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DESIGN AND EVALUATION OF MICROSPHERES LOADED WITH ESOMEPRAZOLE

A. Shylaja Rani[1] and D.V. R. N. Bhikshapathi[2*]

[1]Research Scholar, Mewar University, Chittorgarh, Rajasthan, India

[2]Research Supervisor, Mewar University, Chittorgarh, Rajasthan, India

Abstract:

The current investigation objective was to fabricate Esomeprazole loaded microspheres for the treatment of gastritis. The microspheres were prepared by ionotropic gelation technique using sodium as alginate polymer and calcium chloride as cross-linking agent. The effect of polymer and cross-linking agent on particle size, shape, % yield, entrapment efficiency, and drug release were studied. The prepared microspheres morphology and physicochemical properties of were investigated by Fourier-transform infrared spectroscopy (FTIR) and scanning electron microscopy (SEM). Among the total 14 formulations, S12 formulation was optimized at 1.8% of sodium alginate, 10% of calcium chloride maintained 100rpm for 10min at room temperature. The optimized formulation (S12) In vitro drug release studies were conducted up to 12h in 0.1N HCl showing 95.3% drug release which followed the zero order and Higuchi model (R^2 = 0.994, 0.944) respectively, indicating the possible drug release mechanism to be by diffusion. The optimized S12 formulation subjected to stability studies for 6months as per ICH guidelines. Concluded that before and after stability studies no appreciable difference was observed hence the S12 formulation found stable. The data obtained thus suggest that a microparticulate system can be successfully designed for sustained delivery of Esomeprazole and to improve its bioavailability.

Keywords: *Microspheres, Esomeprazole, release order kinetics, sodium alginate.*

Corresponding Author:

D.V. R. N. Bhikshapathi,

Mewar University,

Chittorgarh, Rajasthan, India

Mobile No: +91-9848514228

E-mail : dbpathi@yahoo.com

QR code

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INTRODUCTION:

Gastritis is a chronic inflammatory disease of the stomach mucosa. Nowadays, the prevalence and incidence of gastritis is increasing, especially in developed countries [1]. There is need to develop new drugs and novel formulations as alternative to existing formulations. But bringing an innovative drug molecule from discovery to commercialization is a tough, lengthy and costly process. Hence, the formulation scientist has been carrying out the research on improving efficacy and reducing toxicity of old drugs by the development of new drug delivery techniques such as microparticulates [2]. The major conventional therapeutic strategies are to reduce inflammatory episodes but dosing frequency is high. Hence, the rapid and extensive drug absorption is required in the upper gastrointestinal tract (GIT), which resulted in better therapeutic effect and the lower side effects had improved to its application [3].

Microspheres are defined as monolithic solid spherical particles in the size range of about 1-1000µm. Microspheres are potential drug carriers for oral controlled release. Microspheres had significant importance in biomedical applications [4]. Microspheres can be produced by using sodium alginate as polymer and calcium chloride as cross linking agent. Drug administration in the form of microspheres usually enhances the treatment by providing the drug substance at the site of action and by sustaining drug release finally reduces gastric irritation [5]. There are several encapsulation methods; among those ionic gelation is an interesting method, given its simplicity and versatility [6].

Esomeprazole magnesium trihydrate is a classical example of proton pump inhibitor used for the treatment of gastroesophageal reflux disease, duodenal ulcers caused by H. pylori, erosive esophagitis, nonerosive reflux disease (heartburn and regurgitation), gastrointestinal ulcers associated with Crohn's disease, and for prevention of gastric ulcers in patients on chronic NSAID therapy [7].

The drug candidate associated with low bioavailability (50%) and quick half life of 1-1.5 h, hence is rapidly metabolized into its inactive metabolite within liver and colonic environment so the efficacy would be reduced and requires multiple dosing for maintaining therapeutic effect throughout the day. One approach to avoid this problem would be control the drug release hence increases the bioavailability at insitu level [8].

Polymeric drug delivery system display several advantages over the conventional dosage forms and it includes enhanced efficacy, patient compliance, reduced toxicity, and also to control the encapsulated drug release [9]. Sodium alginate is anionic natural polysaccharide, prepared by mixture of D-mannuronic acid and L-glucuronic acid. Sodium alginate is extensively used as carrier for drug delivery due to its biocompatibility and low toxicity [10]. The widely used method for microspheres preparation is an ionotropic gelation method. This technique offers several advantages such as simple method of preparation no need to use of organic solvent, and, also easier to control. Sodium alginate could form gel in the presence of multivalent cations such as Ca^{2+}, Zn^{2+}, Ba^{2+} and Al^{3+} etc... by ionic cross-linking to form microspheres, it has been widely used in sustained drug release. Hence in this study calcium chloride is selected as cross linking agent and also because of its nontoxic and biocompatibility [11].

The aim of the present study is to develop Esomeprazole loaded microspheres by ionotropic gelation method in order to obtain an extended retention in the upper GIT, which may result in increased absorption and thereby improved bioavailability. The prepared microspheres were evaluated for particle size, shape, % yield, incorporation efficiency, and *in vitro* release study.

MATERIALS AND METHODS:

Materials:

Esomeprazole was obtained as a gift sample from Dr. Reddy's laboratories, Hyd, India. Sodium alginate was purchased from Pruthvi Chemicals; Mumbai, India, calcium chloride was obtained from SD Fine ltd, Mumbai., India. Remaining all chemicals used in this research study was of analytical grade.

Methods:

Preparation of Esomeprazole microspheres

Microspheres were prepared by the ionotropic gelation technique using various percentages of sodium alginate was ranges from 1% to 2.2% w/v and calcium chloride at 7% and 10% as mentioned in Table 1. Initially, sodium alginate solution (100ml) was prepared in to that dissolved the weighed quantity of Esomeprazole (400mg) at room temperature. The above dispersion was sonicated for 30min to eliminate air bubbles that may have been formed during the stirring process. The above dispersions (100ml) was added drop wise via a 20-gauge needle fitted with a 10ml syringe into 100ml of 7% w/v and 10% w/v of calcium chloride solution, being stirred at 500rpm for 10min. The formed microspheres were collected by filtration, washed repeatedly with distilled water, and dried at 60°C for 2h in a hot air oven [12].

Table 1: Formulation trials for Esomeprazole normal microspheres

FORMULATION CODE	ESOMEPRAZOLE (mg)	SODIUM ALGINATE (%)	CALCIUM CHLORIDE (%)
S1	400	1	7
S2	400	1.2	7
S3	400	1.4	7
S4	400	1.6	7
S5	400	1.8	7
S6	400	2	7
S7	400	2.2	7
S8	400	1	10
S9	400	1.2	10
S10	400	1.4	10
S11	400	1.6	10
S12	400	1.8	10
S13	400	2	10
S14	400	2.2	10

Evaluation of Esomeprazole microspheres:

Size analysis [13]

Microsphere Size plays significant role in determining the drug release from it. Particle size analysis was made by optical microscopy technique, using calibrated eye piece and a stage micrometer, almost 100 particles were measured and the results mentioned in the Table 2.

Flow properties [14]

Flow properties of the prepared microspheres were determined in terms of Angle of repose, Bulk density, Tapped density, Compressibility index and Hausner's ratio by using following equations

1.Angle of repose (θ) = $\tan^{-1}$ (h/r)

θ = Angle of repose, h = height of the microsphere pile and d = diameter of the microsphere pile.

$$\text{2.Bulk density} = \frac{\text{Wt of powder}}{\text{Bulk volume of powder}}$$

$$\text{3.Tapped density} = \frac{\text{Wt of microspheres}}{\text{Tapped volume of microspheres}}$$

4. Carr's compressibility index = Tapped density - Bulk density/ Tapped density X 100

$$\text{5.Hausner's ratio} = \frac{\text{Tapped density}}{\text{Bulk density}}$$

Swelling index studies:

The capacity of the microspheres to absorb water and swell was determined in terms of swelling index. For determining swelling index, the microspheres were weighed initially then suspended in pH 1.2. After 1h microspheres were transferred onto blotting paper to remove the excess moisture then weighed the swollen microspheres using a microbalance. After that swollen microspheres were dried in oven at 60°C for 5h until showed the constant weight. The difference in weight of microspheres was used to calculate the swelling index [15].

Swelling index= (Mass of swollen microspheres - Mass of dry microspheres/mass of dried microspheres) 100.

Drug incorporation efficiency and %yield:

To determine the %E E, microspheres (10mg) were weighed, carefully crushed, triturated and suspended in a required quantity of methanol for dissolving microspheres shell coat. The suspension was suitably diluted with water and filtered to separate shell fragments. The drug content was analyzed after suitable dilution spectrophotometrically at 284nm [16]. The amount of drug incorporated in microspheres was calculated by the following formula

% Drug entrapment = Calculated drug concentration /Theoretical drug concentration x 100

And % yield is calculated by the following formula

% yield = [Total weight of microspheres / Total weight of drug and polymer] x 100

***In vitro* drug release studies:**
In the current study, drug release from microspheres was studied using USP Type2 (paddle) dissolution apparatus at 100rpm in 0.1N HCl (pH 1.2) as dissolution fluids (900ml) maintained at 37 ± 0.5°C. The samples were withdrawn at predetermined time intervals such as 1, 2, 4, 6, 8, 10 and 12h simultaneously same volume replenished each time to maintain the sink condition.
The samples were analyzed spectrophotometrically at 284nm for the estimation of Esomeprazole concentrations in the test samples [17]. All experiments were conducted in triplicate.

Kinetic modeling of drug release:
The optimized formulation was treated with the different release kinetic equations include Zero order, First order, Higuchi's model and Korsmeyer-Peppas. Analysis of drug release from microspheres was determined by calculating the (r^2) correlation coefficient.

Drug excipient compatibility studies:
Drug-excipient compatibility was studied by Fourier transmission infrared spectroscopy (FTIR) and Scanning electron microscopy (SEM).

Fourier Transform Infrared Spectroscopy (FTIR):
The FTIR technique can be used to recognize the functional groups in the sample and drug-excipient compatibility. FTIR spectra of pure Esomeprazole, physical mixtures and optimized formulation were recorded by using FTIR (SHIMADZU). Weighed quantity of KBr and excipients were taken in the ratio 100: 1 and mixed by mortar. The samples were made into pellet/disk by the application of pressure [18]. Then the FTIR spectra were recorded between 400 to 4000 cm^{-1}.

SEM studies:
Surface nature of microspheres includes size and shape was examined with the help of Scanning Electron Microscope (HITACHI, S-3700N). The microspheres were dried completely prior to analysis and SEM was carried out at different magnifications of 15.0 kv × 7.1mm, 15 kv x 6.7mm, 15Kv x 6.9mm [19].

Stability studies:
Stability studies were conducted at 40°C ± 2°C/75% RH ± 5% RH for 6months using stability chamber (Thermo Lab, Mumbai). Samples were withdrawn at predetermined intervals of 0, 30, 60, 120, and 180 days period according to ICH guidelines. Various *in vitro* parameters like % yield, entrapment efficiency and *in vitro* release studies were evaluated [20]^0.

RESULTS AND DISCUSSIONS:

Micromeretic properties of Esomeprazole microspheres:

Fig. 1: **Esomeprazole microspheres**

Normal microspheres of Esomeprazole were formulated by ionotropic gelation method, using different polymers like sodium alginate, calcium chloride in different concentrations and the formulation codes S1 to S14 were prepared and shown in Figure 1. All the formulations were evaluated for their various physical parameters.

Particle size was measured by using optical microscopy. All the formulations S1 to S14 varied from 62.12±0.08µm to 96.13±0.09µm. The formulation 12 shows the particle size 88.29±0.13µm.

The bulk density and tapped density of all the formulations S1 to S14 were measured and they are ranged from 0.62g/cc³ to 0.86g/cc³ and 0.59g/cc³ to 0.782g/cc³. The compressibility index values were found to be in the range of 9 to14.0%. These findings indicated that the all batches of formulation exhibited good flow properties. Angle of repose of all the formulations was found satisfactory. The formulation 12 was found to be 25˚.67 having good flow property. The swelling index of all the formulations was within the limits, the swelling index of the formulation 12 was found to be 93%.

Percentage yield and Entrapment efficiency:

Table 2: Percentage yield, entrapment efficiency of Esomeprazole Normal microsphere

Formulation Code	Percentage yield (%)	Entrapment efficiency (%)
S1	65	73
S2	71	72
S3	73	80
S4	82.7	83.3
S5	86.3	93.2
S6	93	93.3
S7	91	94.1
S8	77	64.3
S9	63	72
S10	76	83
S11	85	85
S12	95.5	96.0
S13	75.3	89.9
S14	85.3	84.8

The % Percentage yield and EE of all formulations varies from 65.0% to 95.5% and 72.00% to 96.0%, respectively, as shown in Table 2 It was resulted that % yield increased with an increase the polymer percentage. In some formulations observed the low %yield may be due to leakage of drug from the microspheres during washing process. It was also showed that as polymer % in the formulation increased, the percentage entrapment efficiency also increased this might be due to an increase in the entrapment of drug in the swollen structure of sodium alginate.

***In vitro* drug release studies**

In vitro release studies were conducted and the results are shown in Table 3 and 4 and in Fig. 2 and 3. The Cumulative release of esomeprazole significantly decreased with increasing polymer concentration. The optimized formulation S7 showed the drug release 97.17±0.28% within 12h whereas drug release from marketed product was 95.23±0.21% within 1h.

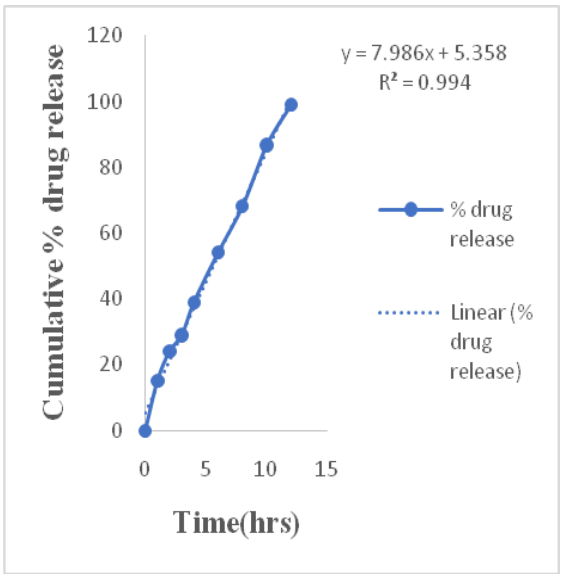
Table 3: *In-vitro* cumulative %drug release of Esomeprazole Normal microspheres formulations S1-S7

Time (h)	S1	S2	S3	S4	S5	S6	S7
0	0±0	0±0	0±0	0±0	0±0	0±0	0±0
1	21.04±0.12	22.41±0.14	15.11±0.14	20.41±0.32	22.16±0.11	16.7±0.13	21.3±0.22
2	26.11±0.23	25.41±1.11	23.15±0.88	24.81±0.11	25.41±0.32	23.1±0.33	25.4±0.32
3	28.32±1.15	30.11±1.13	30.19±1.31	31.31±1.14	30.11±1.16	31.6±0.12	28.3±0.44
4	39.39±2.32	38.21±1.85	38.91±1.83	44.14±2.22	38.21±2.15	38.9±0.11	39.9±0.23
6	50.39±2.82	51.34±2.16	49.97±2.78	51.71±2.82	51.31±2.91	49.9±0.32	51.39±0.33
8	66.23±3.43	63.31±2.75	61.24±2.99	60.32±3.21	63.31±3.22	61.2±0.21	65.23±0.42
10	70.12±3.87	69.97±3.32	70.1±3.76	70.71±3.32	69.19±3.83	70.1±0.33	73.12±0.21
12	83.34±4.12	82.31±4.11	80.2±4.14	83.51±4.22	82.31±0.15	81.2±0.23	82.34±0.24

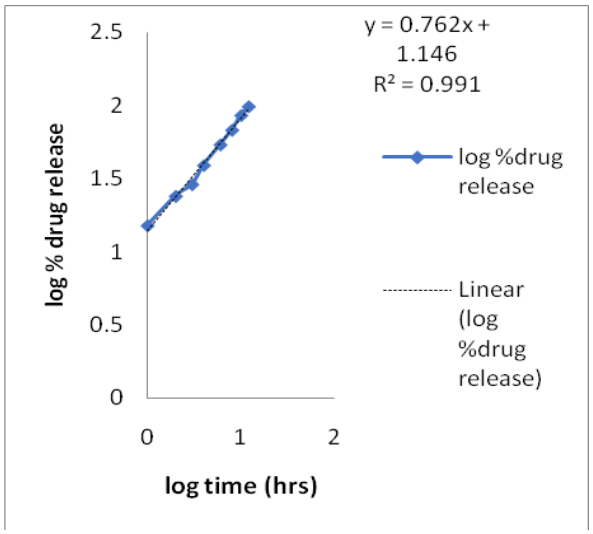
Table 4: In-vitro cumulative %drug release of Esomeprazole Normal microspheres formulations S8-S14

Time (h)	S8	S9	S10	S11	S12	S13	S14	Marketed product
0	0±0	0±0	0±0	0±0	0±0	0±0	0±0	0±0
1	22±0.22	15±0.11	20±0.21	22±0.11	15.2±0.16	20.32±0.13	21.63±0.12	95.23±0.21%
2	25.4±0.32	23±0.32	24.8±0.33	25.4±0.13	24.2±0.15	31.15±0.22	32.01±0.11	
3	30±0.16	30±0.33	31.3±0.41	30±0.21	31.9±0.22	42.19±0.13	37.11±0.23	
4	38.2±0.33	38.9±0.32	44.4±0.11	38.2±0.22	40.1±0.14	49.23±.96	44.24±0.41	
6	51.3±0.21	49.9±0.42	51.7±0.32	51.3±0.44	54.2±0.15	56.73±0.13	57.83±0.62	
8	63.3±0.32	61.2±0.22	60.3±0.18	63.3±0.32	68.2±0.13	65.46±0.13	64.03±0.14	
10	69.9±0.26	70.1±0.13	70.7±0.22	69.9±0.22	86.8±0.11	69.46±0.21	75.24±0.21	
12	82.3±0.22	80.2±0.22	83.5±0.34	82.3±0.14	95.3±0.15	79.29±0.15	85.36±0.44	

Release order kinetics:



ZERO ORDER



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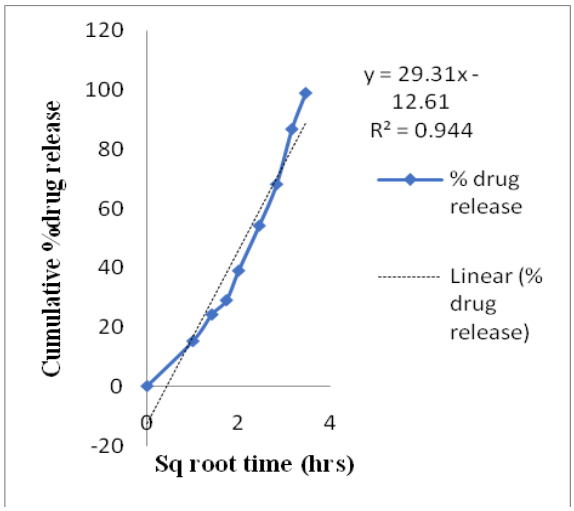

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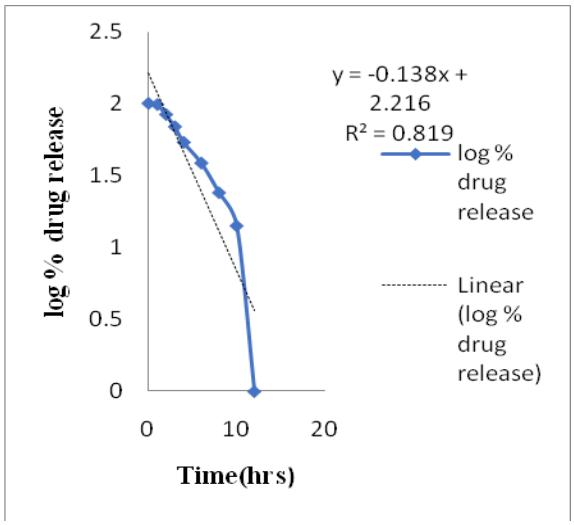
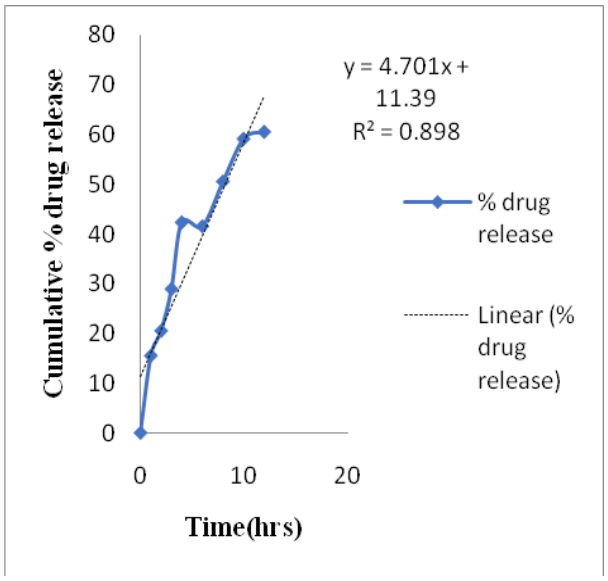


KORSMEYER-PEPPAS

Fig 2: Mathematical modeling of Esomeprazole optimized microspheres (S12)



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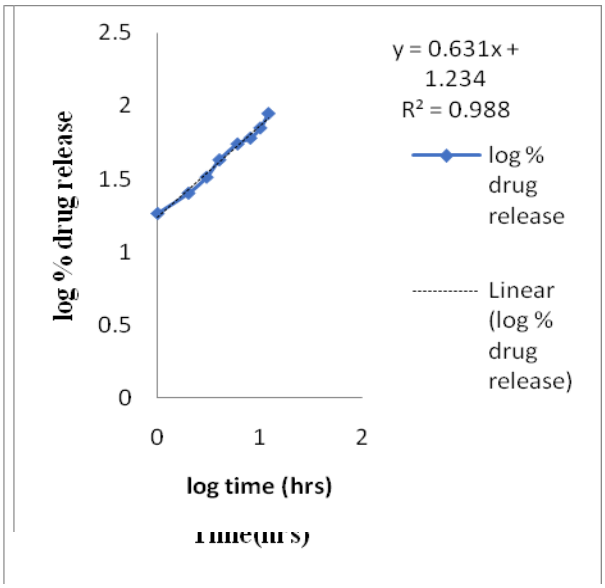

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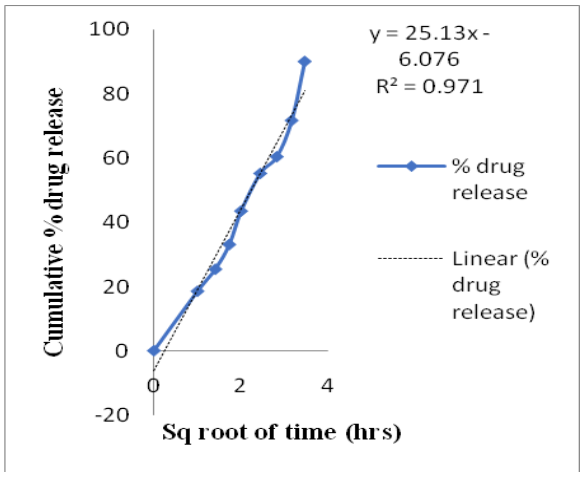

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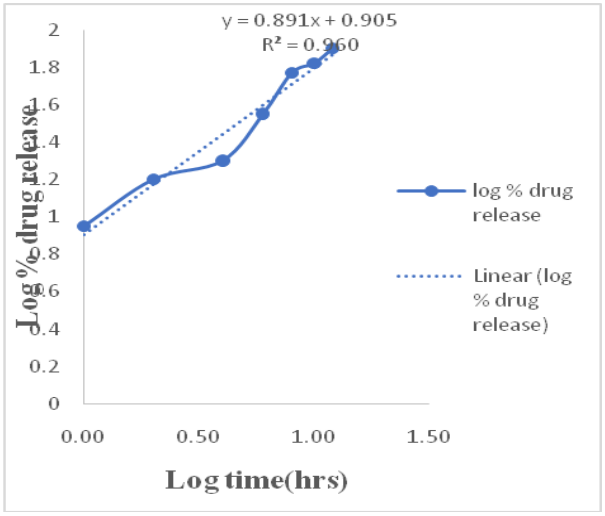

KORSMEYER-PEPPAS

Fig 3: Mathematical modeling of marketed product

The *in vitro* release profiles from optimized formulation and reference standard (Figure 2 & 3 respectively) were applied on various kinetic models. The best fit with the highest correlation coefficient was observed in the Korsmeyer-peppas model and zero-order release kinetics followed by Higuchi model. The '*n*' value of formulation was found to be 0.762 indicating that the drug release was followed by anomalous (non-fickian) diffusion.

Drug excipient compatibility studies

Fourier Transform Infrared Spectroscopy (FTIR)

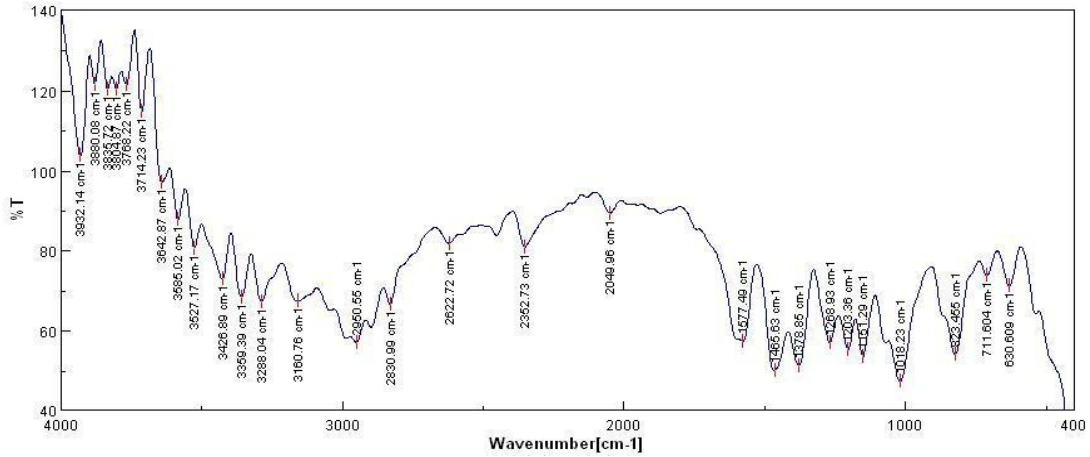

Fig 4: FTIR spectrum of pure drug Esomeprazole

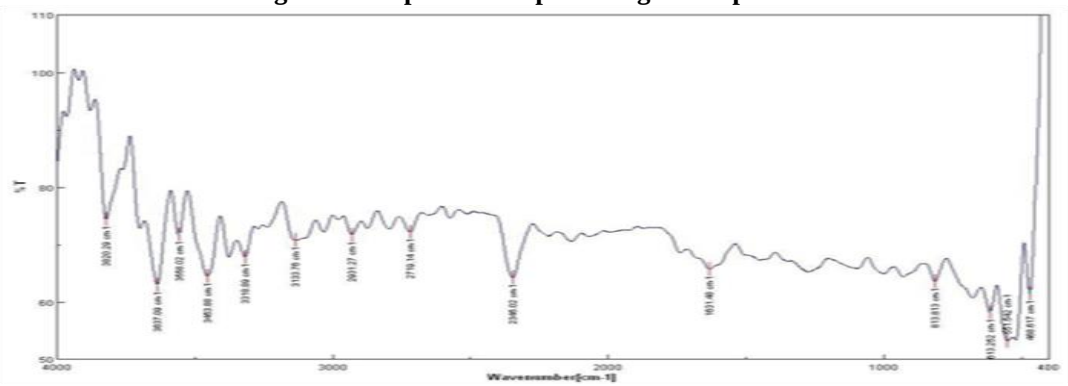

Fig 5: FTIR spectrum of Nizatidine optimized (S12) of microspheres

There were no new significant bands observed in the pure drug (Figure 4) and optimized formulation (Figure 5), which confirms that no interaction takes place between the drug and excipients.

SEM of Esomeprazole microspheres

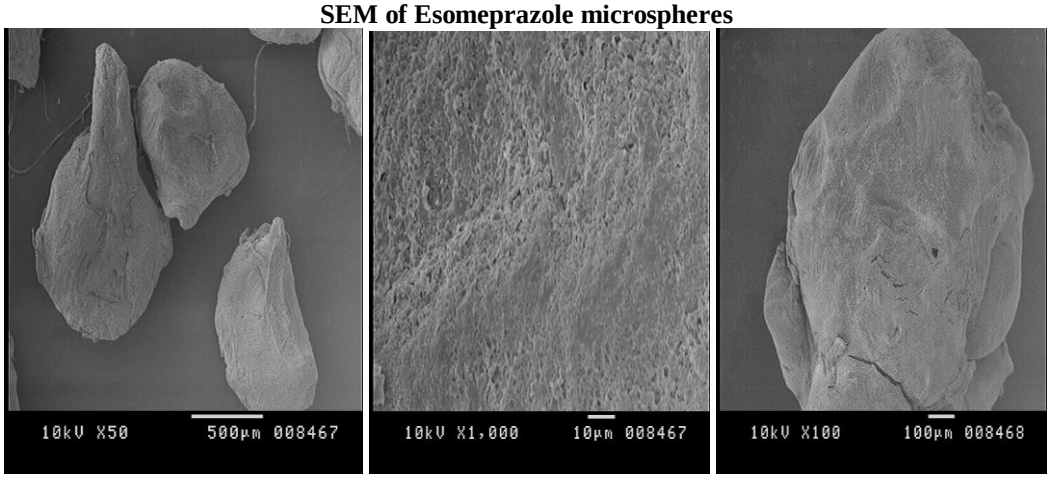

Fig 6: Scanning electron micrographs of Esomeprazole microspheres

The SEM photomicrographs of the dried microspheres were shown in Figure 6. It was observed that shape of microspheres seems to be spherical with fairly rough outer surface. The surface was rough due to polymer matrix density which justifies the controlled release of the drug.

Table 5: Staility studies of optimized esomeprazole normal microspheres

Retest time for optimized formulation (S12)	Percentage yield	Entrapment efficiency	*In-vitro* drug release profile (%)
0 days	95.50	96.00	95.30
30 days	94.21	95.68	95.18
60 days	94.02	95.34	94.03
120 days	93.94	94.85	93.82
180 days	93.22	94.42	93.21

Stability studies:
The results of stability studies was mentioned in Table 5 and indicated that there was no significant change in results before and after stability studies hence the optimized formulation S12 found to be stable.

CONCLUSION:

In this study, stable sustained release Esomeprazole microspheres were prepared successfully by the ionotropic gelation method using sodium alginate (polymer) and calcium chloride (cross linking agent). The Cumulative % drug release of optimized formulation (S12) was found to be slow, controlled release over a period of 12h when compared to marketed product. The drug release followed the zero order and Higuchi model indicated the release was controlled by diffusion. Accelerated stability studies confirmed that the microspheres formed were stable. The results of the present study indicated promising potential of microspheres in the delivery of drugs havin low biological half life and bioavailability with controlled release of Esomeprazole in the management of gastritis.

REFERENCES:

1.Sanjay KM, Shruti C, Sushma T. Chitosan–sodium alginate nanoparticles as submicroscopic reservoirs for ocular delivery: Formulation, optimization and in vitro characterization. Eur J Pharm Biopharm 2008; 68: 513–525.

2.Jiao Y, Pang X, Liu M, Zhang B, Li L, Zhai G. Recent progresses in bioadhesive microspheres via transmucosal administration. Colloids Surf B Biointerfaces 2016; 140:361-72.

3.Hemlata P, Roshan VT, Michael AR. Recent Advancements in Mucoadhesive Floating Drug Delivery Systems: A Mini-Review. J Drug Deliv Sci Tec 2016; 31: 65-71.

4.Junzi Wu, Gareth RW, Christopher BW. Liraglutide-loaded poly (lactic-co-glycolic acid) microspheres: Preparation and in vivo evaluation. Eur J Pharm Sci 2016; 92: 28–38.

5.Wenfang G, Simon T, Martina HS. Synthesis of pH-responsive and thiol-degradable hollow microspheres. Polymer 2013; 54: 1010 – 1017.

6.Bhardwaj P, Singh R, Swarup A. Development and characterization of newer floating film bearing 5-fluorouracil as a model drug. J Drug Del Sci Tech 2014; 24: 486-490.

7.Johnson DA. Review of esomeprazole in the treatment of acid disorders. Expert Opin Pharmacotherapy 2003;4:253-64,

8.Vergin H, Mascher H, Strobel K, Nitsche V. Pharmacokinetics and bioequivalence of different formulations of pirenzepine. Arzneimittelforschung. 1986;36(9):1409-12.

9.Saisivam S, Rahamath UM, Shakeel F**.** Development of floating matrix tablets of losartan potassium: in vitro and in vivo evaluation. J Drug Del Sci Tech 2013; 23: 611-617.

10.Harri H, Peter D, Pitkänen P, Markku H. Effect of concentration and temperature on the properties of the microspheres prepared using an emulsion–solvent extraction process. Adv Powder Technol 2012; 23: 779–786.

11.Megha S, Seema K, Agnimitra D. In-vitro and in-vivo evaluation of repaglinide loaded floating microspheres prepared from different viscosity grades of HPMC polymer. Saudi Pharma J 2015; 23: 675–682.

12.Rishikesh G, Sunil Kumar P, Snigdha P, Bhardwaj P. Formulation and evaluation of novel stomach specific floating microspheres bearing famotidine for treatment of gastric ulcer and their radiographic study. Asian Pac J Trop Biomed 2014; 4: 729-735.

13.Yasunori S, Yoshiaki K, Hirofumi T, Hiromitsu Y. *In vitro* evaluation of floating and drug releasing behaviors of hollow microspheres (microballoons) prepared by the emulsion solvent diffusion method. Eur J Pharm Biopharm 2004; 57: 235–243.

14.Pandya N, Pandya M, Bhaskar VH. Preparation and *in vitro* Characterization of Porous Carrier–Based Glipizide Floating Microspheres for Gastric Delivery. J Young Pharm 2011; 3: 97–104.

15.Kampanart H, Katsuhide T, Sriamornsak P, Nunthanid J. Advanced technologies for assessment of polymer swelling and erosion behaviors in pharmaceutical aspect. Eur J Pharm Biopharm 2013; 83: 315–321.

16.Saravanan M, Anupama B. Development and evaluation of ethylcellulose floating microspheres loaded with ranitidine hydrochloride by novel solvent

evaporation-matrix erosion method. Carbohydr Polym 2011; 85: 592–598.
17.Ninan M, Lu X, Qifang W, Xiangrong Z. Development and evaluation of new sustained-release floating microspheres. Int J Pharm 2008; 358: 82–90.
18.Antonio JR, Ronald JN, Philippe A. Microencapsulation of lipophilic drugs in chitosan-coated alginate microspheres. Int J Pharm 1999;187: 115–123.
19.Sunil K. Jain, Govind. P, Agrawal, Narendra K. Jain. A novel calcium silicate based microspheres of repaglinide: In vivo investigations. Journal of Controlled Release 2006; 113: 111–116.
20.Yan Li, Haibing He, Qiao Wang, Xing Tang. Preparation, stability and pharmacokinetics evaluation of lipid microspheres loading a promising antitumor candidate, Timataxel. Asian J Pharm Sci 2016; 11: 771–779.