DARU Vol. 19, No. 1 2011

Formulation and in vitro characterization of cefpodoxime proxetil gastroretentive microballoons

*[1]Sharma AK., [1]Keservani RK., [2]Dadarwal SC., [3]Choudhary YL., [1]Ramteke S.

[1]School of Pharmaceutical Sciences, Rajiv Gandhi Proudyogiki Vishwavidyalaya, Bhopal, [2]Department of Pharmaceutics, Delhi Institute of Pharmaceutical Sciences and Research, University of Delhi, New Delhi, [3]Jaipur National University, Jaipur, Rajasthan, India.

Received 30 Oct 2010; Revised 2 Feb 2011; Accepted 10 Feb 2011

ABSTRACT

Background and the purpose of the study: The objective of the present work was to improve bioavailability of cepodoxime proxetil through gastroretentive microballoon formulation.
Methods: Microballoons of cefpodoxime proxetil were formulated by solvent evaporation and diffusion method employing hydroxypropylmethyl cellulose (HPMC) and ethyl cellulose (EC) polymers and characterized for particle size, surface morphology, incorporation efficiency, floating behavior, *in vitro* drug release study and differential scanning calorimetry (DSC).
Results: The average particle size of formulated microballoons was in the range of 54.23±2.78-95.66±2.19µm. Incorporation efficiencies of over 83.77±0.85 % were achieved for the optimized formulations. Most of formulations remained buoyant (having buoyancy percentage maximum of 81.36±1.96%) for more than 12 hrs indicating good floating behavior of microballoons. Higher values of correlation coefficients were obtained with Higuchi's square root of time kinetic treatment heralding diffusion as predominant mechanism of drug release.
Conclusion: Inferences drawn from in vitro studies suggest that microballoons may be potential delivery system for cefpodoxime proxetil with improvement in bioavailability in comparison to conventional dosage forms.

Keywords: Floating drug delivery system (FDDS), Solvent evaporation and diffusion method, Microparticulate carriers, Differential scanning calorimetry.

INTRODUCTION

Oral administration is the most convenient and preferred mean of drug delivery to the systemic circulation. Many attempts have been made to develop sustained-release preparations with extended clinical effects and reduced dosing frequency. In order to develop an oral drug delivery systems, it is necessary to optimize both the release rate of the drug and the residence time of the system within the gastrointestinal tract. Various approaches have been used to retain the dosage forms in the stomach (1-3), as a way of increasing the gastric residence time (GRT) including floating (4-7), high density (3), mucoadhesive (8), magnetic (9), unfoldable, extendible, or swellable (10), and superporous hydrogel systems (11). Both natural and synthetic polymers have been used to prepare floating microspheres.

Preparation of hollow microspheres or microballoons of ibuprofen by the emulsion-solvent diffusion method using acrylic polymers has been reported (12). These systems allow prolonged residence time of dosage forms in the stomach and achievement of constant plasma levels; however, it is necessary to analyze the gastrointestinal transit behavior in human to confirm the suitability of the concept as far as the final design is concerned (13).

Cefpodoxime proxetil (CP) is a prodrug of the third generation cephalosporins, which is broad-spectrum antibiotic and is administered orally. In human, the absolute bioavailability of cefpodoxime proxetil administered as a 130mg tablet (equivalent to 100mg of cefpodoxime) is about 50% (14). Reported studies have pointed possible reasons for low bioavailability as: low solubility, typical gelation behavior of CP particularly in acidic environments (15-17), and pre-absorption of luminal metabolism into cefpodoxime acid by the action of digestive enzymes (18, 19). It has been reported that the absorption of cefpodoxime proxetil is optimum at low pH (20).

The objective of the present work was to improve the bioavailability of cefpodoxime proxetil by formulating gastroretentive microballoons (hollow microspheres) in order to sustain the drug release and provide protection from intestinal milieu. In this study the influence of various process variables on particle size, drug loading, incorporation efficiency

Correspondence: sharma_pharm007@yahoo.co.in

and percentage yield, floating behavior and in vitro drug release of microballoon formulations was investigated.

MATERIAL AND METHODS

Materials

Cefpodoxime proxetil was obtained as a gift sample from Lupin laboratories Limited, (Pune, India). Hydroxypropyl methyl cellulose, Ethyl cellulose and Tween 80 were purchased from Loba Chem Private Limited, (Mumbai, India). Ethanol was supplied by S.D. fine Chem Limited, (Mumbai, India). Dichloromethane was purchased from CDH Limited, New Delhi, India. All chemicals/reagents used were of analytical grade.

Methods

Preparation of microballoons

Microballoons (hollow microspheres) were prepared by the solvent evaporation technique according to the reported method. (21). Cefpodoxime proxetil (130mg), HPMC and EC (1:1) were dissolved in a mixture of alcohol and dichloromethane (1:1) at room temperature. The resulting solution was poured into 250 ml of distilled water containing 0.01%(v/v) tween 80, maintained at room temperature and then stirred at different agitation speed for 20 min to allow the volatile solvent to evaporate. The microballoons formed were filtered, washed with water and dried.

Size and Shape of Microballoons

The size of microballoons was determined using a light microscope (BEM-21, Besto Microscope, India) fitted with an ocular micrometer and stage micrometer. Scanning electron microscopy (SEM) (Philips-XL-20, Netherlands) was performed to characterize the surface morphology of the formed microballoons. Microballoons were mounted directly onto the sample stub and coated with gold film (200 nm) under reduced pressure (0.133 Pa).

Drug loading (DL), Incorporation Efficiency (IE), and Percentage Yield

To determine the incorporation efficiency, microballoons (30mg) were thoroughly triturated and suspended in a minimal amount of alcohol, suitably diluted with 0.1 N HCl (pH1.2) and filtered to separate shell fragments. Amount of cefpodoxime proxetil (drug content/drug loading) was analyzed spectrophotometrically at 263 nm. In order to calculate the percentage yield, the prepared microballoons were collected and weighed. The incorporation efficiency and yield were calculated using the following equations:

$$\% \text{ IE} = \frac{\text{Calculated drug conc.}}{\text{Theoretical drug content}} \text{X } 100 \quad (1)$$

$$\% \text{ Yield} = \frac{\text{Total weight of floating microparticles}}{\text{Total weight of all non - volatile components}} \quad (2)$$

$$\text{DL} = \frac{\text{mass of the drug in microballoons}}{\text{mass of the recovered microballoons}} \quad (3)$$

In Vitro Drug Release

A USP paddle apparatus (Lab India, Mumbai, India) using 900 ml of 0.1 N HCl (pH 1.2) maintained at 37±0.5 °C with agitation speed of 75 rpm was used to study in vitro drug release (22). Samples were withdrawn at interval of 2 hrs and analyzed spectrophotometrically at 263 nm. The volume was replenished with the same amount of fresh dissolution fluid each time to maintain the sink condition.

Data analyses

Different kinetic equations (zero-order, first-order and Higuchi's equation) were applied on the release data of optimized batches to interpret the release pattern from matrix system (23-25). Drug released at specified time periods was plotted as percent drug release versus time curve (zero order kinetic treatment). Similarly log of % of the unreleased drug was plotted versus time curve (first order kinetic treatment) and percent of the drug release was plotted versus square root of time (Higuchi's Square root treatment).

Buoyancy test

Microballoons (0.3g) were spread over the surface of a USP (type II) dissolution apparatus (Lab India Mumbai, India) filled with 900 ml of simulated gastric fluid (pH 1.2). The medium was agitated with a paddle rotating at 75 rpm for 12 hrs. The floating and the settled portions of microballoons were recovered separately. The microballoons were dried and weighed. Buoyancy percentage was calculated as the ratio of the mass of the microballoons that remained floating and the total mass of the microballoons.

Differential scanning calorimetry (DSC)

Thermal analysis was carried out using a DSC unit (Pyris 6 DSC, Perkin-Elmer, Netherlands). Indium was used to calibrate the temperature scale and enthalpic response. Samples were placed in aluminum pans and heated at a scanning rate of 5°C/min from 30 to 400°C. Four samples i.e. pure cefpodoxime proxetil, pure HPMC, pure EC and cefpodoxime proxetil-loaded microballoons of HPMC and EC were analyzed.

RESULTS AND DISCUSSION

Preparation and optimization of microballoons

Since ethanol, as a good solvent for the polymers which preferentially diffuses out of dispersed droplets (organic phase) into aqueous phase was

Table 1. Effect of various processing parameters on the particle size, drug loading, incorporation efficiency (IE), yield and percentage of buoyancy of microballoons.

Formula-tion Code	Components			Yield (%)	Drug Loading (µg/mg)	IE (%)	Percentage buoyancy
	Polymer ratio‡	Solvent ratio	Mean particle size (µm)				
P 1	1:1	1:1	54.23±2.78	74.25±1.85	116.0±1.25	78.29±1.13	73.61±2.02
P 2	1:2	1:1	73.81±3.15	76.32±1.95	118.7±1.68	82.36±1.19	74.32±2.08
P 3	1:4	1:1	88.23±4.05	78.21±1.65	117.6±1.81	83.62±0.86	76.62±2.20
P 4	1:6	1:1	95.66±2.19	76.11±1.30	121.1±1.53	83.77±0.85	78.11±1.96
P 5	2:1	1:1	34.17±4.65	75.21±1.39	113.5±1.92	77.61±0.68	73.87±2.32
P 6	4:1	1:1	58.80±4.06	73.68±1.72	115.3±1.26	77.2±1.26	78.68±2.87
P 7	6:1	1:1	73.68±4.06	72.58±1.73	125.5±1.63	82.78±1.09	81.36±2.15
P 9*	1:2	1:1	71.06±3.82	76.32±1.14	114.0±1.58	79.11±1.66	76.32±1.84
P-10*	1:2	1:1	56.10±2.62	72.39±1.92	114.9±1.46	75.63±1.38	74.21±2.10
P 11	1:2	1:1	73.81±3.15	76.32±1.95	118.7±1.83	82.36±1.19	74.32±2.08
P 12	1:2	2:1	67.49±3.19	78.62±1.58	105.4±1.49	75.32±0.98	71.25±2.39
P 13	1:2	1:2	78.71±3.64	76.92±1.53	111.9±1.73	78.25±0.72	72.84±3.11
P 15†	1:2	2:1	71.91±3.49	78.41±1.93	110.4±1.85	78.68±1.04	74.32±1.77
P 16†	1:2	2:1	62.05±3.86	74.21±1.03	108.6±1.24	73.24±0.96	76.29±2.10

***** Formulations were prepared at varying agitation speed (250, 500 and 1000 rpm)

† Formulations were prepared at varying temperatures

‡ Polymer ratio (HPMC: EC)

used in this study, the polymer instantly solidified as a thin film at the interface between the aqueous and the organic phase. The yield of microballoons was a function of diffusion of solvents in the organic phase into aqueous phase. It has been reported that when the rate of the diffusion rate of solvent out of emulsion droplet is too slow, microspheres coalesced together. Conversely, when the diffusion of solvent is too fast, the solvent may diffuse into the aqueous phase before stable emulsion droplets are developed, causing aggregation of embryonic microsphere droplets (26). Results of this study showed that the formation of microballoons is a function of process variables such as polymer concentration, solvent composition, rate of agitation and temperature.

From the results of this study it was found that average particle size and wall thickness of microballoons increased by increase in the polymer concentration as it is apparent from observations of formulations P1 to P4 having average particle size in the range of 54.23±2.78-95.66±2.19 µm (Table 1). This may be attributed to increased viscosity of medium at higher polymer concentration resulting in enhanced interfacial tension. Shearing efficiency was also diminished at higher viscosities (27, 28). Which results in the formation of larger particles. It was obvious that speed of the rotation of the propeller affects the yield and size distribution of microballoons (Table 1). When the rotation speed of propeller was fast (1000 rpm), the average particle size decreased and their morphological characteristics were maintained. At low (250 rpm) rotation speed the shear force was not sufficient to form stable emulsion droplets, as a consequence larger droplets were formed and they were aggregated eventually. Thus optimum rotation speed for formulations of this study was medium i.e.500 rpm as reflected from results of particle size and incorporation efficiency (formulation P9). Solvent composition was found to be a vital factor in the formulation process governing the yield and particle size of microballoons (Table 1). As the amount of dichloromethane increased, the average particle size of microballoons was increased. Since alcohol preferentially diffused out of emulsion droplets, dichloromethane became a major constituent of the internal organic phase. The polymer, not being soluble at the interface between dichloromethane and aqueous phase, started to solidify around dichloromethane rich emulsion droplets and the volume of dichloromethane within the droplets became a size determining factor. The content of dichloromethane also affected the morphology of microballoons and best results were obtained at the ratio of alcohol to dichloromethane of 2:1 (formulation P12).

The temperature of the dispersing medium was an important factor in the formation of microballoons, because it controls the rate of evaporation of the solvents (Table1). At lower temperature (8-10°C), the prepared microballoons had irregularly shaped surface morphology and the shell was translucent

Table 2. Kinetic treatment of drug release data of cefpodoxime proxetil microballoons.

Formulation Code	Zero order*		First order*		Higuchi's square root of time*	
	K_0	r^2	K_1	r^2	K_H	r^2
P-2	0.1215	0.909	10.517	0.983	0.0347	0.990
P-3	0.1597	0.921	23.008	0.954	0.0447	0.966
P-9	0.1242	0.900	11.517	0.983	0.0357	0.991
P-12	0.1211	0.883	10.312	0.964	0.0351	0.981
P-15	0.1215	0.909	10.5173	0.981	0.0.347	0.993

* K_0 (h^{-1}), K_1 (h^{-1}) and K_H ($h^{-1/2}$) are release rate constants for Zero, First and Higuchi's kinetic treatment, respectively.

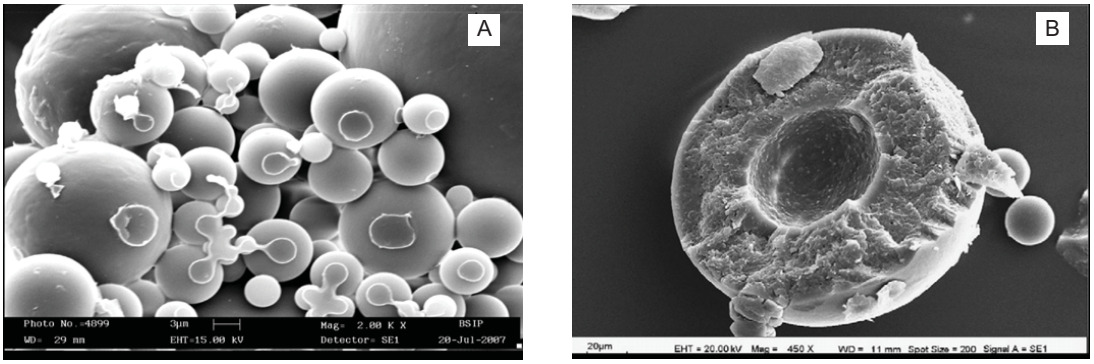

Figure 1. Scanning electron micrograph of microballoons. A. Outer surface of microballoons, B. Inner surface of a broken half of a microballoon.

during the process, due to the slower rate of diffusion of ethanol. At higher temperatures, the shell of the microballoons was very thin and some of them were broken (formulation P16) which might be due to the faster diffusion of alcohol of the droplet into aqueous phase and immediate evaporation of dichloromethane after introduction into the medium. The optimum temperature for present study, to form microballoons with good floating properties was room temperature (formulation P15).

Characterization of microballoons

By observation it was apparent that microballoons composed of HPMC at higher ratio (formulations P5, P6 and P7) were smaller in size as compared to microballoons with high EC content (Table1). Scanning electron micrograph (Fig 1) revealed that by using solvent diffusion and evaporation method spherical shaped microballoons with smooth outer surface and hollow core were formed. Incorporation efficiency of formulated microballoons was a function of process variables as well as the physicochemical properties of drug. It was observed that variation in polymer concentration influenced incorporation efficiency (Table 1). Increase in viscosity at higher polymer concentration restricted the movement of drug from polymer matrix into aqueous phase. Solubility of drug in organic solvents also played an important role in determination of the incorporation efficiency. Cefpodoxime proxetil was soluble in both alcohol and dichloromethane. The drug was hydrophobic, therefore; its leaching into aqueous phase was minimum. The increase in polymer concentration had no impact on the percentage yield of microballoons. There was no significant burst effect from any of the preparations. Guiziou *et al.* have reported significant burst release of the drug from poly (lactide) microspheres prepared by solvent evaporation method (29). Results indicate that proportion of polymers in formulation was the key factor governing release of drug from microballoons. As the concentration of polymer increased, there was an increased in diffusional path length. This may decrease the overall drug release from the polymer matrix. Formulation comprised of EC in higher proportion exhibited much retarded drug release as compared to HPMC formulations (Figs 2A and 2B). Another process variable influencing drug release was agitation speed. At higher rotation speed smaller microballoons were formed resulting in higher drug release. (Fig 3). No significant effect of solvent composition was observed on the in vitro release of cefpodoxime proxetil (Fig. 4). It is apparent that the drug release, from microballoons prepared at high temperatures, was slightly higher by virtue of thin shell of the microballoons (Fig 5).

Higher values of correlation coefficients were obtained in the case of Higuchi's square root of

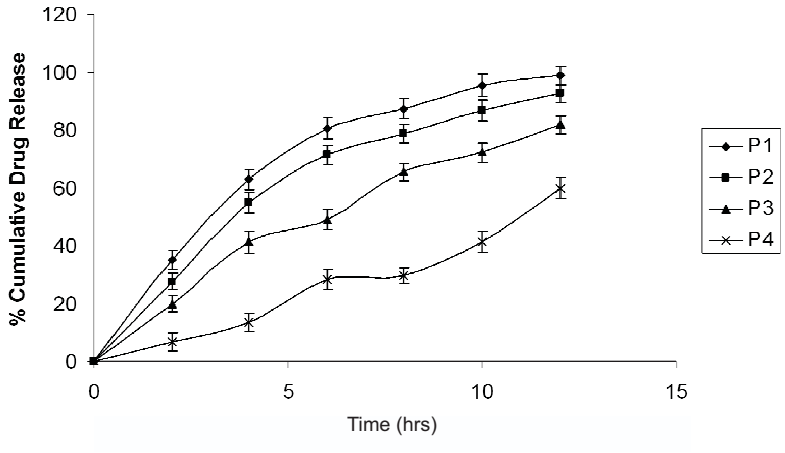

Figure 2a. Release profile of cefpodoxime proxetil from microballoons containing varying concentrations of EC.

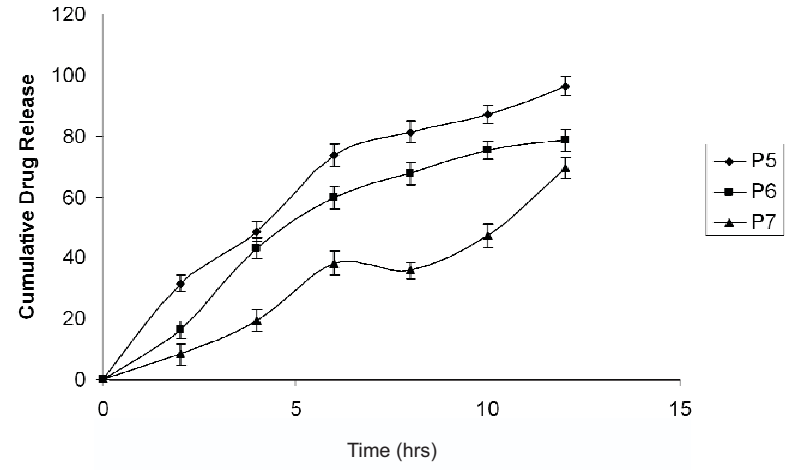

Figure 2b. Release profile of cefpodoxime proxetil from microballoons containing varying concentration of HPMC

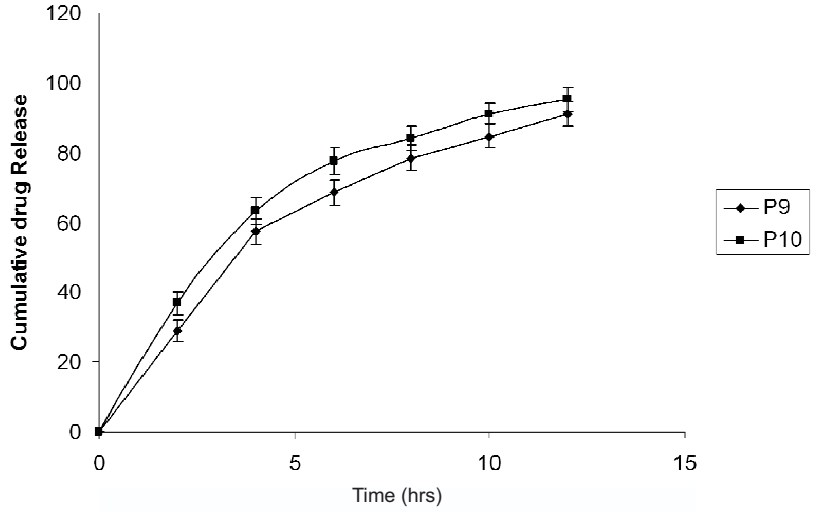

Figure 3. Release profile of cefpodoxime proxetil from microballoons formulated at different agitation speed.

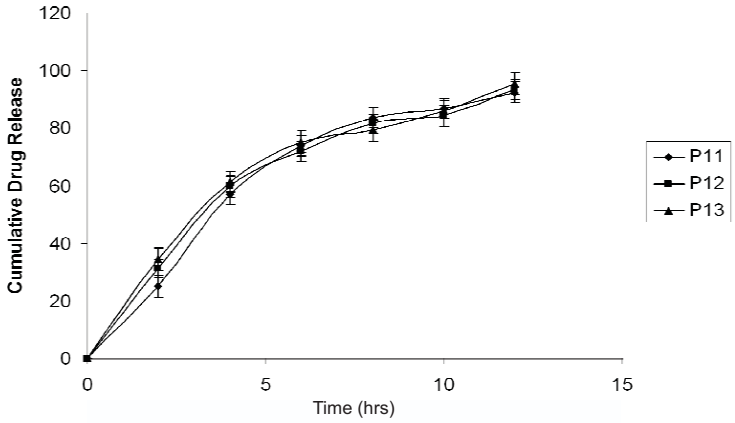

Figure 4. Release profile of cefpodoxime proxetil from microballoons prepared by varying solvent composition.

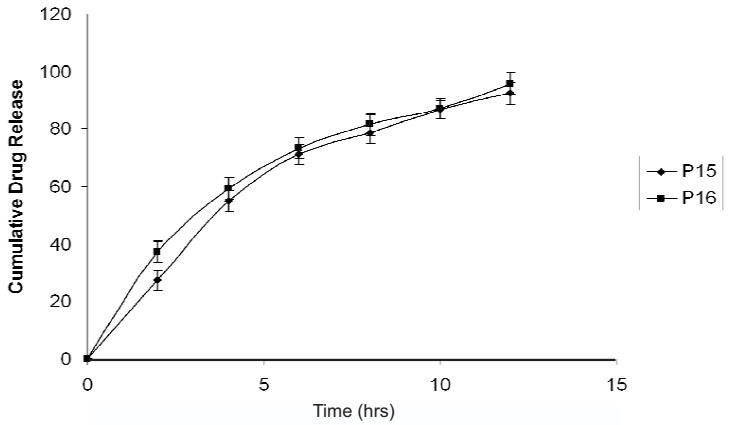

Figure 5. Release profile of cefpodoxime proxetil from microballoons prepared at different temperatures.

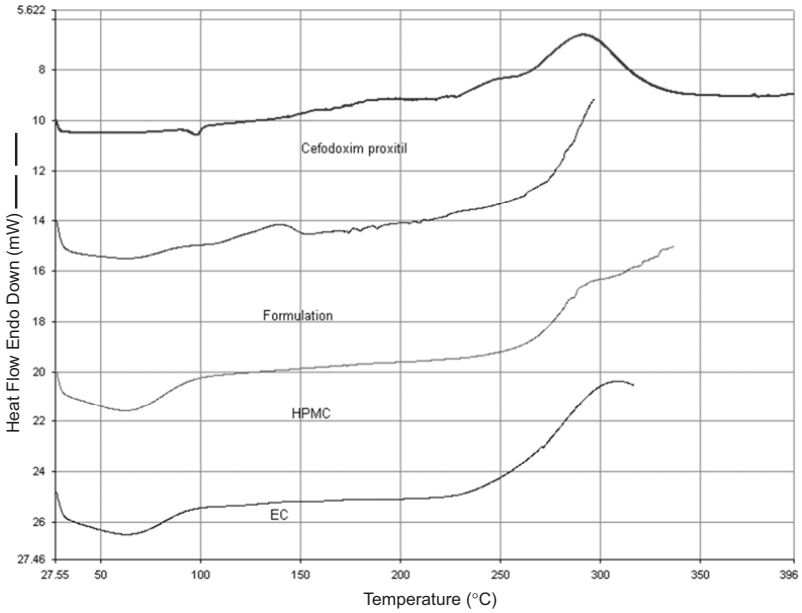

Figure 6. Overlap Differential Scanning Calorimetry (DSC) thermogram of pure drug, polymers and microballoon formulation.

time kinetic treatment (Table 2) which may indicate that diffusion was predominant mechanism of drug release.

The microballoons were spread over the surface of simulated gastric fluid (pH 1.2). The floating and the settled portions of microballoons were recovered separately. It was obvious from results that most of the prepared microballoons remained floating for longer than 12 hrs thereby releasing the drug in dissolution media in sustained manner (Table1). The result also showed a tendency that the larger the particle size, the longer the floating time. It should be noted, however, that the in vivo situation can be quite different and the residence time may vary widely depending on the phase of gastric motility.

DSC thermograms of microballoons along with those of drug and polymers are depicted in Fig 6. DSC thermogram of the cefpodoxime proxetil-loaded microballoons revealed that the drug existed in the amorphous state in the shell of microballoons, irrespective of the drug content, as indicated by peak pattern of DSC thermograms of formulation as well as polymers heralding absence of interactions between drug and polymers under study.

CONCLUSION

The microballoons so prepared will remain buoyant on surface of gastric fluid releasing cefpodoxime proxetil in sustained fashion. Inferences drawn from in vitro studies suggest that microballoons may prove as potential delivery system for cefpodoxime proxetil by improving bioavailability in comparison to conventional dosage forms.

ACKNOWLEDGEMENTS

Authors would like to express sincere gratitude towards Prof. A.R. Kulkarni, head, for providing sophisticated analylitical instrument facility, IIT Bombay, Mumbai for DSC analysis and Dr. N. C. Mehrotra, director, Birbal Sahni Institute of Paleabotany, Lukhnow for SEM study.

REFERENCES

1. Moes AJ. Gastroretentive dosage forms. Crit. Rev. Ther. Drug Carr. Syst. 1993; 10: 143-195.
2. Deshpande AA, Rhodes CT, Shah NH, Malick AW. Controlled-release drug delivery systems for prolonged gastric residence: an overview. Drug Dev. Ind. Pharm. 1996; 22:531-539.
3. Hwang SJ, Park H, Park K.Gastric retentive drug-delivery systems. Crit. Rev. Ther. Drug Carr. Syst. 1998; 15: 243-284.
4. Yuasa H, Takashima Y, Kanaya Y. Studies on the development of intragastric floating and sustained release preparation: I. Application of calcium silicate as a floating carrier. Chem.Pharm. Bull. 1996; 44:1361-1366.
5. Rouge N, Cole ET, Doelker E, and Buri P. Buoyancy and drug release patterns of floating minitablets containing piretanide and atenolol as model drugs. Pharm. Dev. Technol.1998; 3: 73- 84.
6. Lee JH, Park TG, Choi HK. Development of oral drug delivery system using floating microspheres. J. Microencapsul.1999;16: 715-729.
7. Tripathi G, Singh S. Formulation and In Vitro evaluation of pH sensitive oil entrapped polymeric blended gellan gum buoyant beads of clarithromycin, DARU 2010;18: 247-253.
8. Akiyama Y, Nagahara N, Kashihara T, Hirai S, Toguchi H. In vitro and in vivo evaluation of mucoadhesive microspheres prepared for the gastrointestinal tract using polyglycerol esters of fatty acids and a poly (acrylic acid) derivative. Pharm. Res. 1995;12: 397-405.
9. Groning R, Berntgen M, Georgarakis M. Acyclovir serum concentrations following peroral administration of magnetic depot tablets and the influence of extracoporeal magnets to control gastrointestinal transit. Eur. J. Pharm. Biopharm. 1998; 46: 285-291.
10. Fix JA, Cargill R, Engle K. Controlled gastric emptying: Part 3. Gastric residence time of a nondisintegrating geometric shape in human volunteers. Pharm. Res. 1993; 10:1087-1089.
11. Park K. Enzyme-digestible swelling hydrogels as platforms for long-term oral drug delivery: synthesis and characterization. Biomaterials. 1988; 9: 435-441.
12. Kawashima Y, Niwa T, Takeuchi H, Hino T, Itoh Y. Hollow microspheres for use as a floating controlled drug delivery system in the stomach. J. Pharm. Sci. 1992; 81:135-140.
13. Whitehead L, Fella JT, Colletta JH, Sharma HL, Smith AM. Floating dosage forms: an *in vivo* study demonstrating prolonged gastric retention. J. Control Release. 1998; 55: 3-12.
14. Borin T. A review of the pharmacokinetics of cefpodoxime proxetil. Drugs. 1991; 42:13-21.
15. Hamamura T, Kusai A, Nishimura K. Gel formation of cefpodoxime proxetil. S.T.P. Pharm. Sci. 1995; 5: 324-331.
16. Hamamura T, Ohtani T, Kusai A, Nishimura K. Unusual dissolution behavior of cefpodoxime proxetil: effect of pH and ionic factors. S.T.P. Pharm. Sci. 1995; 5: 332-338.
17. Hamamura T, Terashima H, Ohtani T, et al. Decrease in dissolution of cefpodoxime proxetil tablets by gel formation and its improvement. Yakuzaigaku. 1995; 55: 175-182.
18. Crauste-Manciet S, Huneau JF, Decroix MO, Tome D, Chaumeil JC. Cefpodoxime proxetil esterase

activity in rabbit small intestine: role in the partial cefpodoxime absorption. Int. J. Pharm. 1997; 149: 241-249.
19. Kakumanu VK, Arora V, Bansal AK. Investigation of factors responsible for low oral bioavailability of cefpodoxime proxetil. Int. J. Pharm. 2006; 317:155-160.
20. Hughes GS, Heald DL, Barker KB, et al. The effects of gastric pH and food on the pharmacokinetics of a new oral cephalosporin, cefpodoxime proxetil. Clin. Pharmacol. Ther. 1989; 46: 674-85.
21. Streubel A, Siepmann J, Bodmeier R. Multiple unit gastroretentive drug delivery systems: a new preparation method for low density microparticles. J. Microencapsul. 2003; 20: 329-347.
22. US Pharmacopoeia XXVI. US Pharmacopoeial Convention, Rockville, MD, 2003:378-380.
23. Costa P, Lobo JMS. Modeling and comparison of dissolution profiles. Eur. J. Pharm. Sci. 2001; 13:123-133.
24. Wagner JG. Interpretation of percent dissolved-time plots derived from *in vitro* testing of conventional tablets and capsules. J. Pharm. Sci. 1969; 58: 1253-1257.
25. Schefter E, Higuchi T. Dissolution behavior of crystalline solvated and non-solvated forms of some pharmaceuticals. J. Pharm. Sci. 1963; 52: 781-791.
26. Kawashima Y, Iwamoto T, Niwa T, Takeuchi H, Hino T. Role of the solvent-diffusion-rate modifier in a new emulsion solvent diffusion method for preparation of ketoprofen microspheres. .Microencap-sul. 1993; 10:329-340.
27. Reddy BP, Dorle AK, Krishna DK. Albumin microspheres: effect of process variables on the distribution and *in vitro* release, Drug Dev. Ind. Pharm. 1990; 16:1781-1803.
28. Ishizaka T. Preparation of egg albumin microspheres and microcapsules, J. Pharm. Sci. 1981; 70:358-361.
29. Guiziou B, Amstrong DJ, Elliot PNC, Ford JL. Rostron C. Investigation of in-vitro release characteristics of NSAID-loaded polylactic acid microspheres, J. Microencapsul. 1996; 13:701-708.