ISSN 0976 – 3333

Available Online at www.ijpba.info.

International Journal of Pharmaceutical & Biological Archives 2011; 2(3):880-886

ORIGINAL RESEARCH ARTICLE

Formulation, Optimization And *In Vitro* Characterization Of Mucoadhesive Microparticle

Harshad Parmar*, Sunil Bakliwal, Nayan Gujarathi, Bhushan Rane, Sunil Pawar

Department of Quality Assurance, P.S.G.V.P.M's College of Pharmacy, Shahada, Dist.Nandurbar- 425409, Maharashtra, India

Received 12 Mar 2011; Revised 24 May 2011; Accepted 12 Jun 2011

ABSTRACT

Mucoadhesive microparticle of Itopride HCl was design in order to obtain a unique drug delivery system which would remain in the stomach and prolong the residence time at the absorption site by intimate contact with the mucus layer thereby increase bioavailability, reduce the frequency of dose administration and also to prolong the drug release. The mucoadhesive microparticles were prepared by Orifice ionic gelation method using sodium alginate in combination with carbopol 934 and HPMC K15. Entrapment efficiency was in the range of 41.32 to 81.68 %. Microparticle exhibited good mucoadhesive property in the *in vitro* wash off test and revealed that Carbopol 934 had greater mucoadhesive strength than that of HPMC K15. Itopride HCl release from this mucoadhesive microparticle was slow and showed sustained release. SEM study revealed that microparticles were discrete, spherical and free flowing. Stability study of optimized batch was carried out and drug content found was retained with permissible limits and there was no significant difference in the drug content.

Keywords: Itopride HCl, Mucoadhesive microparticles, *in vitro* wash off test, Orifice ionic gelation.

INTRODUCTION

The oral route of drug administration constitutes the most convenient and preferred means of drug delivery to systemic circulation of body. However oral administration of most of the drugs in conventional dosage forms has short-term limitations due to their inability to restrain and localize the system at gastro-intestinal tract. Microparticles constitute an important part of these particulate drug delivery systems by virtue of their small size and efficient carrier capacity. Microparticles are the carrier linked drug delivery system in which particle size ranges from (1-1000 µm) range in diameter having a core of drug and entirely outer layers of polymers as coating material. However, the success of these microspheres is limited due to their short residence time at site of absorption. It would, therefore be advantageous to have means for providing an intimate contact of the drug delivery system with the absorbing membrane[1]. This can be achieved by coupling bioadhesion characteristics to microspheres and developing bioadhesive microspheres. Bioadhesive microspheres have advantages like efficient absorption and enhanced bioavailability of the drugs due to a high surface to volume ratio, a much more intimate contact with the mucus layer and specific targeting of drugs to the absorption site.

Non-ulcer dyspepsia (NUD), gastro-esophageal reflux disease (GERD), gastritis, diabetic gastroparesis and functional dyspepsia are commonly encountered disorders of gastric motility in clinical practice. An acetylcholinesterase inhibitor (often abbreviated AChEI) or anti-cholinesterase represents a novel gastro prokinetc agent that inhibits the enzyme acetylcholine esterase (AChE) responsible for degradation of Acetylcholine. Among them Itopride compose of list of approved Acetylcholine esterase inhibitor is drug of choice for Gastro esophageal reflux disease (GERD) and other disorders of gastric motility.

***Corresponding Author:** Harshad Parmar, **Email:** harshad.parmar19@gmail.com, **Phone No:** +91-9890207404

Itopride HCl has half-life of 5-6 hours and requires frequent administration of dose. Hence it is necessary to develop sustained release formulation to overcome this draw back.

MATERIALS AND METHODS

Itopride HCl was obtained as a gift sample from Optimus Drugs (P) Limited. Carbopol 934 and HPMC K15 were obtained as a gift sample from Corel Pharma Chem, Ahmedabad and Colorcon Asia Pvt. Ltd, Goa, India respectively. Sodium alginate and Calcium chloride were purchased from S.D fine Chemicals, Mumbai.

PREPARATION OF MICROPARTICLE

Microparticles were prepared by orifice ionic gelation method. Sodium alginate and mucoadhesive polymer such as HPMC K15 and Carbopol 934 were dissolved in purified water to form a homogeneous polymer solution. The core material was added to this polymer solution and mixed to form a smooth viscous dispersion. This dispersion was added drop wise into 5% w/v $CaCl_2$ solution through a syringe 22 gauge. The added droplets were retained in the calcium chloride solution for 15 minutes to complete the curing reaction and to produce spherical rigid microparticles. The microparticles were collected and dried in an oven at 45°C for 4 hours. The composition of different formulations is shown in (**Table 1)**.

Table 1. Composition of Alginate reinforced Carbopol 934 mucoadhesive microparticle.

Batch code	Drug: polymer	Drug (mg)	Carbopol 934 (mg)	Sodium Alginate (mg)	Polymer ratio
MC1	1:1	250	125	125	1:1
MC2	1:1.5	250	150	225	1:1.5
MC3	1:2	250	166.66	333.32	1:2
MC4	1:2.5	250	178.57	446.42	1:2.5
MC5	1:3	250	187.5	562.5	1:3

Table 2. Composition of Alginate reinforced HPMC K15 mucoadhesive microparticle.

Batch code	Drug: polymer	Drug (mg)	HPMC K15 (mg)	Sodium Alginate (mg)	Polymer ratio
MH1	1:1	250	125	125	1:1
MH2	1:1.5	250	150	225	1:1.5
MH3	1:2	250	166.66	333.32	1:2
MH4	1:2.5	250	178.57	446.42	1:2.5
MH5	1:3	250	187.5	562.5	1:3

CHARACTERIZATION OF MUCOADHESIVE MICROPARTICLES

FT-IR Spectroscopy:

The interaction between the Itopride and polymers were determined by using the FT-IR spectroscopy wherein infrared spectra of Itopride HCl, Sodium Alginate, Carbopol 934, HPMC K15 and Microparticles were carried out using the KBr disk method. The scanning range was 400 to 4000 cm^{-1} and the resolution was 1/cm.

Production yield:

The product yields of microparticles of various formulations were calculated using the weight of final product after drying with respect to the initial total weight of the drug and polymer used for preparation of microparticles and percent yields were calculated as per the formula mentioned below [2].

$$\text{Product yield} = \frac{\text{Practical mass (microparticles)}}{\text{Theoretical mass (Polymer + Drug)}} \times 100$$

Drug entrapment efficiency:

The drug content of alginate microspheres was determined by crushing 50 mg Microparticles in 100 ml 0.1 N HCl followed by agitation with a magnetic stirrer for 12 hours to dissolve the polymer. The solution was then gently warmed for two hours to extract the drugs completely, filtered, and the resulting solution was analyzed by UV spectrophotometer for itopride at 257 nm[3].

$$\text{DEE (\%)} = \frac{\text{Practical drug content}}{\text{Theoretical drug content}} \times 100$$

Swelling property:

The swelling indices of formulations were determined by immersing preweighed dried microparticles (20 mg) in10 ml of 0.1 N HCl at a temperature of 37º C and flask was shaken at 100 rpm by rotary shaker for 12 hrs. After 12 hours, the sample was removed, blotted with a piece of tissue paper to absorb excess water on surface and then reweighed. The difference in weight before and after soaking was found out. The swelling

index was calculated from the following expression[4].

$$SI = \frac{WS - WD}{WS} \times 100$$

Where WS = weight of the swollen microparticles,

WD = weight of dried microparticles.

Micromeritic Properties [5, 6]:

Density

The bulk density and tapped density of microparticles were measured in 10 ml of graduated cylinder. Weighed quantities of microparticles were introduced into a 10 ml of measuring cylinder. After noting down the initial volume, the sample contained in the measuring was tapped mechanically onto a hard surface from the height of 2.5 cm at 2 second intervals. The tapping was done for 100 times. The initial bulk volume and tapped volume were noted from which, their respective densities were calculated.

The bulk density and tapped density were calculated using following equation

$$\text{Bulk density} = \frac{\text{Weight of microparticles}}{\text{Volume of microparticles}} \times 100$$

$$\text{Tapped density} = \frac{\text{Weight of microparticles}}{\text{Tapped Volume of microparticles}} \times 100$$

Compressibility Index

Compressibility index of all formulations was calculated by following equation

$$\text{Compressibility Index} = \frac{\text{Tapped density} - \text{Bulk density}}{\text{Tapped density}} \times 100$$

Hausner's Ratio

Hausner's ratio was also calculated by using following equation

$$\text{Hausner's Ratio} = \frac{\text{Tapped density}}{\text{Bulk density}}$$

Angle of Repose

The angle of repose was determined by the fixed funnel method. Accurately weighed microparticles were taken in funnel. The height of funnel was adjusted in such a way that the tip of the funnel just touched to apex of the heap of the microparticles. It is the maximum angle possible between the surface of pile of microparticle and horizontal plane. The microparticle was allowed to flow through the funnel freely onto the surface. The diameter of microparticle cone was measured. The Angle of repose is calculated by using the following equation

$$\tan\theta = \frac{h}{r}$$

Where, θ = Angle of repose

h = height of pile

r = radius of pile

Particle size and shape:

The particle size and shape of Microparticles were carried out using optical microscope. The particle diameters of 50 microparticles were measured randomly by optical microscope[7].

Mucoadhesive measurement study:

The mucoadhesive properties of microparticles were evaluated by *in vitro* adhesion testing method, known as wash off method. The pieces of goat intestinal mucosa (2×2 cm) were tied on glass slides (3×1 inch) by rubber band. About 50 microparticles were counted and spread over the wet rinsed tissue specimen and wait for 10 min. and immediately thereafter the support were hung on the arm of a USP tablet disintegrating test machine. By operating the disintegration machine the tissue specimen was given slow regular up and down movement in the 1 L vessel containing 0.1 N HCl at 37± 2°C. At the end of each hour, the machine was stopped and number of microparticles still adhering on the tissue was counted [8, 9].

$$\text{Percentage mucoadhesion} = \frac{Nt - N0}{N0} \times 100$$

Where, Nt = Number of microparticles adhered to tissue after each time interval.

N_0 = Number of microparticles applied

***In vitro* drug release study:**

The dissolution studies of mucoadhesive microparticles were carried out in a USP dissolution apparatus II (TDT 08L Electrolab) at a rotation speed of 100 rpm in a 900 ml medium at 37 ±0.5°C. The microparticles were placed in muslin cloth and tied to the paddle and transferred to dissolution medium 0.1 N HCl and samples were taken at selected time intervals, filtered through Whatmann filter paper no. 41 and analyzed by UV spectrophotometer 2210 (Systronics, Ahmedabad) at 257 nm.

The drug release data of the *in vitro* dissolution study was analyzed with various kinetics equations like zero order, first order, matrix, Peppas and Hixon crowell model. Coefficient of correlation (r) values were calculated for linear curves obtained by regression analysis of the plots.

Scanning Electron Microscopy:

The SEM photographs of microparticles of optimized formulation were obtained by scanning electron microscope using platinum sputter technique. A working distance of 500 µm and the particles were vacuum dried and 5-kV accelerating voltage was set as a processing parameters.

Stability study [10] :

Stability studies were carried out at various temperatures. The samples were wrapped in a butter paper and placed in petri dishes. These were stored at room temperature (27 ± 2° C) and at elevated temperature (45 ± 2° C) for a period of 1 month. Then microparticles were analyzed for physical changes such as color, texture and Entrapment efficiency.

RESULTS AND DISCUSSION

The formulated microparticles were evaluated for production yield, drug entrapment efficiency, swelling index. The results are shown in **(Table 3)**. The result revealed that the production yield of batch MC1 to MC5 was found in the range 74 to 92 % and that of MH1 to MH5 was found in the range of 70 to 89 %.

Drug polymer compatibility studies were carried out using FTIR spectroscopy to establish any possible interaction of Itopride HCl with the polymer used in the formulation. Thus results indicated that the characteristic absorption peak due to pure Itopride HCl have appeared in the formulated microparticle, without any significant change in their position indicating no chemical interaction between Itopride HCl and polymers.

Percentage Drug Entrapment efficiencies (%DEE) of batch MC1- MC5 was found in the range of 54.12 ± 0.48 to 81.68 ± 0.15 % and of batch MH1- MH5 was found in the range of 41.32 ± 0.33 to 78.4 ± 0.33 %. The data is shown in Table 3. It was observed that the drug entrapment efficiency of the prepared microparticles increases progressively with an increase in the proportion of respective polymers. Alginate concentration along with mucoadhesive polymer increases may also reduce loss of drug in curing medium due to formation of dense matrix structure. Increase in polymer proportion increases the viscosity of the dispersed phase. The higher viscosity of the polymer solution at the highest polymer proportion would be expected to decrease the diffusion of the drug into the external phase which would result in higher entrapment efficiency. The higher incorporation efficiency was observed as the proportion of alginate increased. This may be attributed to the greater availability of active calcium binding sites in the polymeric chains and consequently the greater degree of cross linking as the quantity of sodium alginate increased, resulting in the formation of nonporous microspheres.

The degree of swelling is expressed as the percentage of water in hydrogel at any instant during swelling. The swelling property of microparticles was studied in 0.1 N HCl. As the polymer to the drug ratio increases, the degree of swelling increased from 40 to 105 % for microparticles of batch MC1- MC5 and 25 to 95 % for microparticles of batch MH1- MH5. It can be concluded from the data shown in Table 3 that with increase in the polymer ratio, the degree of swelling also enhanced. So it can be stated that amount of polymer directly affects the degree of swelling.

The mean particle size increased with increasing polymer proportion which is due to a significant increase in viscosity, thus leading to an increased droplet size and finally a higher microparticle size. Microparticles of Itopride HCl using sodium alginate in combination with Carbopol 934 exhibited a size range of 780.18 µm to 963.5 µm and microparticles of Itopride HCl using sodium alginate in combination with HPMC K15 exhibit a size range of 765.7 µm to 939.4µm. Increase in the ratio of polymer tends to form the particles more spherical and obtained uniform size spheres. The difference in the shape of microcapsules is observed, representing that microparticle containing higher amount of alginate are more spherical and regular as compared to that of microparticle having lower percent of alginate. Such results may be due to as the polymer (alginate) proportion increases the spherical nature of microparticle also increases.

Optimized batch MC4 showed 50 % of microparticles were still adhering and MH4 showed 46 % of microparticle remains adhere at the end of 8 hour. Mucoadhesive microparticle are anticipated to take up water from the underlying mucosal tissue by absorbing, swelling and capillary effects leading to considerable stronger adhesion. From the technique which was used for mucoadhesive strength determination it was found that Carbopol 934 had greater mucoadhesive strength than that of HPMC K15. Carbopol possess various carboxyl group, when mobile at the wet mucosal surface, they orientate these mucoadhesive site towards mucosa and make interaction through hydrogen bonding. Also greater swelling rate of Carbopol 934 results in large surface of polymer that is expand to the mucosal layer resulting in increase in no. of hydrogen bonding between the polymer and mucosal layer. Thus increase the mucoadhesive

strength of polymer. Graphical representation is shown in (**Figure 1 and 2**).

The formulation coded as MC4 and MC5 containing sodium alginate along with Carbopol 934 sustained release of drug up to 12 hr found to be 98.31 ±0.43 and 88.80 ±0.65 respectively. The formulation MH4 and MH5 containing sodium alginate along with HPMC K15 also showed sustained release of 98.91 ±0.43 and 90.15 ±0.46 was observed with increase in ratio of polymer at the end of 12 hr. It is attributed to fact that alginate hydrated faster under acidic condition and built up the diffusion barrier more rapidly resulting in slower release in the acidic phase. As the polymer to the drug ratio was increased the extent of drug release decreases. A significant decrease in the rate and extent of the drug release is due to the higher density of polymer matrix that results in increased diffusion path length through which the drug molecule have to traverse. The release would depend on diffusion of Itopride through the insoluble matrix of alginate polymer in 0.1 M HCl and a sustained drug release behavior was observed.

The photographs of the optimized batch taken by Scanning electron microscopy are shown in (**Figure 5 and 6**). The SEM Photograph indicated that the microparticles were spherical and completely covered the coat polymer. The results revealed that the microparticles containing higher amount of alginate are more spherical and regular as compared to microparticle containing lower amount of alginate, this explains the dependence of the polymer concentration on the spherical nature of microparticle.

Two formulation coded MC4 and MH4 were chosen for stability studies. The stability of preparation is an important factor to estimate the quality of dosage form. The stability data did not show any significant change in color, texture and Entrapment efficiency. Thus we may conclude that, the drug does not undergo degradation on storage.

Table 3. Production yield, Entrapment efficiency and swelling index of mucoadhesive microparticle.

Batch Code	Product yield (%)	DEE (%)*	Swelling index (%)
MC1	74	54.12 ± 0.48	40
MC2	92	64.10 ± 0.21	55
MC3	90	67.28 ± 0.36	85
MC4	90.28	78.71 ± 0.39	95
MC5	85	81.68 ± 0.15	105
MH1	70	41.32 ± 0.33	25
MH2	83.2	53.1 ± 0.62	45
MH3	86.6	68.4 ± 0.49	70
MH4	78.8	72.1 ± 0.86	90
MH5	89	78.4 ± 0.33	95

Table 4. Micromeritics properties of mucoadhesive microparticle

Batch ratio	Bulk density (g/ml)	Tapped density (g/ml)	Carr`s index (%)	Hausner ratio	Angle of repose(θ)
MC1	0.975	1.170	16.66	1.20	33.69
MC2	0.871	1.016	14.33	1.16	29.74
MC3	1.012	1.157	12.53	1.14	29.05
MC4	0.969	1.145	15.41	1.18	27.14
MC5	1.039	1.228	15.39	1.18	26.00
MH1	1.13	1.36	16.91	1.20	31.21
MH2	0.885	1.03	14.32	1.16	29.05
MH3	0.928	1.08	14.07	1.16	27.75
MH4	1.344	1.512	11.14	1.12	27.14
MH5	0.950	1.055	9.95	1.11	24.94

Fig 1. Percentage mucoadhesion measurement of MC1-MC5

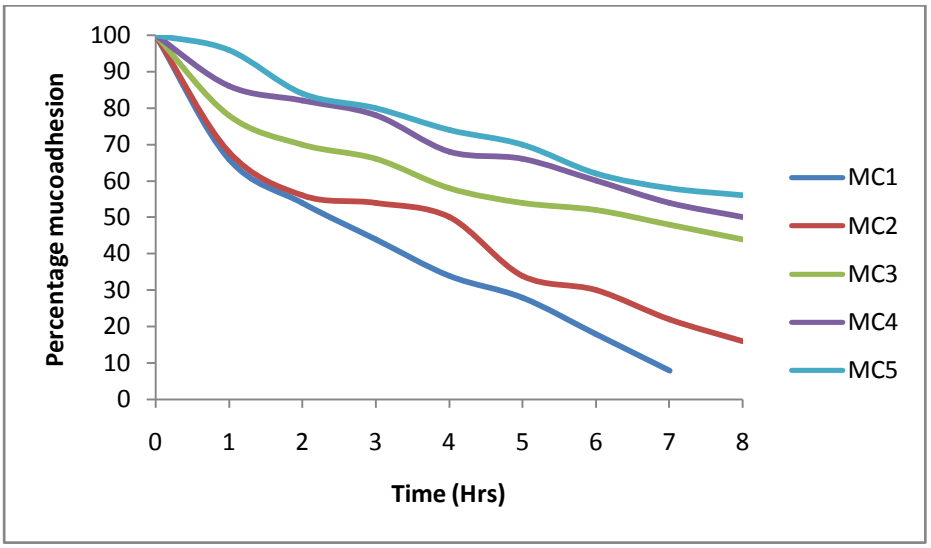


Fig 2. Percentage mucoadhesion measurement of MH1-MH5

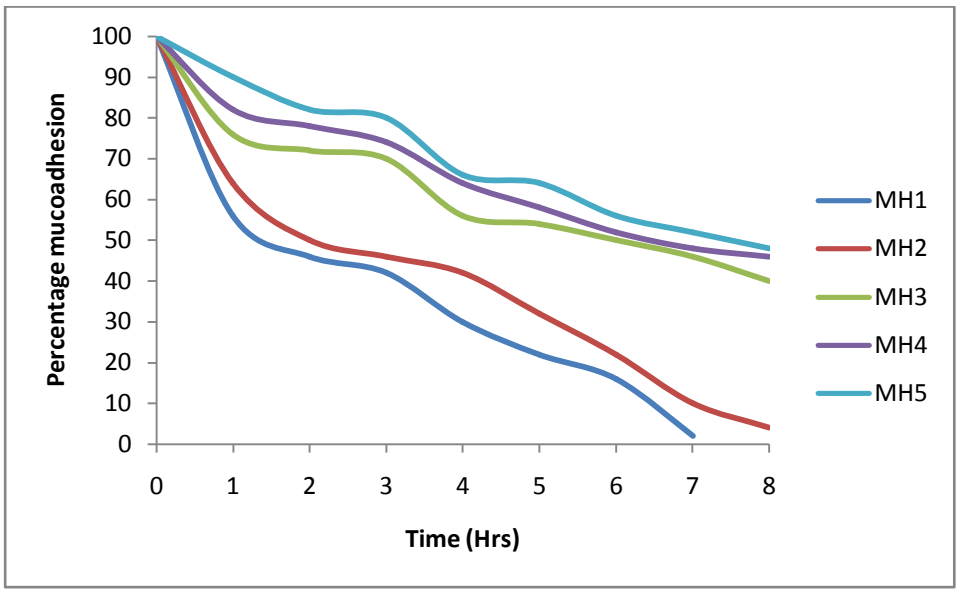


Fig 3. *In vitro* drug release (%) of batch MC1-MC5

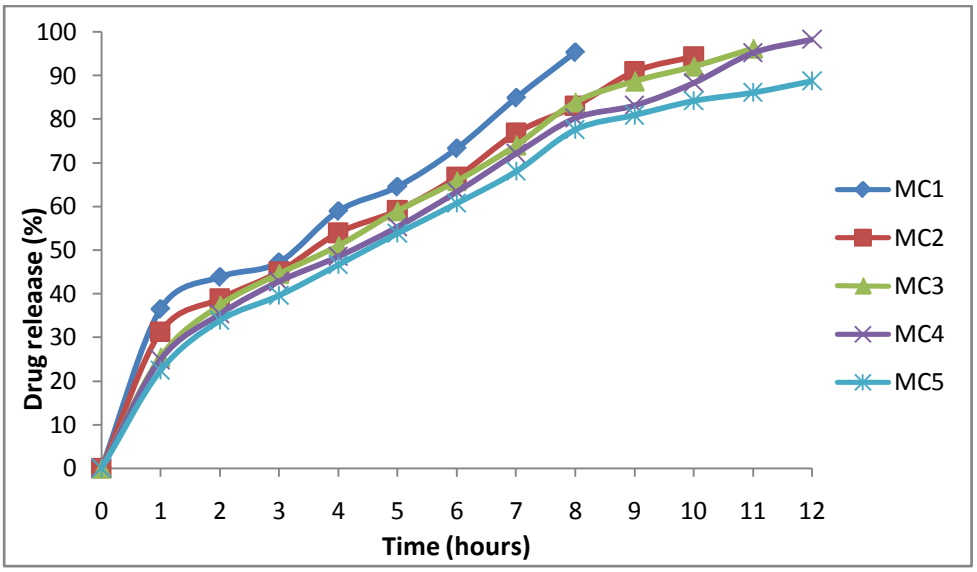


Fig 4. *In vitro* drug release (%) of batch MH1-MH5

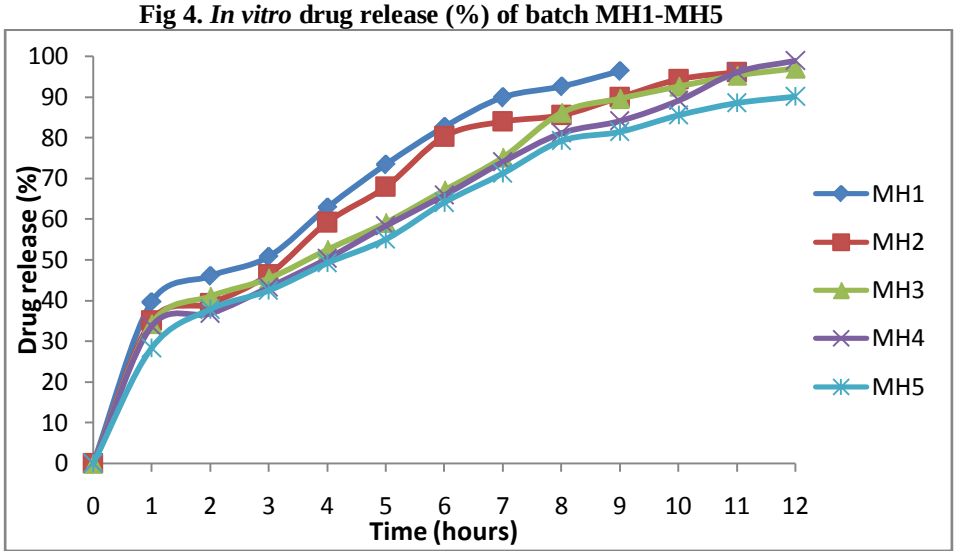


Figure 5. SEM of optimized batch MC4

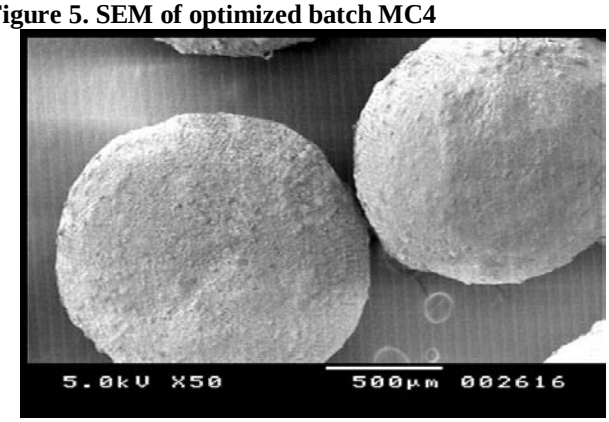
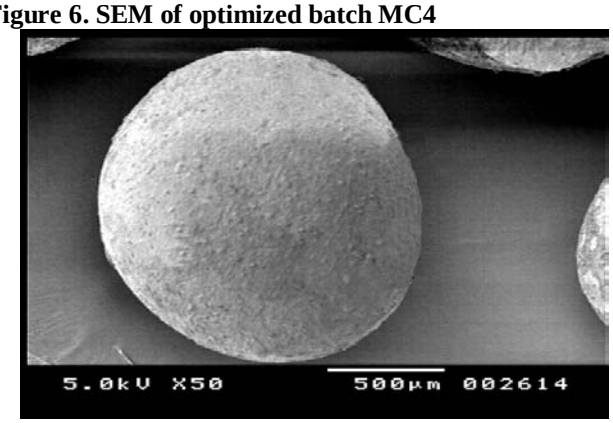


Figure 6. SEM of optimized batch MC4



REFERENCE:

1. Murata K. Kobayashi M, Noda K. Enhancement of bioavailability of dopamine via nasal route in beagle dogs. Chem Pharm Bull (Tokyo). 1992; 40: 2155-2158.
2. Ko JA, Park HJ, Hwang SJ, Park JB, Lee JS. Preparation and characterization of chitosan microparticles intended for controlled drug delivery. Int J Pharm 2002; 249: 165- 174.
3. Amit K. Goyal and Goutam Rath. Evaluation of anti-gerd activity of gastro retentive drug delivery system of itopride hydrochloride. Artifi cial Cells, Blood Substitutes, and Biotechnology, 38: 200–207
4. Shaji J, Poddar A. Intranasal clonazepam mucoadhesive microspheres: factorial designing and primary evaluation. 2009 [cited 2009, Feb.12]
5. Alton A. Pharmaceutics, The science of dosage form, dosage design, 2nd ed; 1996.
6. Manjanna KM, et al. Formulation of oral sustained release Aceclofenac sodium microbeads, Intern J of Pharm tech research 2009; 1(3): 940- 952.
7. Yellanki SK, Singh J, Syed JA, Bigala R, Goranti S, Nerella NK. Design and Characterization of Amoxicillin trihydrate Mucoadhesive Microspheres for Prolonged Gastric retention. International Journal of Pharmaceutical Sciences and Drug Research 2010; 2(2): 112-114
8. Chowdary KPR and Rao YS. Preparation and evaluation of mucoadhesive microcapsules of Indomethacin. Saudi Pharmaceutical journal 2003; 11 (3):97-103.
10. M.A. Altaf, Sreedharan and N. Charyulu. Ionic gelation controlled drug delivery system for gastric mucoadhesive microcapsules of captopril. Indian journal of pharmaceutical sciences. Sep-oct 2008.
11. Ahmed MG, Satish KBP, Kiran KGB. Formulation And Evaluation Of Gastric-Mucoadhesive Drug Delivery Systems Of Captopril. Journal of Current Pharmaceutical Research 2010; 2(1): 26-32.