ISSN: 0975-8585

Research Journal of Pharmaceutical, Biological and Chemical Sciences

Formulation and *In-vitro* evaluation of enteric microspheres of rabeprazole sodium

Tank Nishit A[1*], GS Shantha Kumar[1], Prakasam Kalyani[1], Tank Nimit A[1], Paradava Dhaval S[1]

[1]Acharya & B.M Reddy College of Pharmacy, Chikkabanavara Post, Bangalore, Karnataka, India, -560090.

ABSTRACT

The aim of the present study was to prepare and evaluate microspheres of Eudragit (S100 and L100) containing an PPIs drug Rabeprazole Sodium. Microspheres were prepared by O/O solvent evaporation method using methanol/liquid paraffin system. Magesium stearate was used as the droplet stabilizer and n-hexane was added to harden the microspheres. The prepared microspheres were characterized for their micromeretic properties and entrapment efficiency; as well by Fourier transform infrared spectroscopy (FTIR), scanning electron microscopy (SEM) revealed the crystalline nature of drug in a final stage. The *in vitro* release studies were performed in pH 1.2 (0.1 N HCl) for 2 h followed by 7.2 pH phosphate buffer for 10 h. The best fit release kinetics was achieved with a Zero order. The yield of preparation and entrapment efficiencies were very high with a larger particle size for all the formulation. Mean particle size, entrapment efficiency and production yield were highly influenced by the type of polymer and polymer concentration. It is concluded from the present investigation that Eudragit S100 are promising controlled release carrier for Rabeprazole Sodium.

Keywords: Rabeprazole Sodium, Eudragit S100, Eudragit L100, microspheres, controlled release.

Corresponding author

ISSN: 0975-8585

INTRODUTION

Rabeprazole Sodium belongs to the class of anti-secretory compounds that do not exhibit anticholinergic or histamine H_2-receptor antagonist properties, but suppress gastric acid secretion by inhibiting the gastric H^+/K^+-ATPase at the secretory surface of the gastric parental cell [1-2]. Rabeprazole Sodium is a gastric labile drug and get degraded in stomach pH[1]. These dis-advantages make it an appropriate candidate for microencapsulation. Microspheres are one of the multiparticulate delivery systems and are prepared to obtain prolonged or controlled drug delivery to improve bioavailability or stability and to target drug to specific sites. Microspheres can also offer advantages like limiting fluctuation within therapeutic range, reducing side effects, decreasing dosing frequency and improving patient compliance [3]. Eudragit polymers are a series of acrylate and methacrylate polymers available in different ionic forms. Eudragit S100 and Eudragit L100 are insoluble in aqueous media, but they are permeable and have pH dependent release profile. The aim of the study was to prepare Eudragit microspheres containing Rabeprazole Sodium to achieve a prolonged release and specific site targeting drug delivery system profile suitable for oral administration. The microspheres were prepared by a solvent evaporation technique using Eudragit as a matrix polymer. Liquid paraffin and methanol system were used for the preparation of microspheres. Magnesium stearate was used as a droplet stabilizer to prevent droplet coalescence in the oil medium and n-hexane was added as a non-solvent to the processing medium to solidify the microspheres [4]. Firstly, we investigated formulation variables (polymer type and drug:polymer ratio) to obtain spherical particles. The effects of various Eudragit on the yield of production, particle size distribution, encapsulation efficiency, surface properties and Rabeprazole Sodium release rate from microspheres were investigated. The influences of formulation variables on the microspheres properties were examined. The prepared spherical microspheres were evaluated for micromeretic properties and drug content, and also by FTIR and SEM, as well as for *in vitro* drug release studies [4].

EXPERIMENTAL

Materials

Eudragit S100, Eudragit L100, Evonik Industries; Rabeprazole Sodium, Enal Drug Pvt Ltd; Magnesium Stearate, n-hexane, methanol, liquid paraffin, petroleum ether, Karnataka Fine Chemicals; Other substances used were all of analytical grade.

Preparation of microspheres

The technique used in preparation of microspheres is a “O/O emulsion solvent evaporation technique”. As shown in Table 1, three different formulation of each polymer (Eudragit S100, Eudragit L100) and three formulations of a mixture of Eudragit S100 and Eudragit L100 with drug (Rabeprazole Sodium, 300 mg) were prepared. The polymers were dissolved in 15 ml of methanol separately. Pure Rabeprazole Sodium was dissolved in 5 ml of

methanol. Both the solutions were mixed and 30 mg of magnesium stearate was dispersed in solution containing polymer and Rabeprazole Sodium. The dispersion was then stirred for 15 min using a magnetic stirrer. The resultant dispersion was then poured into a 500 ml beaker containing the external phase (40 ml of liquid paraffin heavy + 3 ml of n-hexane) while stirring, using a three blade mechanical stirrer. Stirring (at 700-800 rpm) was continued for 3-4 h until methanol had evaporated completely. After evaporation of the solvents, the microspheres formed were filtered using Whatman no. 41 filter paper. The residue was washed 4-5 times in 25 ml of n-hexane followed by 4-5 times in 50 ml of petroleum ether. Therefore, the microspheres were dried in a desiccator for 24 h at room temperature [4].

Table 1. Formulae for Rabeprazole Sodium loaded Eudragit microspheres

Formulation code	Eudragit S100 (mg)	Eudragit L100 (mg)	Rabeprazole Sodium (mg)
F1	300		300
F2	600		300
F3	900		300
F4		300	300
F5		600	300
F6		900	300
F7	300	300	300
F8	600	300	300
F9	300	600	300

Production yield

The yield was calculated by dividing the weight of the collected microspheres by the weight of all the non-volatile components used for the preparation of microspheres and expressed in the terms of percentage [5].

Percent Yield = (the amount of microspheres obtained/the theoretical amount) * 100

Particle size distribution analysis

Formulations of the microspheres were analyzed for particle size by optical microscope. The instrument was calibrated and found that 1 unit of eyepiece micrometer was equal to 13.33 µm. 100 microspheres sizes were calculated under 10x magnification [6].

Drug entrapment efficiency (DEE)

About 10 mg equivalent Rabeprazole Sodium loaded microspheres were dissolved in 100 ml of PBS (pH 7.2) by shaking on bottle shaker for 12 h. The solution was filtered through Whatman no. 41 filter paper. An aliquot was assayed spectrophotometrically (UV-1701 Schimadzu corporation, Japan) for Rabeprazole Sodium at 284 nm. Drug entrapment efficiency was determined by using the following relationship.

% Entrapment = (Actual content/Theoretical content) * 100

In vitro drug release study

The dissolution rate of Rabeprazole Sodium from the microspheres was studied at pH 1.2 for 2 h followed by pH 7.2 for 10 h using the basket method. Accurately weighed microspheres (equivalent to 20 mg of Rabeprazole Sodium) were taken for dissolution studies. The dissolution medium was kept at 37 ± 0.5°C. Aliquots of sample were withdrawn at predetermined intervals of time and analyzed for drug release by measuring the absorbance at 284 nm. The volume withdrawn at each time intervals was replaced with the same amount of fresh dissolution medium.

Release kinetics

Data obtained from *in vitro* release studies were fitted to various kinetic equations to discover the mechanism of drug release from microspheres. The kinetic models used were Zero order, Korsemeyer-Peppas, and Higuchi. The rate constants were also calculated for the respective models [4].

FTIR study

Drug polymer interactions were studied by FTIR spectroscopy. IR spectra for drug and drug loaded Eudragit microspheres were recorded in a Fourier transform infrared (FTIR) spectrophotometer (Bruker, Tensor-27, Germany). The Scanning range was 400-3500 cm^{-1}.

Scanning electron microscopy (SEM)

Scanning electron microscopy was used to examine the surface morphology of microspheres. Dried microspheres were mounted on to stubs by using double-sided adhesive tape. The microspheres were coated with gold and observed under a scanning electron microscope (Joel, JSM-5600 LV, Japan) for surface characteristics.

RESULTS AND DISCUSSION

Mean particle size

In the present work, the microspheres of Eudragit S100 and Eudragit L100 were prepared by "O/O-emulsification solvent evaporation" technique using methanol/liquid paraffin system. The drug was dissolved in 1 ml methanol and polymers were dissolved in 15 ml of methanol separately, then 30 mg magnesium stearate was dispersed into it. It was then dispersed into the external phase containing 40 ml of heavy liquid paraffin and 3-4 ml of n-hexane. The effects of parameters like the type of polymer and polymer concentration on the

ISSN: 0975-8585

production yield, entrapment efficiency, particle size distribution, *in vitro* drug release, surface characteristic and drug polymer interaction were studied. As shown in table 2, the mean size of the formulation of Eudragit S100 (F1-F3) found in the range of 341.10 to 453.39 µm, for Eudragit L100 (F4-F6) found in the range of 154.40 to 164.33 µm and for Eudragit S100 and L100 combination the range was 143.59 to 146.73 µm. The data revealed that particle size was highly influenced by the type of polymer and polymer concentration [3,7-10].

Production yield

The production yields obtained were very high for all the formulations. As shown in table 2 and in figure 1, the % yield of the formulations of Eudragit S100 (F1-F3) found in the range of 76.85% to 82.12%, for Eudragit L100 (F4-F6) found in the range of 77.20% to 83.25% and formulation of Eudragit S100 and L100 combination (F7-F9) found in the range of 80.02% to 96.16%.

Entrapment efficiency

Table 2. Physicochemical Parameters of Rabeprazole Sodium loaded Eudragit microspheres

Formulation Code	Percentage Yield(%)	Percentage DEE*(%)	Particle Size(µm)	Angle of Repose*(θ)	Flow Properties
F1	76.8	94.29 ± 0.26	341.10	22.58° ± 0.45	Excellent
F2	77.22	92.06 ± 2.68	344.80	26.56° ± 0.48	Good
F3	82.12	91.33 ± 3.87	453.39	29.24° ± 0.54	Good
F4	83.25	93.64 ± 0.66	154.40	27.51° ± 0.63	Good
F5	79.2	88.67 ± 0.51	164.33	29.24° ± 0.54	Good
F6	77.20	83.08 ± 0.53	136.42	30.11° ± 0.69	Passable
F7	96.16	94.13 ± 0.69	146.40	24.7° ± 0.42	Excellent
F8	80.02	88.96 ± 0.69	143.59	29.24° ± 1.06	Good
F9	92.42	84.59 ± 0.52	146.73	28.3° ± 0.55	Good

*All readings are mean of three reading

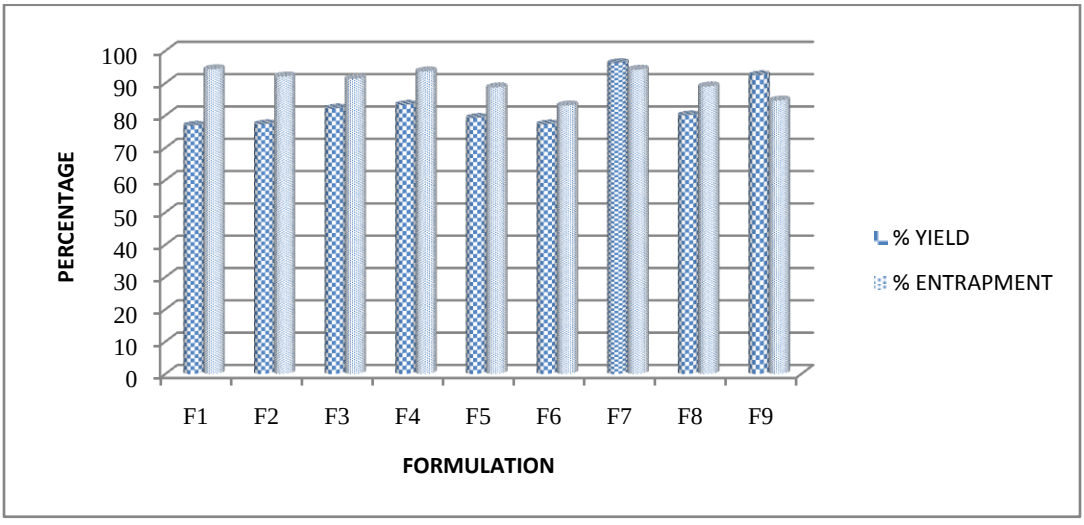

Fig.1.Yield of preparation and encapsulation efficiency data of formulation F1-F9.

ISSN: 0975-8585

As shown in table 2 and figure 1, high entrapment efficiency of the drug was obtained for all Eudragit formulations. The % entrapment efficiency of the formulations of Eudragit S100 (F1-F3) found in the range of 91.33 ± 3.87% to 94.29 ± 0.26, for Eudragit L100 (F4-F6) found in the range 83.08 ± 0.535% to 93.64 ± 0.666% and for Eudragit S100 and L100 combination (F7-F9) found in the range 84.59 ± 0.529% to 94.13 ± 0.69%. The data revealed that entrapment efficiency was highly influenced by the type of polymer, solvent used to dissolve the drug and polymer, polymer concentration, and method use to prepare the microspheres[3,7-10].

***In vitro* release study**

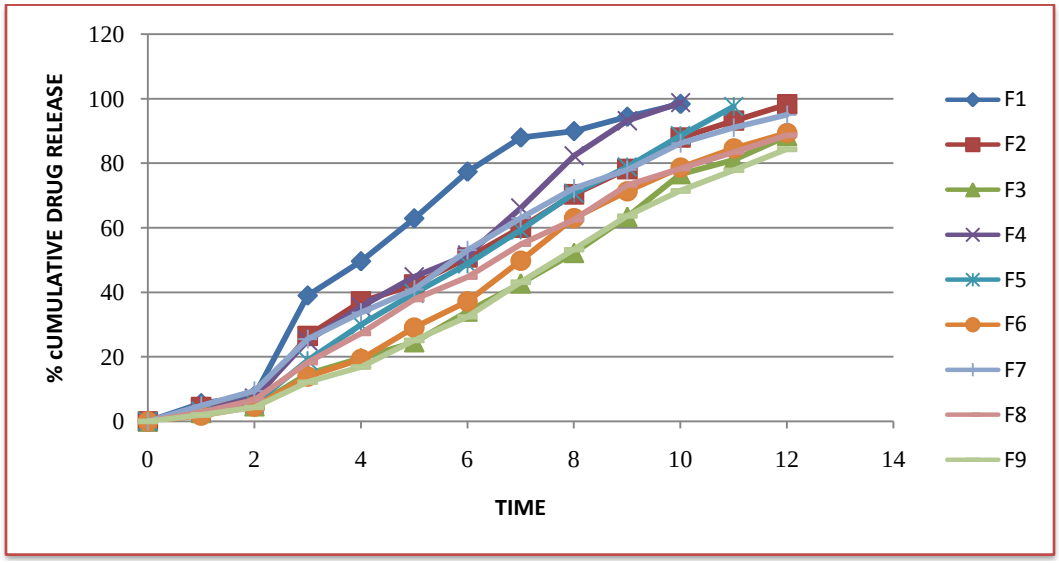

Fig. 5. Cumulative Drug release versus time profile

In vitro release studies of the formulation of Eudragit were carried out in the pH 1.2 (0.1 N HCl) at 37 ± 0.5°C for 2 h followed by in phosphate buffer (pH 7.2) at 37 ± 0.5°C for 10 h. As shown in figure(5), the initial release of Rabeprazole Sodium from all the formulation might have resulted from the dissolution of the drug presented on the surface of the microspheres[10].

The formulations of Eudragit S100, F1 showed the complete drug release after 10 h. Formulation F2 showed the complete release in 12 h while formulation F3 failed to release completely in 12 h. The formulations of Eudragit L100, F4 and F5 showed complete drug release after 10 h and 11 h respectively. Formulation F6 failed to release completely in 12 h. The formulations of Eudragit S100 and L100 combination, F7 showed almost complete release in 12 h while formulation F8 and F9 failed to release completely in 12 h. Though formulation F2 and F7 showed complete and sustained release in 12 h. F2 was considered as the optimized formulation for Eudragit S100 and Eudragit L100 polymer because of higher entrapment.

ISSN: 0975-8585

Release kinetics

Table 3.Kinetic Release study profile

Formulations Code	For Higuchi Equation	For Peppas equation	For Zero Order
	R^2	n	R^2
F1	0.847	1.368	0.946
F2	0.838	1.321	0.987
F3	0.731	1.493	0.955
F4	0.774	1.535	0.973
F5	0.779	1.517	0.978
F6	0.748	1.641	0.961
F7	0.848	1.238	0.988
F8	0.814	1.429	0.986
F9	0.732	1.576	0.956

The release kinetics of all the formulations were checked by fitting the release data to various kinetic models, and the release was best fitted to the Higuchi model. It was further confirmed by fitting the data to the Korsmeyer-Peppas equation and the n value for all the formulations obtained between 1.238 to 1.641, and this revealed that the release followed the zero order mechanism. The R^2 values for all the models are shown in table 3.

FTIR spectroscopy

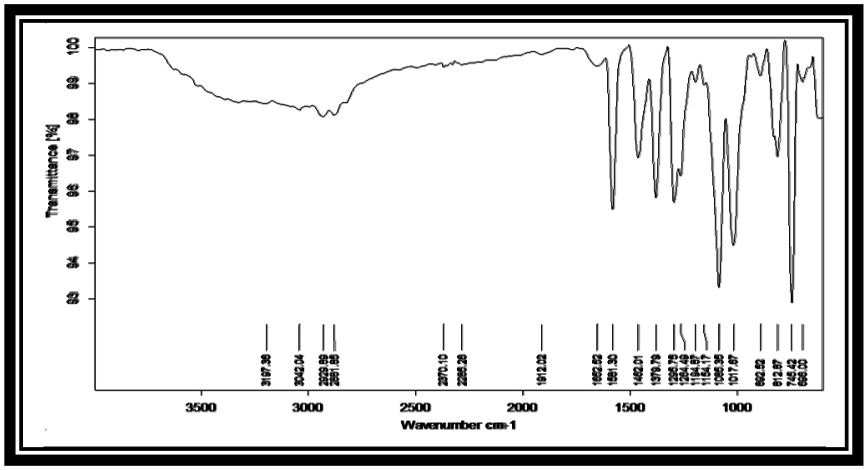

Fig.2. FTIR spectra of pure Rabeprazole Sodium

Drug polymer interaction was checked by the IR spectrum of the optimized formulation with the IR spectrum of pure drug. The IR spectrum of pure drug shows the characteristic peaks at 1017 cm^{-1} for C-O-C band, 3042 cm^{-1} for –C-H aromatic stretching, 2929 cm^{-1} and 2881 cm^{-1} for C-H aliphatic stretching, 1462 cm^{-1} for C-N stretching as shown in fig. 2 and 3.

ISSN: 0975-8585

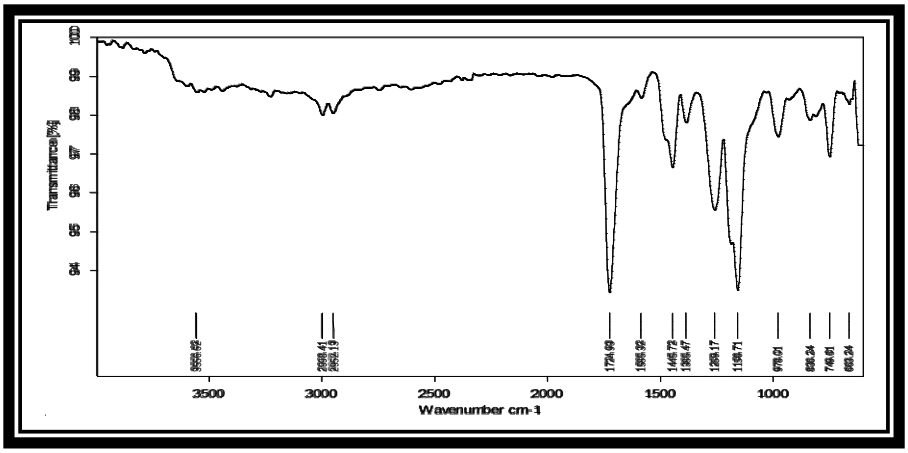

Fig. 3. FTIR spectra of formulation F2

SEM

ISSN: 0975-8585

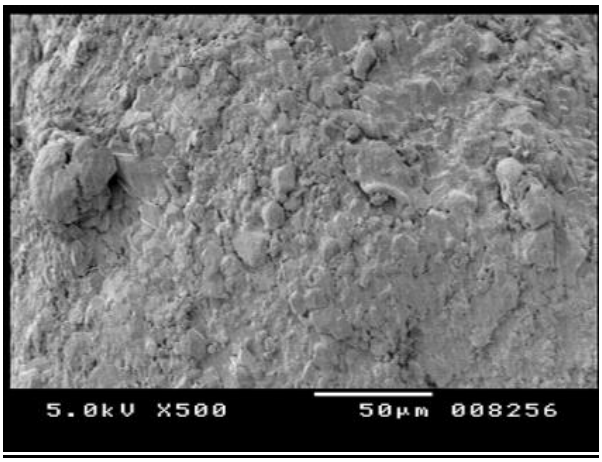

Fig. 4. Scanning electron microscopy of F2

Scanning electron microscopy of the formulation F2 were carried out. For the formulation F2 Figure 4, it showed the spherical shape of the microspheres with a rough surface. The rough surface was due to the presence of the drug crystals on the surface [11].

CONCLUSION

Rabeprazole Sodium were prepared easily and successfully using the solvent evaporation method. The yield and entrapment efficiency was high for all the formulation prepared. Particle size, entrapment efficiency and production yield were highly influenced by the type of polymer and polymer concentration. *In vitro* dissolution of optimized formulations F2 of Eudragit S100 in PBS (pH 7.2) has the potential to target Rabeprazole Sodium in the intestine. According to the results of FTIR, no drug interaction occurred with polymer and Rabeprazole Sodium.

ACKNOWLEDGMENTS

The author thanks Enal Drug Pvt Ltd, for providing gift samples. The authors are also thankful to Chairman, Principal and Staff members of Acharya and B.M. Reddy College of Pharmacy, Bangalore, for providing required facilities to carry out this research work.

REFERENCE

[1] Indian Pharmacopoeia. The Indian Pharmacopoeia commission; Ghaziabad. 2010; Vol 3: p. 2037-38.
[2] Anastacio HM, Hector AT, ImogeneG,Thomas HJ.Clin.Ther1999;21(4):691-701.
[3] Haznedar S, Dortunc B. Int J Pharm 2004; 269: 131-140.
[4] Behera BC, Sahoo SK, Dhal S, Barik BB, B.K. Gupta. Trop J Pharm 2008;7(1):879-885.
[5] Manjunatha N, Vasant S, Rajesh N, Uma N. Int J Pharm Tech Res 2010;2(1):856-862.

[6] Polk A,Amsden K, Yao D,Peng T, Goosen MF. JPharmSci1998; 83(2): 178–185.
[7] Said M, Baomi, Al-badar AA. Fluorouracil.In: Florey K. Analytical Profile of Drug Substances. Elsevier Publication; New Delhi, 2005; 18: 599–632.
[8] Bhalerao SS, Lalla JK, Rane MS. J Microencapsul 2001; 18(3): 299–307.
[9] Sengel CT, Hascicek C, Gonul N. J. Microencapsul2001; 23(2): 135–152.
[10] Rahman Z, Kohli K, Khar RK, Ali M, Charoo NA,Shamsher AAA. AAPSPharmaSciTech2006;7(2)