Available online at www.scholarsresearchlibrary.com

Scholars Research Library

Der Pharmacia Lettre, 2010, 2(5): 387-398
(http://scholarsresearchlibrary.com/archive.html)

ISSN 0975-5071
USA CODEN: DPLEB4

Preparation and Evaluation of HPMC and Eudragit Microparticles loaded with Diltiazem Hydrochloride for Controlled Release.

Gowda D.V*. Mohammed S. Khan, Venkatesh M.P, Sowjanya A.S., Shivakumar H.G

Dept. of Pharmaceutics, JSS College of Pharmacy, JSS University,
Sri Shivarathreeshwara Nagar, Mysore, Karnataka, India

ABSTRACT

The aim of the present study was to prepare and evaluate microparticles of Diltiazem Hydrochloride (DTZ) using blend of HPMC and Eudragit RS 100 by solvent evaporation technique for controlled release. Sieve analysis data indicated that the prepared microparticles were in the ranges of 265 to 187µm. The angle of repose, % Carr's index and tapped density were well within the limit, indicating reasonable good flow potential for the prepared microparticles. SEM photographs and calculated sphericity factor confirms the prepared formulations are spherical in nature. DSC studies and FT IR spectra showed that the encapsulated drug was stable in the prepared formulations. The prepared formulations were analyzed quantitatively for the amount of encapsulated drug. From the drug loading, encapsulation efficiency and in vitro drug release data, optimum formulation F7 was selected. The optimum formulation shows the drug release of 88.30 % up to 24 h having drug loading and encapsulation efficiency of 56.28% and 91.50 % respectively. It was also observed that, there was no significant release of drug at gastric pH. The release kinetics for all the formulations indicated that drug release followed non -Fickian diffusion. The release performance was greatly affected by the ratio of materials used in preparation of microparticles. Diltiazem Hydrochloride loaded microparticles have desirable release profiles and worthy of further investigation as an oral controlled release dosage form.

Key words: Diltiazem Hydrochloride, Microparticles, controlled release, Solvent Evaporation.

INTRODUCTION

High blood pressure is the leading risk factor for mortality around the world; it is much more than a "cardiovascular disease" as it not only affects heart but other organ systems of the body

such as kidney, brain, and eye. High blood pressure, termed "hypertension," is a condition that afflicts almost 1 billion people worldwide and is a leading cause of morbidity and mortality. [1]

Hypertensive is defined as an abnormal elevation in diastolic pressure and/or systolic pressure; mean arterial pressure is also elevated in hypertension, but it is not usually measured in people. In past years, the diastolic value was emphasized in assessing hypertension. However, elevations in systolic pressure ("systolic hypertension") are also associated with increased incidence of coronary and cerebrovascular disease (e.g., stroke). Therefore, we now recognize that both systolic and diastolic pressure values are important to note [2].

Hypertension is also commonly treated with drugs that decrease cardiac output. These cardio inhibitory drugs either block beta-adrenoceptors on the heart or L-type calcium channels, which decreases cardiac output by decreasing heart rate and contractility (inotropy). Vasodilator drugs, which decrease systemic vascular resistance, are also used to treat hypertension. Included in these drugs are alpha-adrenoceptors antagonists, direct, angiotensin-converting enzyme inhibitors and blockers. [3]

Conventional dosage forms often suffer from several drawbacks like repeated drug administration, fluctuations in drug blood levels which lead to untoward side-effects. Keeping this in mind, there has been an increasing effort to develop controlled release dosage forms for many drugs. Controlled release dosage forms have many advantages in safety and efficacy over immediate release drug products in that the frequency of dosing can be reduced, drug efficacy can be controlled and the incidence and/or intensity of adverse effects can be decreased.

Microencapsulation is a well-known method that is used to modify and delay drug release from pharmaceutical dosage forms. A great number of microencapsulation techniques are available for the formation of sustained release microparticulates drug delivery systems. One of the popular methods for the encapsulation of drugs within water insoluble polymers is the emulsion solvent evaporation method.

DTZ is a calcium ion cellular influx inhibitor [4] (slow channel blocker), widely used for the treatment of angina pectoris, hypertension and arrhythmias. To achieve maximum therapeutic effect with a low risk of adverse effects, controlled released preparations are preferred [5]. The solubility of diltiazem significantly decreases as the pH increases in the gastrointestinal tract. The controlled drug delivery system delivers the drug at a constant rate as it is transported from the region of low pH in the stomach to a region of higher pH in the intestine. Such controlled delivery results in a decrease in the frequency of drug administration thereby improving patient compliance. Furthermore, controlled drug delivery systems produce constant plasma levels of active ingredients as compared to fluctuations seen when multiple doses of a conventional formulation are prescribed. Thus, controlled drug delivery systems may decrease the severity and frequency of side effects. In addition to the use of a combination of defined ratios of hydrophilic and hydrophobic polymers, the use of a high quantity of polymers contributes towards ensuring a constant rate of release of the drug.

In the present study, an attempt has been made here to reduce the dosing frequency and to maintain the drug level at therapeutic concentration range, by formulating a controlled drug

delivery system of Diltiazem hydrochloride in the form of microparticles using HPMC and Eudragit RS100 polymer using solvent evaporation technique.

MATERIALS AND METHOD

Diltiazem HCl was obtained from Stadmed Pvt Ltd, Kolkata, India as a gift sample. Hydroxypropylmethylcellulose (HPMC) was obtained from Loba Chemie. Eudragit RS 100 was obtained from Vikram Thermo (Ahmedabad). All others reagent used are of analytical grade.

Preparation of Diltiazem Hydrochloride microparticles [6]

Drug-loaded microparticles were prepared by Solvent evaporation method. Weighed amounts of Diltiazem Hydrochloride and HPMC were dissolved in 10ml of water and this solution was added to 20ml of methanol solution containing Eudragit RS100 polymer under constant stirring at 1500 rpm using stirrer fitted with four blade impeller of approximately of about 53mm diameter .The resulted solution was poured into the liquid paraffin solution of 160ml and Tween 80 was added for emulsification of solution. The formed emulsion was stirred for about 4 h at 50ºC until the formation of microparticles. The resultant microparticles were vacuum filtered and then oven dried at 55ºC. The dried microparticles were stored in desiccators at room temperature. However, in the present study, various parameters were optimized such as drug and polymer ratio, stirring speed and time, addition of surfactant were studied during the preparation of microparticles. Therefore the influence of the above parameters can be highlighted. Formulation chart of the prepared formulations are shown in Table 1.

Table 1: Formulation chart of Diltiazem Hydrochloride loaded microparticles

INGREDIENTS	F1	F2	F3	F4	F5	F6	F7	F8	F9
Diltiazem HCl	240	240	240	240	240	240	240	240	240
HPMC	180	120	60	360	240	120	480	300	120
Eudragit RS100	60	120	180	120	240	360	120	300	480
Methanol (ml)	10	10	10	10	10	10	10	10	10
Water (ml)	20	20	20	20	20	20	20	20	20
Liquid Paraffin (ml)	160	160	160	160	160	160	160	160	160

Micromeritic Properties and Particle size analysis [7, 8]

Micromeritic properties like tap density, Carr index, angle of repose were calculated. Tap density of the prepared microparticles was determined using tap density tester and percentage Carr index (%I) was calculated. Angle of repose (θ) was assessed to know the flowability of the microparticles, by a fixed funnel method.

The particle size of the prepared microparticles was measured using a Malvern MASTERSIZER 2000 version 5.1 (Malvern, UK.) The drug loaded Diltiazem Hydrochloride microparticles were dispersed in 1:20 with methanol and measured at temperature of 37°C.

Sphericity of the microparticles [9]

The sphericity of the prepared drug loaded microparticles was determined by using a camera Lucida using the traces of the microparticles on a black paper. The tracings help to calculate the circulatory factor and confirm the sphericity of microparticles if the obtained values are nearer to 1.

The sphericity was determined by tracings of drug loaded microparticles (magnification 45x) which were taken on a black paper using camera Lucida, (Model -Prism type, Rolex, India) and circulatory factor (S) was calculated as

$$S = \frac{P^2}{12.56 \text{ X } A} \qquad (3)$$

Where A is area (cm^2) and P is the perimeter of the circular tracing

Scanning electron microscopy (SEM) [10, 11]

SEM photographs were recorded using scanning electron microscope Model Joel-LV-5600, USA, at suitable magnification at room temperature. The photographs were observed for morphological characteristics and to confirm shape of the microparticles.

Fourier Transform Infrared Spectroscopy (FT-IR) [12]

Drug polymer interactions were studied by FT-IR spectrophotometer (Shimadzu, 8033, USA) by KBr pellet method. The IR spectrum of the pellet from 400 – 4000 cm^{-1} was recorded.

Differential Scanning Calorimetry (DSC) [13]

All dynamic DSC studies were carried out on DuPont thermal analyzer with 2010 DSC module. The instrument was calibrated using high purity indium metal as standard. The dynamic scans were taken in nitrogen atmosphere at the heating rate of 10ºC/min heating rate of 10ºC/min.

Drug loading and encapsulation efficiency [14]

100 mg of Diltiazem Hcl microparticles were weighed and transferred to 100 ml volumetric flask containing pH 7.4 phosphate buffers. From this, 1 ml of solution was transferred to 10 ml volumetric flask and diluted up to the mark. Further 1 ml of this solution is diluted to 10 ml and absorbance was measured at 236 nm. The drug content was calculated by using the formula

$$\text{Amount of drug} = \frac{\text{Conc. from standard graph X dilution factor}}{1000} \qquad (5)$$

Percentage encapsulation efficiency is found out by calculating the amount of drug present in 100 mg of microparticles. It is further calculated by using formula:

$$\% \text{ Encapsulation Efficiency} = \frac{(b)}{A} \text{ X100} \qquad (6)$$

Where, A is the theoretical drug content and b is the drug entrapped.

***In vitro* drug release studies [15]**

The *in vitro* release of drug from the Diltiazem loaded microparticles (equivalent to 120mg of DTZ) was carried out in basket type dissolution tester USP XXIII, TDT-08L, with auto sampler containing 900 ml of pH 1.2 buffer for the first 2 hrs and in 7.4 pH phosphate buffer for the next 22 hrs. The volume of the dissolution media was maintained at 900 ml with constant stirring (100 rpm) and temperature of bath was maintained at 37 ± 0.5°C. Aliquots (10 ml) of dissolution media were sampled at specified time points and replaced with fresh media immediately after sampling. Samples were analyzed for drug content by UV Visible spectroscopy .The release data obtained were fitted into various mathematical models to know which mathematical model is best fitting for the obtained release profile.

The release profiles of DTZ from microparticles were studied and compared with Cardizem CD®.

RESULTS AND DISCUSSION

Microparticles are prepared using blends of hydrophobic and hydrophilic polymers using solvent evaporation method, and by using a non toxic solvent to entrap the drug which is different from other methods. Diltiazem Hcl is water soluble drug could be entrapped into water soluble and insoluble polymers by Solvent evaporation method.

Various formulation and process variables that could affect the preparation and properties of the microparticles were identified and optimized to get small, discrete, uniform, and spherical microparticles. The formulation variables included drug: polymers ratio: 1:1, 1:2, 1:2.5; HPMC and Eudragit RS100 ratio 1:3 ,1:1, 3:1 concentration of emulsifier Tween 80 (ml): 1, 2, 3, 4. The process variables included stirring speed: 1000, 1500, 2000, and 2500 rpm; stirring time: 1 h, 2 h, 3 h, 4 h; and temperature of the system: 30°C, 40°C, 45°C, and 50°C.

Results revealed that the average diameter and size of microparticles was controlled by agitation speed, stirring time, concentration of the polymers and surfactant used in the preparation. The average microparticles size increases with the polymer concentration while reduced with increase in agitation speed and stirring time; and at the higher speed gives irregular shape of the particles.

The formation of a stable emulsion in the early stages is important if discrete microparticles are to be isolated. It has also been found that the choice of solvent influences microparticles morphology depending on the rate at which it migrates from the polymer solution into the non-solvent phase and is removed by evaporation. The solubility of the polymer in the chosen solvent, affects the particles solidification. During this process the forming "particles" will evolve from being liquid emulsion droplets, to semi-solid "sticky" particles, to solidified, discreet particles. The length of time the particles exist in the semi-solid form is expected to influence coalescence of the forming particles and the overall morphology of the end product.

An optimal concentration of emulsifier is required to produce the finest stable dispersion. Below optimal concentration the dispersed globules/droplets tend to fuse and produce larger globules because of insufficient lowering in interfacial tension, while above the optimal concentration no

significant decrease in particle size is observed, because a high amount of emulsifying agent increases the viscosity of the dispersion medium. The optimal concentration of Tween 80 was found to be 2.0%. The average mean diameter of the microparticles showed 186.4µm which fall in the arbitrary scale of micro particles (1-1000 µm).The use of the surfactant permits the remarkable reduction in the size of the microparticles as the result of decrease in the interfacial tension.

Micromeritic properties

The obtained data angle of repose (θ) and % compressibility index (CI) along with related parameters are presented in Table 2. The values of θ^0 and CI ranged from 25.52 to 28.83 and 11.01 to 14.11% respectively indicating that the obtained values were well within the limits. This result clearly shows that the prepared microparticles have reasonably good flow potential.

The values of tapped density ranged between 0.492 to 0.543 g/cm^3. Density difference between the formulations is negligible and the density values of formulations were well within the limits, indicating that the prepared microparticles were non-aggregated and spherical in nature.

Table 2: Micromeritic properties Diltiazem Hydrochloride Microparticles

Formulation	θ^0 mean ± SD*	Carr's Index (%) mean ± SD*	Tapped density (gm/cm^3) mean ± SD*
F1	26.32± 0.46	12.35 ± 0.26	0.499 ± 0.04
F2	25.52 ± 0.19	11.01 ± 0.56	0.525 ± 0.01
F3	26.72 ± 0.37	14.11 ± 0.67	0.535 ± 0.03
F4	27.37 ± 0.35	13.33 ± 0.23	0.515 ± 0.02
F5	26.51 ± 0.27	12.22 ± 0.17	0.492 ± 0.06
F6	27.46 ± 0.32	12.45 ± 0.26	0.535 ± 0.06
F7	28.83 ± 0.65	11.22 ± 0.65	0.515 ± 0.02
F8	27.92±0.44	12.36 ± 0.44	0.495 ± 0.06
F9	27.99±0.92	13.42 ± 0.32	0.543 ± 0.02

*Standard deviation, n = 3

Particle size determination

The average particle size/volume mean diameter (D [4, 3]) and volume median diameters (D [v, 0.5]), (D [v, 0.9]) of the microparticles formulation of Diltiazem Hydrochloride (F7) are given in

Table 3 and the particle size graphs are given in Figure 1. D [4, 3] is the volume mean diameter and is the diameter of the sphere having the same volume as that of the microparticles whose size is being determined. D [v, 0.50] is the median diameter and it is the value of particle size that divides the population in to two equal halves i.e., there is 50% of distribution above this and 50% below this value. D [v, 0.90] is the median diameter and it is the cut off value for the distribution, which means 90% of the distribution is below this value. The microparticles showed the mean size of 186.4µm - 264.7 µm which fall in the arbitrary scale of microparticles (1-1000 µm).

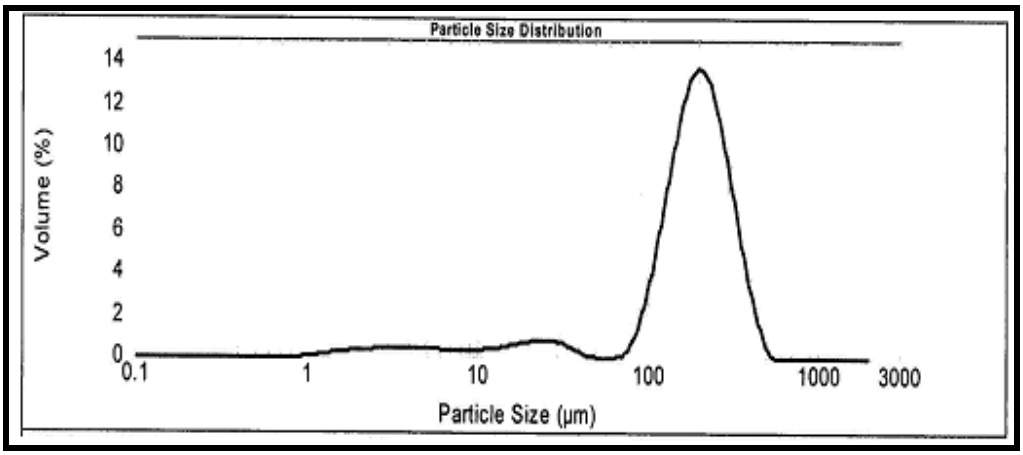

Figure 1: Particle size distribution of Diltiazem Hydrochloride microparticles formulation F7.

Table 3: Particle size distribution parameters of Diltiazem HCl microparticles Formulation F7

Formulation	Volume mean diameter (D[4,3]) µm	Volume median diameter (D[v,0.50]) µm	Volume median diameter (D[v,0.90]) µm
F7	186.4	200.3	264.7

Effect of Stirring speed on Particle size:

It was observed that with an increase in the stirring speed from 1500 to 2000 rpm, average size of the spheres decreased and difference in the sizes of the particles were significant. When the stirring speed was slower than 1000 rpm, the average size of the particles was found to be increased and resultant microparticles were composed of irregular masses, which were not possible to distinguish individual microparticles as shown in the Figure 2.

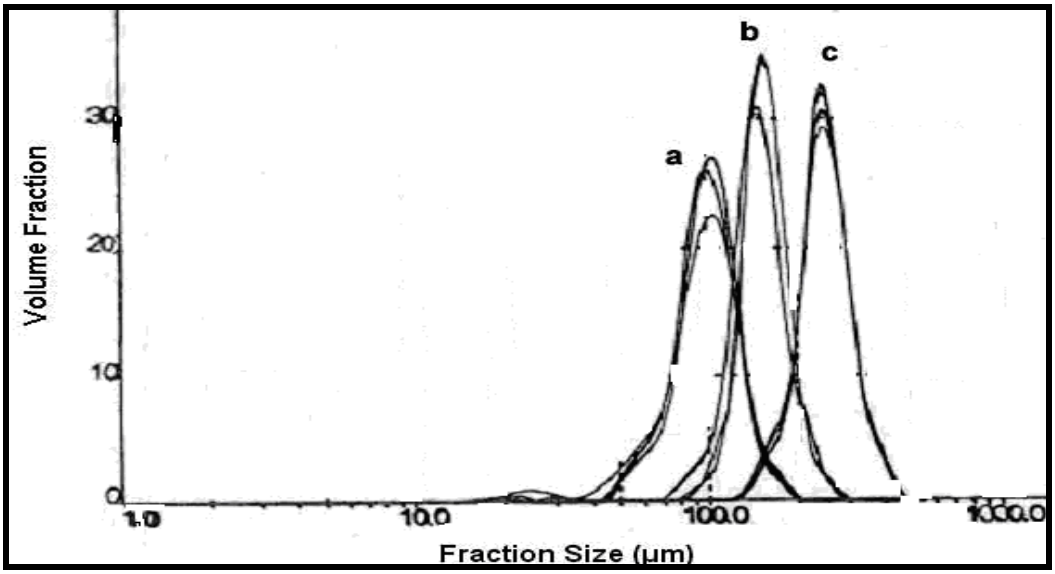

Figure 2: Effect of stirring rate on particle size of microparticles, a.1000rpm b.1500 rpm, c. 2000rpm

SEM and Sphericity

The scanning electron microscopy (SEM) studies were carried out to identify the morphology of the microparticles and the obtained microphotographs are presented in Figure 3. The SEM photographs showed that the microparticles were spherical in nature, having a smooth surface. SEM photographs reveal the absence of drug particles on the surface of microparticles showing uniform distribution of the drug in the walls of the microparticles. The sphericity factor was obtained in the range 1.00 to 1.06mm, indicating that the prepared formulations were spherical in nature.

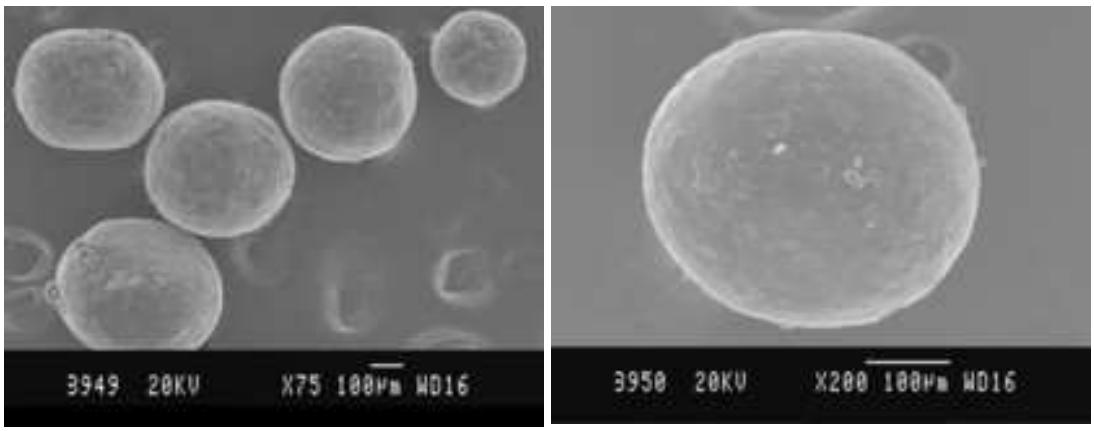

Figure 3: SEM photograph of the prepared formulation F7 at different magnifications.

Differential scanning calorimetric (DSC) studies

To understand the compatible state of the drug, DSC studies were carried out on pure drug, drug loaded microparticles and empty microparticles. The thermograms obtained are shown in Figure 4. Diltiazem Hcl exhibits a sharp endothermic peak at to 217.39^{0}C. It was observed that presence

of the endothermic peak at to 218.85^{0}C in the drug loaded microparticles indicated, that the drug retains its identity in the prepared microparticles. The melting points of the drug and Polymers were estimated by open capillaries and found agrees well with the DSC data.

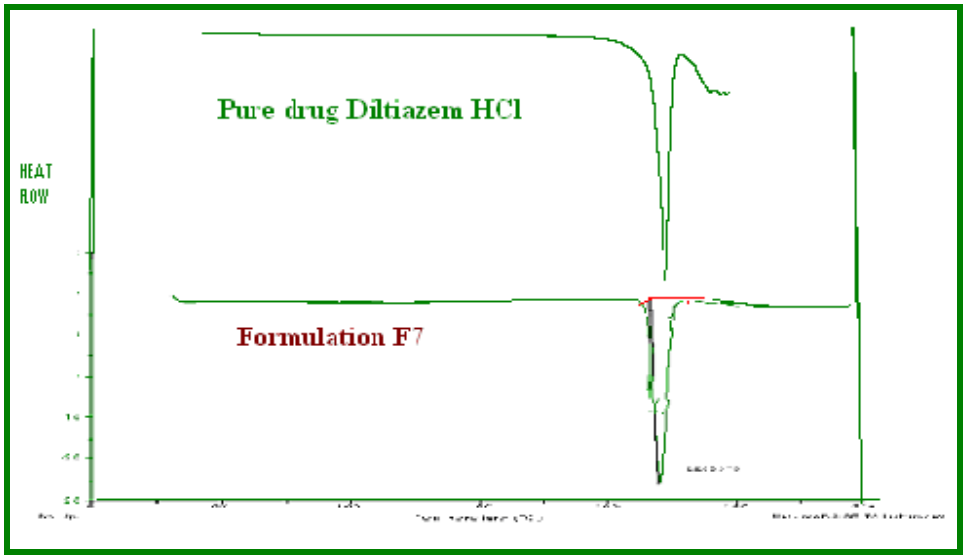

Figure 4: DSC comparison of Diltiazem Hydrochloride pure drug and formulation F7.

Fourier Transformed Infrared (FT IR) Spectroscopic Analysis:
FT IR spectra were obtained for of Diltiazem HCl pure drug and Diltiazem HCl loaded microparticles and are presented in **Figure 5**. The characteristic peaks of the pure drug were compared with the peaks obtained for formulation F7. From the data it is observed that a similar characteristic peak of Diltiazem Hydrochloride and Formulation F7 was appears with minor differences. The characteristics peaks found both in pure drug of Diltiazem and formulation F7, hence it appears there is no chemical interaction between drug and polymer and it can be concluded that the characteristics bands of pure drugs were not affected after successful loading.

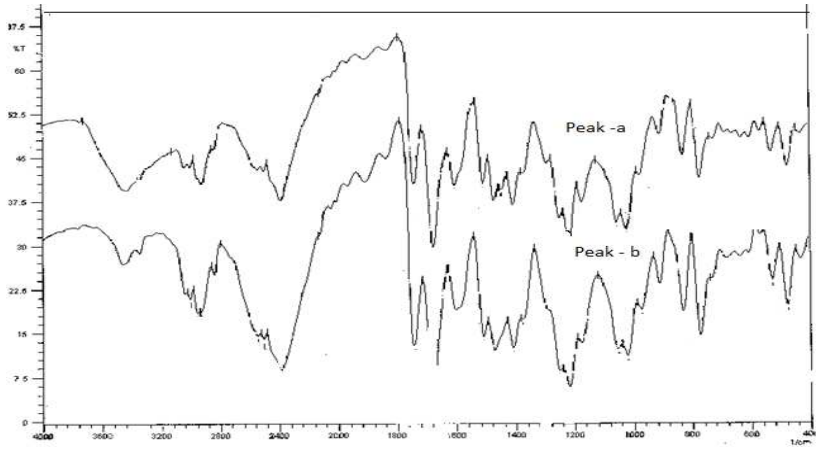

Figure 5: FTIR spectra of Diltiazem hydrochloride (peak a) and Diltiazem hydrochloride loaded microparticles (peak b – F7).

Drug loading and Encapsulation efficiency

The test for drug content was carried out to ascertain whether the drug is uniformly distributed in the formulation. Drug loading and entrapment efficiency increase with increase in the polymer concentration.

The percent of drug loading in the formulations was found to be in the range of 43.30% to 56.28 %. The percentage encapsulation efficiency was found to be 77.55 to 91.50 %. The results obtained are given in Table 4.

Table 4: Drug loading and encapsulation efficiency of prepared microparticles

Formulation	Drug loading(mg) mean ± SD*	Encapsulation efficiency (%) mean ± SD*
F1	43.30 ±0.12	77.55 ±0.32
F2	44.65 ±0.34	80.52 ±0.24
F3	46.44 ±0.24	74.32 ±0.25
F4	47.32 ±0.32	77.55 ±0.41
F5	49.16 ±0.16	80.50 ±0.24
F6	54.59 ±0.28	89.55 ±0.12
F7	56.28 ±0.22	91.50 ±0.24
F8	52.58 ±0.26	87.50 ±0.26
F9	49.64 ±0.38	83.70 ±0.32

*Standard deviation, n = 3

Table 5: % Yield of Diltiazem Hydrochloride loaded microparticles.

Formulation	% Yield ± SD*
F1	84.36 ± 1.24
F2	86.13 ± 1.84
F3	89.78 ± 1.43
F4	84.56 ± 1.63
F5	88.94± 1.33
F6	86.53 ± 1.21
F7	92.3± 1.63
F8	86.4± 1.82
F9	87.12 ± 1.38

*****Standard deviation, n = 3

Percentage yield

During the process of microencapsulation, the mechanical variables cause loss of final product and hence process yield may not be 100%. Microparticles were weighed after drying and the percentage yield was calculated. The obtained data is shown in **Table 5**.

***In-vitro* drug release**

From the release studies it was observed that, there is no significant release of drug at gastric pH from microparticles. Drug was released in a biphasic manner consisting of initial burst release followed by a slow release in intestinal pH from the microparticles. At the end of 24th hr, *in vitro* drug release from F1 (95.32%), F2 (93.51 %), F3 (90.30 %), F4 (89.64%), F5 (91.51 %) F6 (92.51 %), F7 (88.30 %), F8 (89.64%), F9 (93.32%), was slower than Cardizem CD® (96.30%) in the intestinal environment.

The release profile of microparticles in both media clearly indicates that the as the concentration of polymers increases, a decrease in release of DTZ from microparticles was observed. Increase in the concentration of HPMC retards the release of drug from the microparticles due to the hydroxy groups of the polymers which swells in contact with the aqueous media. The drug release profile of DTZ microparticles formulation (F7) were compared with marketed formulation Cardiazem CD shown in **Figure 6**.

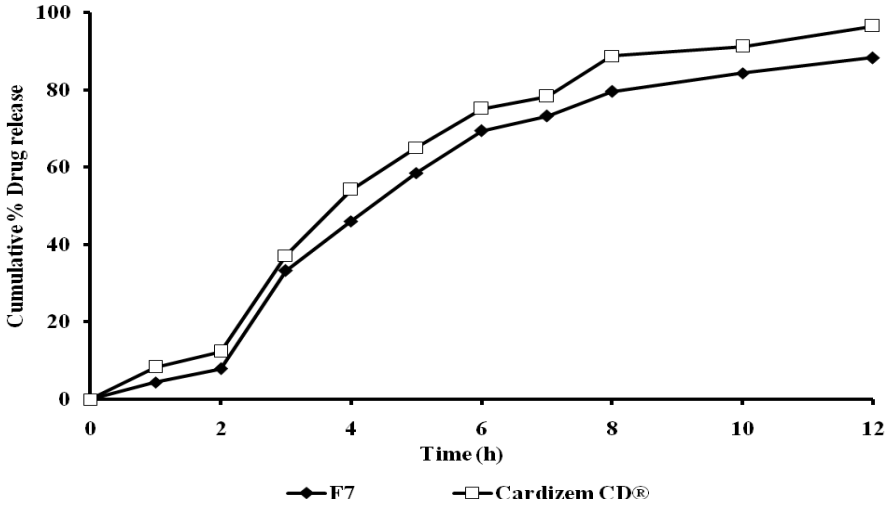

Figure 6: Percent drug release profiles of DTZ from formulation F7 and Cardizem CD® capsule in the gastric and intestinal environment against the time.

The *in vitro* release studies data was fitted in to various mathematical models to determine the best-fit model. The results indicated that, the best-fit models were found to be Peppas and Higuchi models. In all the cases the value of intercept, A were found to be more then 1.0. This indicates that the release of drug from all the formulations followed super case II transport. Super case II release is the drug transport mechanism associated with stresses and state-transition in hydrophilic glassy polymers which swell in water or biological fluids.

CONCLUSION

Diltiazem Hydrochloride loaded microparticles using HPMC and Eudragit RS100 was prepared using emulsification solvent evaporation method using different ratios of drug to polymer. The method is quite simple, rapid, and economical and does not imply the use of toxic organic solvents. The method used was most suitable for highly water-soluble drugs. The prepared spherical Diltiazem Hydrochloride exhibited good micromeritic properties. From the FT-IR spectra, it was observed that similar characteristic peaks appear with minor differences for the drug and their formulations. Hence, it appears that there was no chemical interaction between the drug and the polymer used. The DSC thermograms obtained for the pure drug and for the formulation shows no significant shift in the endothermic peaks confirming the stability of the drug in the formulation. The SEM studies clearly showed that the obtained microparticles exhibited good spherical nature. The *in vitro* drug release studies showed that, the release of drug was found to be diffusion controlled and the process followed zero order kinetics. Hence it is stated that Diltiazem Hydrochloride could be formulated into microparticles as controlled drug release dosage form.

REFERENCES

[1] A.V. Chobanian. *JAMA*. 289: 2560-72.
[2] J.A.Oater,''Antihypertensive agents and the drug therapy of hypertension "Goodman and Gillman's 'the pharmacological basis of Therapeutics"9thEdition, McGrew Hill, NewYork, 780-981(**1996**).
[3] G.A. Sagnella, P.A .Swift. *Current Pharmaceutical Design*. 12 (14): 2221-2234.
[4] A . Sordahl, E.F .LaBelle, K.A.Rex. *Cell Physiology* , 246: 172-176, (**2002**).
[5] Amnon Hoffman. *Adv Drug Del Rev* , 33: 185-199, (**1998**).
[6] J. Hermann, R. Bodmeier. *J.Pharm.Sci.* 13 (6): 747-60, (**1995**).
[7] K. Takayama, T. Nagai. *Chem. Pharm. Bull*. 37: 160-167, (**1989**).
[8] S .G. Yogesh, Durgacharan A Bhagwat, Akhil P Maske , *AAPS PharmSciTech*, 2 (4) : 228-231, (**2008**).
[9] T.W. Wong, L.W. Chan, H.Y. Lee, P.W. Heng. *J. Microencapsulation*. 19: 511-522, (**2002**).
[10] X. Yan, R.A. Gemeinhart. *J Control Rel*, 106: 198–208, (**2005**).
[11] K. Amit., S. Sitanshu, B. Lahiri., S. Harpal. *Int J Pharm* , 323: 117–124, (**2006**)).
[12] Y.Sultana, M. Shalini, D.P. Maurya, D. Kumar, M. Das. *Pharm Dev Tech*, 14: 321-331, (**2009**).
[13] T. Hekmatara, , G. Regdon, P. Sipos, I. Ers & K. Pintye-Hódi. *J. Therm. Anal. Cal*. 86: 287-90, (**2006**).
[14] X. Yan, R.A Gemeinhart. *J. Control Release*. 106: 198–208, (**2005**).
[15] D. Perumal, C.M. Dangor, R.S. Alcock, N. Hurbons , K.R.Moopanar. *J Microencap*. 16:475-87, (**1996**).