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Original Article

Studies on Rebamipide Loaded Gastroretentive Alginate Based Mucoadhesive Beads: Formulation & *In-vitro, In-vivo* Evaluation

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ABSTRACT

Introduction: Oral route is the most common and convenient route to deliver the drug. Many oral drug delivery systems were developed to improve drug bio availability; gastro retentive drug delivery system is one of them. Gastroretentive drug delivery system is the system in which a drug can remain in the gastric region for several hours in order to prolong its gastric residence time. The purpose of this work was to develop and evaluate rebamipide loaded gastro retentive alginate based muco adhesive beads. **Materials and Methods:** Rebamipide loaded alginate beads were prepared by ionotropic gelation and polyelectrolyte complexation method. Prepared beads were characterized for IR, DSC, SEM and evaluated for dissolution and stability studies. **Results:** The FTIR spectra revealed that there were no interaction between drug and excipients. The particle size analysis showed that beads with 3% of sodium alginate were spherical in shape. The mucoadhesion study reveals that as concentration of alginate and carbopol 934 increases percentage of mucoadhesion also increases. **Conclusion:** From *in-vivo* study it was concluded that the prepared formulation showed better control on ulcer than that of pure rebamipide. Rebamipide was successfully formulated as gastroretentive floating and mucoadhesive alginate beads by using ionotropic gelation and polyelectrolyte complexation method.

Key words: Rebamipide, Gastroretentive, Alginate beads, Mucoadhesion and Carbopol 934.

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INTRODUCTION

Amongst several routes of administration oral route is the common and convenient to deliver the drug. Many oral drug delivery systems including controlled release has been developed to improve drug bioavailability. However, some of these systems failed to work as planned becauseoral sustained release formulation is subjected to frequently changing environments in the gastrointestinal (GI) tract and variation of the stomach emptying time.[1] It is clear from recent research and patent literature that there is an increased interest in novel dosage forms that are retained in the stomach for a prolonged and predictable period of time. Gastroretentive drug delivery system (GRDDS) is one of the approaches in this area. GRDDS is the system in which a drug can remain in the gastric region for several hours in order to prolong its gastric residence time.[2]

There are various approaches of gastro retention viz. high density, low density, swelling, bio adhesive, expandable, magnetic and ion exchange resin system and all such systems have their own merits and demerits. To achieve a proper gastro retention of the system a combined approach is now day's preferred.[2] Single unit system such as tablets and capsules possess drawbacks like high variability of the gastrointestinal transit time due to their all or none emptying processes, while multiple unit dosage forms shown to reduce the inter- and intra-subject variability in drug absorption with lower risk of dose dumping therefore they may be used as alternative.[3] Multi-particulate system offers advantages such as improvement in bioavailability, better and reproducible pharmacokinetic behavior and these are some reasons to formulate a drug as a multiparticulate system. Muco adhesive multi-particulate have advantages such as efficient absorption and enhanced bioavailability of drugs owing to a high surface-to-volume ratio, a much more contact with mucus layer and specific targeting of drugs for local action.

Rebamipide commonly referred as a muco-protective agent, is an anti-ulcer drug that protects the gastric mucosa against acute damage caused by various noxious agents and accelerates gastric ulcer healing without affecting gastric acid secretion. It also prevents ulcer relapse and delay of gastric ulcer healing caused by *H. pylori* infection.[4] Gastro protective drugs like rebamipide have less adverse effects than anti-secretory drugs such as PPI and H_2-receptor antagonist. They are also superior to prostaglandin derivatives.[5] Rebamipide is BCS class IV drug. The oral bioavailability of rebamipide is very low at around10%, therefore this drug is used as a model drug to plan different strategies for improving bioavailability. Hence in present study attempts were made to improve bioavailability of rebamipide by formulating particulate gastro retentive drug delivery system assuming that increase in gastric residence time will increase maximum absorbable dose.[6]

The objectives of present investigation were to formulate and evaluate the alginate based gastro retentive beads of rebamipide, to evaluate performance of beads *in-vivo* and to carry out stability studies.

MATERIALS AND METHODS

Rebamipide was supplied as a gift sample from Macleods Pharmaceuticals Pvt. Ltd. Mumbai, India. Sodium alginate, carbopol 934 and sodium bicarbonate were purchased from Loba Chemie Pvt. Ltd. Mumbai, India. All other ingredients were of analytical grade.

Preparation of alginate beads

The beads were prepared by ionotropic gelation and polyelectrolyte complexation method. The weighed amount of the drug was thoroughly mixed with de-ionized water by using mechanical stirrer maintained at the speed of 500-600 rpm in one beaker. In another beaker sodium alginate was dispersed in de-ionized water (5 ml) under continuous stirring for 30 min. To this dispersion the desired amount of carbopol 934 was mixed and the entire mixture was stirred for 15 mins. The resulted bubble free, homogeneous dispersion was then added to beaker containing drug and finally sodium bicarbonate was added. The resultant

mixture was extruded into 80 ml of solution containing, calcium chloride (6% w/v) and citric acid (2%w/v) , through hypodermic syringe with needle (21G) at a height of 6 cm and stirred at 100 rpm using magnetic stirrer. The formed beads were allowed to cure for 30 min in the calcium chloride solution to complete the gelation reaction before being filtered and then dried in oven at 50°C.

The formulation composition of rebamipide loaded alginate beads is shown in Table 1.

Evaluation of prepared alginate beads

Percentage yield[7]

The percentage yield of all formulation batches were calculated by using formula

$$\text{Percentage yield} = \frac{\text{Practical yield}}{\text{Theorotical yield}} \quad 100$$

Drug entrapment efficiency

Drug entrapment efficiency was determined by indirect method. The method utilizes filtrates of polyelectrolyte solution (i.e. calcium chloride) from each batch. One ml of the filtrate was taken which was further diluted up to 10 ml with 0.1 N HCL: methanol in 1:1 proportion (media) and absorbance was recorded by UV-visible double beam spectrophoto meter(Shimadzu-1601, Tokyo) at 230 nm.

Particle size determination

Particle size distribution of the rebamipide loaded alginate beads was determined by optical microscopy using calibrated ocular eyepiece. Fifty beads were evaluated and the experiment was performed.

Floating behavior

The *in-vitro* floating behavior of rebamipide loaded alginate beads were studied by using 900 ml 0.1N HCl at $37^0 \pm 0.5^0$C. The speed of rotation was maintained at 50 rpm. The floating lag time (the period between placing beads in the medium and buoyancy beginning) and floating duration of beads were determined by the visual observation.

In-vitro wash-off test for mucoadhesion[7,8]

The muco adhesive properties of the rebamipide loaded beads were evaluated, using an *in-vitro* wash-off test. A 2×2 cm piece of goat stomach mucosa was tied onto a glass slide. Beads were spread (approximately 50) onto the wet rinsed tissue specimen and the prepared slide was hang onto one of the groves of a USP tablet disintegration test apparatus, with continuous oxygen supply. The disintegration test apparatus was operated, giving the tissue specimen regular up and down movements within the beaker of the disintegration apparatus, which contains 0.1 N HCL (pH=1.2). At specific time interval bead adhered onto the tissue was counted.

$$\text{Percentage of adhesion} = \frac{\text{Number of beads adhered}}{\text{Number of beads applied}} \quad 100$$

In-vitro dissolution studies

The *in vitro* release of the rebamipide from alginate beads was studied in 900 ml of media 0.1 N HCL at 37 ± 0.5^0C by using USP type II apparatus. A sample of beads equivalent to 300 mg of rebamipide was used in each test. 5ml of aliquots of dissolution fluid were withdrawn and the same amount of fresh medium was replaced to maintain the sink condition at different time intervals. Then samples were filtered and assayed at 230 nm for rebamipide content using a Shimadzu-1700 UV-visible double beam spectrophotometer.

Kinetic modeling and mechanism of drug release

The release data obtained were fitted to zero order, first order, higuchi, Hixson crowell and koresmeyerpeppas equation to determine the mechanism of drug release from the rebamipide loaded alginate beads.

Micromeritic studies

The prepared beads were evaluated for micromeritic properties such as angle of repose, Carr's index and hausner's ratio.

Scanning electron microscopy

To detect the surface morphology, the surface morphological study was done using Jeol, JSM-6360 (LA) model, Japan. The rebamipide and rebamipide loaded beads were mounted onto stubs using double sided adhesive tape. The mounted samples were sputter coated under an inert atmosphere with gold palladium and examined at 20 kV accelerating voltage.

Fourier Transform Infrared spectroscopy

FTIR spectroscopy was conducted using a Jasco FTIR 4100 Spectrophotometer and the spectrum was recorded in the wavelength region of 4000–400 cm^{-1}. The procedure consists of dispersing a sample (Rebamipide alone, physical mixture and rebamipide loaded alginate beads) in KBR and compressing into discs by applying a pressure of 5 t for 5 min in a hydraulic press. The pellet was placed in the light path and the spectrum was recorded.

Differential Scanning Calorimetry

Thermo grams of rebamipide, physical mixture and rebamipide loaded beads were obtained by using SDT Q600 V20.9 Build 20 Universal V4.5A TA instrument. Sample was placed in an aluminum pan and then hermetically sealed with an aluminum lid. The thermograms were obtained at a scanning rate of 10°C/min over a temperature range of 40°C to 500°C.

X-Ray Diffraction

Rebamipide, physical mixture and rebamipide loaded beads were subjected to X-ray diffraction study in a D2 phaser X-ray diff racto meter.

Stability studies

Short-term stability studies were performed on optimized formulation. Stability study was carried out by storing the alginate beads at room temperature (RT) for 1 month period. At the end of month the beads were examined for any physical change and changes in *in-vitro* dissolution studies.

In-vivo indomethacin induced antiulcer activity in rats[9,10]

Groups of 6 Wistar rats weighing 100-200 gms were used. Three groups were prepared and labeled as control, standard and test. The stock solution of indomethacin was prepared in 0.1% tween 80 and RBM in 0.5% of carboxy methyl cellulose. The animals from control group received only Indomethacin at a dose of 25 mg/kg, p.o. The standard drug (RBM, 30 mg/kg, p.o.) was administered to the animals of standard group and immediately after 10 min Indomethacin (25 mg/kg) was administered orally. The test formulation i.e. alginate beads were administered orally to the animals of test group at a dose of 30 mg/kg in a water. Six hours later the rats were sacrificed and stomach form each rat was isolated. Formol saline (2% v/v) is then injected into the totally legated stomach for overnight storage. On next day the stomachs were opened along greater curvature and then washed in warm water. The number of lesions were measured and summated to give total lesion score. The numbers of ulcers were noted and the severity recorded with the scores like 0=no ulcer, 1=superficial ulcers, 2=deep ulcers, 3=perforation. An ulcer index U_I is calculated by using following formula:

$$\boldsymbol{U_I = U_N + U_S + U_P \times 10^{-1}}$$

Table 1: Formulation composition of rebamipide loaded alginate beads

Batch code	Drug (mg)	Sodium alginate (mg)	Carbopol 934 (mg)	Sodium bicarbonate (mg)	Calcium chloride (% w/v)	Citric acid (% w/v)
G_1	300	100	25	750	6	2
G_2	300	100	50	750	6	2
G_3	300	100	75	750	6	2
G_4	300	300	25	750	6	2
G_5	300	300	50	750	6	2
G_6	300	300	75	750	6	2
G_7	300	500	25	750	6	2
G_8	300	500	50	750	6	2
G_9	300	500	75	750	6	2

Table 2: Evaluation parameters of rebamipide loaded alginate beads

Sr. no	Batch code	Percentage yield* (%)	Entrapment efficiency (%)	Mean particle size (mm)	Floating lag time (Sec)	Total floating time (Hrs.)
1	G_1	51.28 ± 0.022	95.79 ± 0.070	0.06 ± 0.02	180 ± 5	24 ± 0.5
2	G_2	48.33 ± 0.020	95.40 ± 0.068	0.07 ± 0.018	187 ± 3	21 ± 0.25
3	G_3	60.78 ± 0.024	94.16 ± 0.072	0.09 ± 0.017	196 ± 7	20 ± 0.75
4	G_4	78.86 ± 0.018	96.85 ± 0.075	0.11 ± 0.014	245 ± 4	18 ± 0.5
5	G_5	66.66 ± 0.021	94.23 ± 0.074	0.13 ± 0.018	251 ± 6	16 ± 0.75
6	G_6	74.86 ± 0.024	94.93 ± 0.076	0.14 ± 0.015	269 ± 2	15 ± 0.0
7	G_7	62.06 ± 0.021	95.34 ± 0.071	0.15 ± 0.02	307 ± 5	14 ± 0.25
8	G_8	69.52 ± 0.023	95.58 ± 0.070	0.16 ± 0.019	322 ± 9	12 ± 0.5
9	G_9	65.96 ± 0.020	95.48 ± 0.074	0.18 ± 0.017	362 ± 6	12 ± 0.75

* Indicates values in mean ± SD (n =3)

Where, U_N = average of number of ulcers per animal, U_S = average of severity score, U_P = percentage of animals with ulcers.

RESULTS

The results of evaluation parameters of rebamipide loaded alginate beads like percentage yield, entrapment efficiency; mean particle size, floating lag time and total floating time were shown in Table 2. The percentage yield of all batches ranged between 48.33% ± 0.020 to 78.86% ± 0.018. Batch G_4showed highest percentage yield of about 78.86% ± 0.018.

All formulations showed entrapment efficiency ranging from 94.16% ± 0.072 to 96.85%±0.075. There was no significant difference in the entrapment efficiency of all nine batches, and showed excellent entrapment of drug which may be due to the "Egg Box" type arrangement.[12]

The mean particle size of batches ranged from 0.06 mm ± 0.02 to 0.18 mm ± 0.017. As the concentration of sodium alginate increased during the formulation of beads, the shape of the beads become almost spherical and mean bead diameter increased due to increase in micro-viscosity of the polymeric dispersion. The beads containing 3%w/v of sodium alginate was spherical in shape and had good mechanical strength. As concentration of sodium alginate increased further above 3%w/v, tailing phenomenon was observed.[12]

The floating lag times of all batches were less than 6 mins. The total floating time was ranged from 12 ± 0.75 hrs to 24 ± 0.5 hrs as shown in Table 2. As the concentration of polymers increased total floating time decreased along with increase in the floating lag time. Figure 1 shows floating behavior of rebamipide loaded alginate bead.

The floating property of beads was induced by evolution of carbon dioxide when it comes in contact with acidic environment followed by the ability of polymer gel to entrap it which decreased their density below one.[12]

Floating lag time is nothing but the period between placing beads in the medium and buoyancy beginning. The system should float within few minutes after contact with gastric fluid to prevent dosage form from emptying into small intestine along with food. From above results

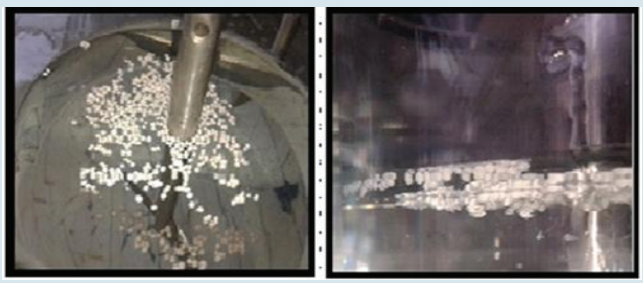

Figure 1: Floating behavior of rebamipide loaded alginate beads.

Figure 2: Muco adhesion of rebamipide loaded alginate beads on goat stomach mucosa.

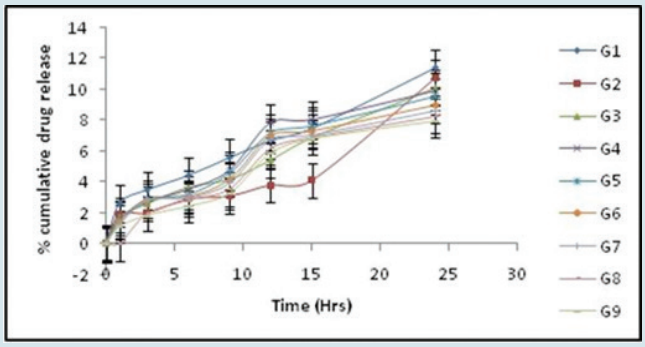

Figure 3: *In-vitro* dissolution profile of batches (G_1-G_9).

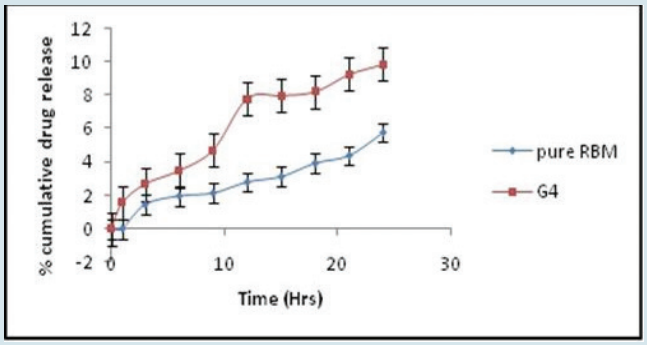

Figure 4: *In-vitro* dissolution profile of pure drug and G4 batch.

of floating lag time it was observed that the beads were floated within few minutes. Here, floating lag time increased and total floating time decreased from G_1 to G_9 batch, this was due to higher concentration of polymers from G1 to G9 as higher amount of sodium bicarbonate was needed to lift the polymers.

The percentage of muco adhesion at the end of 30 mins was found to be 60 ± 0.020 % for G1 batch and 86 ± 0.025 % for G9 batch. The percentage of muco adhesion at the end of 6 hrs was found to be 36 ± 0.020 % for G1 batch and 58 ± 0.014 % for G9 batch. The Figure 2 shows muco adhesion of beads on goat stomach mucosa.

From above results it was indicated that as the concentration of polymers increases percentage of mucoadhesion also increases. The results of *In-vitro* wash-off test for muco adhesion of G1 to G9 batch is shown in Table 3.

As the concentration of carbopol 934 and alginate increased the percentage of muco adhesion also increased since a greater amount of polymer results in a higher amount of free–COOH group which are responsible for binding to the sialic acid group within the mucus network. Hence it resultedinto an increase in muco adhesive properties of beads.[7] Hence there was an increase in percentage of muco adhesion from G_1 to G_9 batch.

The cumulative percentage of drug released of G_1 batch was highest among all the nine batches 11.39 ± 0.014% and that of G_9 batch was lowest 7.94 ± 0.017%, where that of G_4 batch was 9.88 ± 0.015%.

The drug release was decreased as concentration of sodium alginate increased. This was due to the 3D gel network, which was formed due to reaction between calcium ions and alginate, which restricted further diffusion of drug molecules and resulted in the sustained drug release. Pure drug showed very less cumulative percentage of drug release i.e. 5.78% ± 0.018 at the end of 24 hr. It was known that as the solubility of drug was low the time available for drug dissolution becomes less adequate and thus transit time become a significant factor affecting drug absorption, hence gastro retention was made to increase absorption of rebamipide by increasing the residence time by taking the support of maximum absorbable dose equation. This can be further supported by *in-vivo* studies.

$$MAD = K_a S_{GI} V_{GI} t_r$$

Where, MAD is the maximum absorbable dose in mg, K_a is the intrinsic absorption rate constant, S_{GI} is the solubility of the drug in the GI fluid, V_{GI} is the volume of GI fluid present, t_r is the residence of drug in GI.[20] Therefore, by taking the support of above equation, hypothesis is assumed that by increasing the residence time there will be increase in the maximum absorbable dose which can be further supported by *in-vivo* study.

Table 3: *In-vitro* wash-off test for muco adhesion of G1 to G9 batch

Time (Hrs)	Percentage of muco adhesion* (%)								
	G_1	G_2	G_3	G_4	G_5	G_6	G_7	G_8	G_9
0.5	60 ± 0.020	62 ± 0.023	64 ± 0.014	68 ± 0.014	74 ± 0.020	76 ± 0.012	78 ± 0.018	82 ± 0.022	86 ± 0.025
1	54 ± 0.019	58 ± 0.021	56 ± 0.018	60 ± 0.021	64 ± 0.022	68 ± 0.023	72 ± 0.014	74 ± 0.021	78 ± 0.023
2	42 ± 0.024	52 ± 0.024	56 ± 0.022	58 ± 0.022	60 ± 0.024	62 ± 0.025	68 ± 0.019	66 ± 0.025	64 ± 0.021
3	38 ± 0.018	48 ± 0.023	52 ± 0.021	58 ± 0.021	56 ± 0.019	58 ± 0.018	64 ± 0.022	58 ± 0.024	62 ± 0.024
4	38 ± 0.022	44 ± 0.021	48 ± 0.025	52 ± 0.017	56 ± 0.024	58 ± 0.022	64 ± 0.024	56 ± 0.023	62 ± 0.018
5	38 ± 0.024	40 ± 0.024	42 ± 0.026	46 ± 0.023	52 ± 0.024	58 ± 0.014	54 ± 0.024	50 ± 0.024	60 ± 0.019
6	36 ± 0.020	40 ± 0.020	40 ± 0.024	46 ± 0.025	48 ± 0.021	46 ± 0.025	42 ± 0.019	44 ± 0.021	58 ± 0.014

* Indicates values in mean ± SD (n=3)

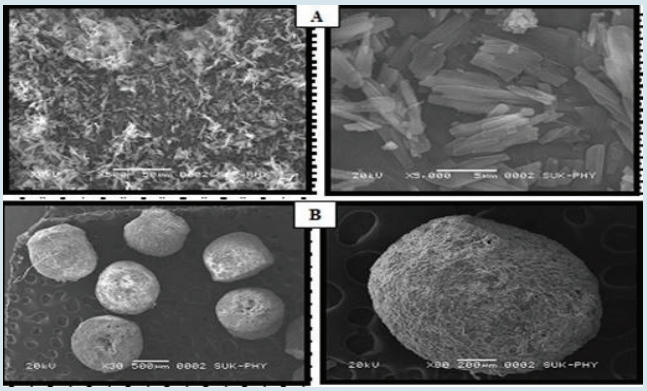

Figure 5: Scanning Electron Microphotographs of Rebamipide (A), Drug loaded alginate beads (B).

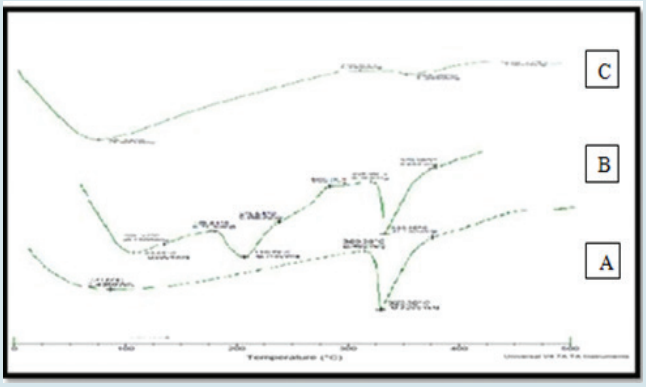

Figure 7: DSC spectra of rebamipide (A), physical mixture (B) and rebamipide loaded alginate beads (C).

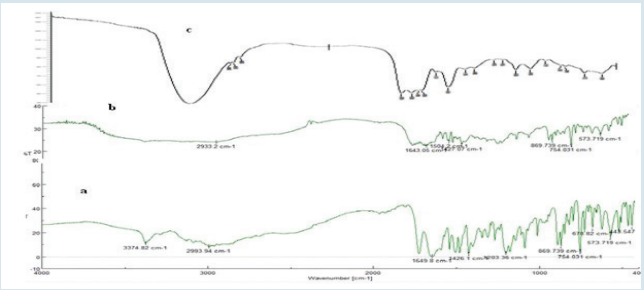

Figure 6: FTIR spectra of rebamipide (a), physical mixture (b) and rebamipide loaded alginate beads (c).

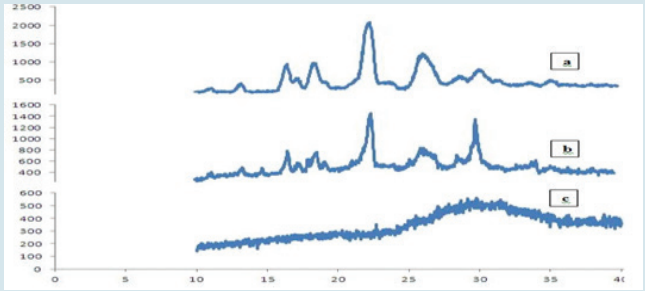

Figure 8: XRD patterns of rebamipide(a), physical mixture(b) and rebamipide loaded alginate beads(c).

From the results of percentage yield, entrapment efficiency, it was found that batch G_4 showed highest percentage yield and entrapment efficiency amongst all nine batches. From floating behavior it was found that the batch G_4 has a total floating time 18 hrs ± 0.5 and floating lag time 245 sec ± 4. *In-vitro* wash off test for muco adhesion shows percentage of muco adhesion 68 ± 0.014% for G_4 batch with 9.88 ± 0.015% drug release at the end of 24 hr. Hence, G_4 batch was selected as optimized batch for further evaluation.

Figure 2 and 4 shows the *in-vitro* dissolution profile of all nine batches (G_1-G_9) were shown in Figure 3 and comparison between pure drug and batch G_4 respectively.

From the released kinetic data table it was found that regression coefficient for zero order was 0.9414, for first order 0.8539, for Higuchi 0.9623 and for KorsmeyerPeppas model 0.9688 as shown in Table 4.

From Table 4 it was concluded that the kinetic data was best fitted to Korsmeyer and Peppas model and good regression coefficient was observed i.e. 0.9688. The value of diffusion exponent (slope) n was found to be 0.664 indicating drug releases from beads by non-fickian mechanism.

Determination of angle of repose, Carr's index and Hausner's ratio is important because it may influence compressibility and dissolution. The Table 5 shows the micromeritic properties of optimized batch i.e. batch G_4.

The prepared beads showed improvement in flow properties than pure drug. A powder characteristic of drug indicates poor flow property and turned to excellent when formulated in the form of alginate beads.[13,14]

Figure 5 shows the appearance of surface morphology of the rebamipide and rebamipide loaded alginate beads using scanning electron microscopy which was indicated that the rebamipide have needle type small crystals and beads were spherical in shape with slightly rough surface.

The results of scanning electron microscopy revealed that the rebamipide was crystalline in nature and visible large wrinkles on rebamipide loaded alginate beads have a sandy appearance which may be due to the surface-associated crystals of drug.[15]

FTIR spectra of rebamipide, physical mixture and rebamipide loaded alginate beads were shown in Figure 6. In case of rebamipide, the bands were observed in the region of 3300-3500 cm^{-1} (3374.82) due to N–H stretching vibration. Bands were observed at 2993.94 cm^{-1} due to =C-H stretching of aromatic hydrocarbons, 1726.94 cm^{-1} due to =C-O stretching of carboxylic acid, 1203.36 cm^{-1} due to C-N stretching, 754.031 cm^{-1} due to C-H bending vibrations of aromatic ring.[11] The same peaks were observed in the physical mixture, but in case of rebamipide loaded alginate beads a slight change in wave number and intensity of some peaks was observed.

From FTIR spectra's it was observed that all characteristic peaks of rebamipide were present in physical mixtures, which indicates that there were no interaction involving bond formation between drug and excipients. In case of rebamipide loaded alginate beads the cross-linking process of sodium alginate with calcium caused an obvious shift of wave number and change in the intensity of peak. This indicated the presence of an ionic bond between the calcium ion and carboxyl group of sodium alginate and partial covalent bonding between calcium and oxygen atom.[15]

DSC spectra of rebamipide showed endothermic peak at 323.30°C and same endothermic peak was observed in physical mixture. DSC spectra of rebamipide loaded alginate beads showed disappearance of endothermic peak of rebamipideat 323.30°C (Figure 7).

From the DSC spectra of rebamipide and physical mixture it was con-

Table 4: Study of various kinetic models on in-vitro drug release of Rebamipide loaded alginate beads

Formulation Code	Regression Coefficient (R^2)				
	Zero order	First order	Higuchi Model	Hixson Crowell Model	Korsmeyer-Peppas Model
G_4	0.9414	0.8539	0.9623	0.9414	0.9688

Table 5: Micromeritic properties of optimized batch G_4.

Sr. no	Powder characteristic	Value*	Flow property
1	Angle of repose	23.74° ± 0.01	Excellent
2	Carr's index	14.13% ± 0.014	Excellent
3	Hausner's ratio	1.14 ± 0.012	Excellent

* Indicates values in mean ± SD (n = 3)

Table 6: *In-vitro* dissolution study of optimized batch G_4 after one month

Parameter	Before*	After*
In-vitro dissolution study (G_4)	9.88% ± 0.015	8.95% ± 0.014

* Indicates values in mean ± SD (n = 3)

Table 7: Ulcer index in control, standard and test groups

Groups	Ulcer index (U_I)
Control (Indomethacin)	13.5
Standard (Rebamipide)	12.5
Test(Formulation)	0

firmed that there were no interaction between drug and excipients.

Absence of sharp endothermic peak of rebamipide in formulation supports the PXRD studies for reduced crystallanity. The broadening of peak was observed in the DSC spectra of rebamipide loaded alginate beads which may be due to the dilution effect by polymers.

By X-ray diffraction a pattern of pure rebamipide was compared with rebamipide loaded alginate beads (Figure 8).

The XRD scan of pure rebamipide showed sharp intense peaks indicating crystallanity. But the XRD patterns of the drug loaded beads showed halo pattern with less intense and denser peaks compared to plain RBM which was indicative of reduction in crystallanity or partial amorphinization of the drug in beads.

The relative degree of crystallanity (RDC) of RBM in prepared beads was calculated according to the equation RDC = Isam/Iref, where Isam is the peak height of the sample under investigation and Iref is the peak height at the same angle for the reference with the highest intensity. The peak height at 20.36° was used for calculating the RDC. The RDC was found to be 0.119. The XRD pattern of prepared beads exhibited reduction in intensity of peak compared to pure rebamipide indicating reduced crystallanity of the drug in prepared beads.

Short-term stability studies of the formulation G_4 (Table 6) shows that there were no significant changes in physical appearance and *in vitro* dissolution studies at the end of one month.

From stability study it was concluded that the prepared formulation was stable for one month.

The results obtained from *in-vivo* study are shown in Table 7.

The prepared formulation showed better control on ulcer than that of pure rebamipide indicating improved oral bioavailability with local effects due to gastro retention.

In case of *iv-vivo* study the improvement in the ulcer healing of prepared rebamipide loaded alginate beads than that of pure rebamipide was due to the local effect of formulation because of gastro retention or improved absorption. The reason for improved ulcer healing or gastro protection in case of alginate beads might be due to the increased contact with mucus membrane due to muco adhesion.

CONCLUSION

Rebamipide was successfully formulated as alginate mucoadhesive beads using ion tropic gelation and polyelectrolyte complexation method. The gastro retention of rebamipide by muco adhesive approach proven a permissible mean of drug delivery. From *in-vivo* study it was concluded that the prepared formulation showed better control on ulcer than that of pure rebamipide.

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

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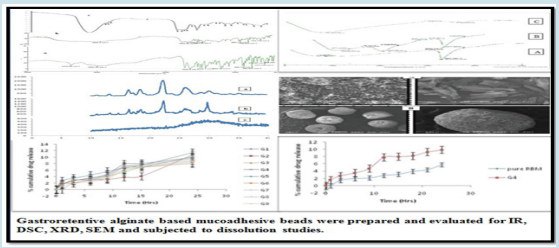
PICTORIAL ABSTRACT



SUMMARY

- Rebamipide loaded gastroretentive alginate based mucoadhesive beads were prepared and evaluated.
- No chemical interaction was found between drug and polymers.
- The prepared formulation showed better control on ulcer than that of pure rebamipide.

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