. 3. Formulation

1. Dispensing: All the ingredients were dispensed accurately as per formula quantity.

2. Sifting: Measured quantity (refer table no. 4. 5) of Cilostazol, Microcrystalinecellulose (Avicel PH-101), HPMC K100M, were passed through 30#, Microcrystalinecellulose(PH-102), Aerosil-200, through 40#, yellow oxide of iron and Magnesium stearate were passed through 60# .

3. Mixing: Measured quantity (refer table no. 4. 5) of Cilostazol, Microcrystalinecellulose (Avicel PH-101), were mixed in polybag for 15 min, to it added yellow oxide of iron and mixed for 5 min. in RMG at 2. 4 RPM .

4. Preparation of Binder solution: Measured quantity (refer table no. 4. 5) of IPA and Water were poured in stainless steel beaker, to it added PVPK-30 with stirring continuously by glass rod till dissolved completely and clear solution is obtained.

5. Wet Granulation: Granules were prepared by wet granulation method in RMG at 2. 4 RPM for 15 min, using step 4 binder solution. Prepared granules dried at 60 ºc till LOD reaches less than 2. 5% and finally sifted through 30#.

6. Mixing: Mixed the measured quantity (refer table no. 4. 5) of Microcrystaline cellulose (Avicel PH-102), HPMC K100M, Aerosil-200, in polybag for 15 min with granules obtained in step 5.

7. Lubrication: Above granules are lubricated with measured quantity of magnesium stearate

In the trial 5 concentration of HPMC K100M is reduced from 10% to 8% and trials 6, 7, 8 from 10% to 7% .

2. 4. Physical Characterization of Tablets

The prepared tablets were subjected to various physical characterization studies. Weight variation test was performed with 20 tablets with an electronic balance. Tablets hardness was determined using Monsanto (Standard type) tablet hardness tester. Thickness was measured by a venier caliper (Mitutoyo, Japan). Friability was calculated using a Roche friabilator (Basel, Switzerland)

2. 5. Drug Content of Tablets (Assay by HPLC)

Cilostazol USP:

Chromatographic Conditions:

The drug content of the formulated tablets was estimated by HPLC method. Column: Stainless steel column packed with octadecylsilane silica gel for chromatography , c18 , 150×4. 6 mm, 5µm(Inertsil ODS-3 is suitable) Mobile phase:: Acetonitrile: Mthanol: Water(7: 3: 10by volume), filter and degas.

Flow rate: 1. 0ml / min Wavelength: 254nm Diluent: Methanol Injection Volume: 10µl Temperature: 27 ^{0} C± 1 ^{0} C Retention time: Cilostazol- about 9. 4 minutes. Cilostazol was analyzed by HPLC at a wavelength of 254 nm.

2. 6. In Vitro Dissolution Studies

In-Vitro dissolution Studies (Dissolution analysis by HPLC)

Dissolution testing for the amount of drug-substances released was studied using the following dissolution parameters:

Table –: Dissolution parameters and specifications for Cilostazol

Drug Name	Apparatus	Speed (RPM)	Medium	Volume (ml)	Acceptance Criteria	
Cilostazol	USP-2 paddle	75	6. 8 phosphate buffer	900	Time (hrs)	Amount dissolved
1	NMT 20%					
4	25% – 45%					

8	55% – 75%
12	NLT 80%

Acceptance criteria : As given table no. 4. 15

Dissolution Parameters: Medium: 900 ml, 0. 3% SLS in 6. 8 Phophate bufferApparatus: USP Apparatus 2Paddle Speed: 75 RPM Temperature: 37 ^{0} C ± 0. 5 ^{0} c. Duration: 1, 4, 8, 12, 24 hours Chromatographic Conditions: Instrument: HPLC(Hitachi) Column: Stainless steel column packed with octadecylsilane silica gel for chromatography, C-18, 150cm× 4. 6mm, 5µm (Inertsil ODS-3 is suitable) Mobile Phase: Acetonitrile: Mthanol: Water(7: 3: 10 by volume) filter and degas. Flow rate: 1. 0 ml/min Wavelength: 254nm Injection Volume: 20µl Diluent: Methanol, Dissolution Medium Temperature: 27 ^{0} C ± 1 ^{0} C. The release studies were conducted in duplicate. Mean % cumulative drug release was plotted against time (hours).

2. 7. Drug release Kinetics and Mechanism

Kinetics of drug release was determined by fitting data to

Table 1. Composition of extended release matrix tablet of Cilostazol different models such as zero order (M = kt), first order equation (M = lnM $_{0}$ + kt), Higuchi model (M = k√t) and KorsemeyerPeppas equation (M = ktn). The

value of n = 0. 5 denotes case I diffusion (Fickian), 0. 5 < n < 1 is for (non-Fickian) anomalous diffusion.

n = 1, for case II transport and n > 1 for super case II transport. Where M is the amount of drug (%) released after time t; Where M_0 is the amount of drug released at (0) zero time; k is the release rate constant, n is the exponent. Drug release following particular mechanism is judged by the linearity of plot

2. 8. Stability Studies

Stability studies were conducted on SR Tablets of select batch to assess their stability with respect to their physical appearance, drug content and release characteristics after storing at 25ËšC under 60% relative humidity (RH) and 40ËšC under 75% RH for 6 months [8].

2. 9. Pharmacokinetic Evaluation

The animal studies were performed as per guidelines for the Care and Use of Animals that were approved by the Animal Ethics Committee. Male rabbits (Albino) with average weight of 2. 5 kg were housed in standard cage individual, which well ventilated with air, humidity and temperature control.

50 mg equivalent weight of Cilostazol sustained Release Tablets with and 50mg equivalent weight of Cilostazol 50mg IR tablet was administered to 2 groups orally (N = 4) via silicone rubber gastric intubation tube.

A wooden rod was kept between the jaws of rabbit’s mouth. A gastric tube was centrally placed over the hole in mouth (21. 22). With the help of gastric

intubation tube the tablets were administered in to the stomach of rabbit by set on the tip in it. After administered the oral dose, 5 ml of water was given to facilitate the admittance of the tablets. Rabbits were kept fasting over night but access to water *ad libitum* ; In a heparin zed branule (G22, G24) 2 ml of blood samples were collected, which placed in the marginal ear vein , at each of the pre determined times i. e., 0. 25 Hr, 0. 5 Hr, 1 Hr, 2 Hr, 4 Hr, 6 Hr, 8 Hr and 24 Hr after administration: Samples were transferred to eppendrof centrifuge tube and centrifuged at 3000 rpm for 10 min. The separated organic layer will be collected and evaporate to dryness under a gentle steam of nitrogen gas. The obtained residues will be reconstituted in organic solvent with vortex mixing, from which aliquot will be injected to HPLC system.

supernatant plasma was separated and transferred and stored at –20ËšC until Analyzed.

in to 96 well plate

2. 10. In Vivo Data Analysis

The maximum plasma concentration (C_{max}) and the time to reach the maximum concentration (t_{max}) were directly obtained from the observed values. The area under the curve up to 24 h after administration (AUC) was calculated by the trapezoidal rule from the observed values.

3. Results and Discussion

In this study, the matrix tablets were prepred using various types of polymers and different composition. of polymers (Table 1) of matrix forming polymers (HPMC, sodium CMC and MCC) with the help of granulating agent, PVP was used as Binder. In vitro studies conducted for all the formulations. Extended release of drug was in the order of CW1 < CW2 < CW3 < CW4 < CW5 tablets with code no CW5 exhibited extended drug release up to 24 hours (Figure 1). Rate of drug release was significant (p < 0. 005) when combination of Povidone and HPMC Matrix builder gave the effect of hydrophilic type prolonged release of drug (Figure 1, CW5). It seems the mechanism is by diffusion method. Physical characteristics of matrix tablets were shown inTable 2.

There was no any significant burst effect in the optimized HPMC matrix tablets that showed a low possibility of dose dumping and avoids toxicity (in vivo). The Release kinetics of matrix tablets was determined by fitting the drug release data in different established models they are zero order, first order, Higuchi model, Korsemeyer-Peppas equation. Table 3shows values of regression coefficient, release constant and exponent n. First order release data was not satisfactory. The data suggested that kinetics of drug release of DVF5 was best explained by Korsemeyer-Peppas equation (R^2 = 0. 991, n = 0. 60). This indicated combined effect of diffusion and erosion mechanism on the release of drug.

The stability results of storing at 25ËšC/60% RH and 40ËšC/75% RH for 6 months as per ICH guidelines evidenced any change in physical parameters

and appearance and very slight change in dissolution pattern. Based on the available stability data 2 years shelf life can be provided.

Figure 1. In vitro release profile of Cilostazol SR tablets.

Table 2. Drug release kinetic of Cilostazol SR tablet formulations.

Next, the stable formula were designated for its in vivo test in rabbit. Plasma concentration and pharmacokinetic parameters after oral administration of formulated ER matrix tablet CW5 and Cilostazol IR tablets 50mg were summarized inFigure 2andTable 3. No sustained blood level was observed after oral administration of the IR formulation. The formulated matrix Tablet (CW5) showed significant lowerC $_{max}$ than the IR formulation ($P < 0.05$) and it required more time to reach C $_{max}$ (t $_{max}$ is 6 hr) as compared with immediate release formulation (t $_{max}$ is 0. 55 hr). The AUC increased from 11190. 30 hr*ng/ml to 295396. 49 hr*ng/ml for ER tablets. Values of C $_{max}$ and t $_{max}$ clearly indicated that the drug release was sustained to about 24 hours after oral administration in rabbits (n = 4). CW5 Tablets maintained prolonged plasma concentration up to about 24 hours. The sustained plasma concentration of new formulation (CW5) indicates its extended drug release in vivo absorption.

Table 3. Mean (±SD)
pharmacokinetic parameters of
Cilostazol in Rabbits (n = 4) orally
administered with IR tablets (50

mg) and ER tabletsCW5 (50 mg).

PK Parameters	Cilostazol	Cilostazol
Cmax (ng/mL)	3350 ± 284. 3	2400 ± 58
Tmax (hr)	0. 5	4
AUClast (hr*ng/mL)	11190. 30 ± 1629. 1	295396. 49 ± 1053. 62

The Results demonstrated that the hydrophilic polymers were successfully utilized for formulating Cilostazol extended release matrix tablets. By wet granulated with povidone . Moreover the extended release matrix tablets have a unique advantage of lessening chance of dose dumping and to avoid side effects. The investigated extended release matrix tablets were adequate to maintaining constant plasma level of Cilostazol up to 24 hours in rabbits.

Figure 2. Profile shows mean plasma concentration of Cilostazol against time, following oral administration of IR tablets and SR Tablets (CW5) to rabbits. Data are represented as mean ± SD (n = 4).

Table 3. Mean (±SD) pharmacokinetic parameters of Cilostazol in Rabbits (n = 4) orally administered with IR tablets and ER tablets CW5 (50 mg).

4. Conclusion

A new sustained release formulation of Cilostazol has been developed for its in-vitro drug release and in-vivo absorption. Extended release matrix tablet were found to be an effective to maintain the drug level in plasma. Bioavailability studies can be carried out to assess the usefulness of this formulationand in comparison with existing IR products in the market formulations on healthy human volunteers.