

# RESEARCH AND REVIEWS: JOURNAL OF PHARMACY AND PHARMACEUTICAL SCIENCES

## Preparation and Characterization of Tegaserod Maleate Containing Targeted Microspheres for Colonic Drug Delivery System.

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### Research Article

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#### ABSTRACT

The aim of study was to develop and evaluate tegaserod maleate (TM) microspheres for colonic drug delivery system. Microspheres of TM were prepared by with little modification in the coacervation phase separation method by using polymer sodium alginate. These microspheres were evaluated for particle size, percentage drug entrapment and *in-vitro* drug release studies. The mean particle size of prepared microspheres was found to be in a range of 713.00 - 823.66  $\mu\text{m}$ . Percentage drug entrapment observed in all formulations was between 62.28 - 66.88 %. *In-vitro* drug release studies showed that the release of tegaserod maleate from the microspheres was mainly influenced by the polymer concentration. It is concluded from the present investigation that TM-alginate microspheres are promising controlled release carriers for colon targeted delivery.

#### INTRODUCTION

There are various approaches in delivering a therapeutic substance to the target site in a controlled release fashion. One such approach is using microspheres as carriers for drugs. Microspheres are characteristically free flowing powders consisting of proteins or synthetic polymers which are biodegradable in nature. Microsphere based drug delivery have received considerable attention in recent years. The most important characteristic of microspheres is the micro phase separation morphology which endows it with a controllable variability in degradation rate and also drug release. Biodegradable microspheres can be prepared from certain synthetic as well as natural polymers. An important requirement of such polymers is that the degradation products should be non-toxic because such products eventually enter circulation or result in tissue deposition. Biodegradability carrier matrices can be designed to deliver the therapeutic agent for periods ranging from a few days to a few years.

Tegaserod is being developed as a treatment for constipation-predominant irritable bowel syndrome (IBS).<sup>[1]</sup> Tegaserod is a potent partial agonist of serotonin type-4 (5-HT<sub>4</sub>) receptors located in the GI tract. The mechanism of action of Tegaserod is reflected in its stimulation of the peristaltic reflex intestinal secretion, as well as inhibition of visceral sensitivity via activation of 5-HT<sub>4</sub> receptors in the gastrointestinal tract. Tegaserod acts as a partial agonist at neuronal 5-HT<sub>4</sub> receptors triggering the release of further neurotransmitters such as calcitonin gene related peptide from sensory neurons.<sup>[2,3]</sup>

The recommended dosage of Tegaserod is 6 mg (i.e equivalent to 8.3 mg of Tegaserod maleate) taken twice daily orally before meals for 4-6 weeks. Tegaserod is rapidly absorbed following oral administration; peak plasma concentrations are reached after approximately 1 hour. Tegaserod is approximately 98% bound to plasma proteins, primarily to  $\alpha$ 1-acid glycoprotein. The plasma clearance of Tegaserod is 77  $\pm$ 15 L/h, with an estimated terminal half-life (t<sub>1/2</sub>) of 11  $\pm$  5 h following intravenous

administrations. Approximately two-thirds of the orally administered dose of Tegaserod is excreted unchanged in the feces, with the remaining one third excreted in the urine.<sup>[2,3,4]</sup>

Aim of this study was to explore the feasibility of colon-specific drug delivery for treatment of irritable bowel syndrome and constipation with single daily dose of tegaserod. Hence, it is worthwhile to formulate and evaluate colon specific microspheres by using suitable polymer. So, an attempt will be made to formulate a dosage form, which could be formulated easily, with little modification in the method of processing of the ingredients.

## MATERIALS AND METHODS

### Materials

Tegaserod maleate, sodium alginate, ethanol and CaCl<sub>2</sub>.

### Preparation of Microspheres

A solution was prepared by dissolving 0.60 gm of tegaserod in 5ml ethanol. The solution was dispersed in 100 ml sodium alginate solution (3% w/v). The resulting solution was dropped through a 26G syringe needle into 1% (w/v) CaCl<sub>2</sub> solution. The solution containing suspended microspheres was stirred with a magnetic stir bar for 10 min to improve the mechanical strength of the microspheres. The fully formed microspheres were collected, washed with water and subsequently freeze dried. Then they were used for further analysis.<sup>[5]</sup>

The preliminary study performed for microspheres formulation with 1:0.5, 1:1, 1:1.5 ratio for drug to polymer. It was found that 1:0.5 ratio for drug to polymer release the drug faster and 1:1.5 ratio for drug to polymer retard the release of drug, while 1:1 ratio for drug to polymer release the drug appropriately so, 1:1 ratio was decided for the final batches of microsphere formulations by using 5%, 10% calcium chloride as crosslinking agent.

### Evaluation of microspheres

The microsphere was formulated with above composition were evaluated for following micromeritics properties:

#### Angle of repose

The angle of repose for the microsphere of each formulation was determined by the funnel method. The microsphere was allowed to flow out of the funnel orifice on a plane paper kept on the horizontal surface, this forms a pile of microspheres on the paper. The angle of repose was calculated by substituting the values of the base radius 'R' and pile height 'H' in the following equation.<sup>[6,7]</sup>

$$\tan\theta = H / R$$

Where, H = pile height, R = radius of pile

Therefore;  $\theta = \tan^{-1}(H / R)$

#### Bulk density and tapped density

Both loose bulk density (LBD) and tapped bulk density (TBD) were determined. A quantity of 2g of microsphere from each formula was lightly shaken to break agglomerates if any and then was introduced into a 10 ml-measuring cylinder. It was allowed to fall under its own weight onto a hard surface from the height of 2.5 cm at 2- second intervals. The tapping was continued until no further change in volume was noted. Loose bulk density (LBD) and tapped bulk density (TBD) were calculated using the following formulae.<sup>[7,8,9]</sup>

$$\text{LBD} = \text{Weight of the microsphere} / \text{volume of the packing}$$

$$\text{TBD} = \text{Weight of the microspheres} / \text{tapped volume of the packing}$$

#### Compressibility index

The compressibility indices of the formulation blends were determined using Carr's compressibility index formula.<sup>[6,7]</sup>

$$\text{Carr's compressibility index (\%)} = \frac{[(\text{TBD} - \text{LBD}) / \text{TBD}] \times 100}{}$$

### Hausner's ratio

Hausner's ratio of microspheres was determined by comparing the tapped density to the bulk density using the equation.<sup>[7,8]</sup>

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

### Particle size analysis

The particle size was measured using a stage micrometer, and the mean particle size was calculated by measuring 200 particles with the help of a calibrated stage micrometer. A small amount of dry microspheres was suspended in liquid paraffin (10 ml). A small drop of suspension thus obtained was placed on a clean glass slide. The slide containing microspheres was mounted on the stage of the microscope and diameter of at least 100 particles was measured using a calibrated optical micrometer.<sup>[10]</sup>

### Percentage yield

The percentage yield of different formulations was determined by weighing the microspheres after drying. The percentage yield was calculated as follows

$$\% \text{ Yield} = (\text{Total weight of microspheres} / \text{Total weight of drug and polymer}) \times 100$$

### Drug entrapment

The various batches of the microspheres were subjected to estimation of drug content. The microspheres equivalent to 100 mg of Tegaserod, were accurately weighed and crushed. The powdered microspheres were placed in 100 ml of methanol for overnight. This solution is then filtered through whatmann filter paper. After filtration, use this clear supernatant solution to measured absorbance at 291 nm by using UV-visible spectrophotometer. The percentage drug entrapment was calculated.

$$\% \text{ Drug entrapment} = (\text{Calculated drug concentration} / \text{Theoretical drug concentration}) \times 100$$

### In-vitro release studies

The release rate of TM was determined using USP dissolution testing apparatus II (Paddle type). The dissolution test was performed using 900 ml of 0.1 N HCl, at  $37 \pm 0.5^\circ\text{C}$  and speed of 75rpm for 2hrs. Aliquot (10 ml) of the solution was collected from the dissolution apparatus hourly and were replaced with fresh dissolution medium. After this the dissolution medium was replaced with 900 mL phosphate buffered (pH 6.8), and the dissolution continued for another 6 hrs. Then study at a pH of 7.4 buffer medium is carried out till completion of 12-15hr at 75rpm and  $37 \pm 0.50\text{C}$ . The experiments in rat fecal slurries and in without rat fecal slurries were carried at pH 7.4. The aliquots were filtered through whatmann filter. Absorbance of these solutions was recorded at 220nm and 221nm for pH 6.8 and pH 7.4 respectively. Aliquots were withdrawn at one hour interval from a zone midway between the surface of dissolution medium and the top of rotating paddle not less than 1 cm apart from the vessel wall. Drug content in dissolution sample was determined by software (PCP disso v2.08) version.<sup>[11]</sup>

## RESULTS AND DISCUSSION

### Angle of repose

Angle of repose of TM-alginate microspheres was determined by fixed funnel method. Angle repose of these microspheres was observed in range of 23 and  $26^\circ$  suggesting good flow properties of microspheres. (Table No.1)

### Bulk density and tapped density

Bulk and tapped density was determined by tapping method. The bulk density value of different batches of TM-alginate microspheres was determined which was summarized in Table No.1

### Carr's compressibility index

The Carr's compressibility index values for microspheres were ranged between 13.50-16.50. The value less than 20 for all formulation suggested good flow property of microspheres. (Table No.1)

### Hausner's ratio

Hausner's ratio of microspheres was determined by comparing the tapped density to the bulk density. It was in ranged from 1.1-1.25; i.e. all the formulation showed that they had excellent flow properties. (Table No.1)

**Table 1: Properties of TM-alginate alginate microspheres.**

Formulations	Parameter	Angle of repose (°)	Bulk density (g/ ml)	Tapped density (g/ ml)	Compressibility Index (%)	Hausner's ratio
		Mean ± SD (n=3)				
F1		24.47± 0.67	0.448±0.019	0.575± 0.005	22.197± 2.57	1.286± 0.043
F2		24.38 ±0.94	0.452± 0.008	0.575± 0.006	21.358± 2.25	1.272± 0.037
F3		24.84± 0.32	0.448± 0.01	0.564± 0.016	20.562± 4.11	1.261± 0.066
F4		24.49± 0.73	0.450± 0.009	0.571± 0.007	21.061± 2.01	1.267± 0.032

### Particle size

Particle size of different batches of microspheres was determined by stage micrometer equipped with optical microscope. Analysis was carried out by observing the slide containing microspheres under the microscope. The average particle size of the microspheres was calculated and it was in the range between 711-826 µm. (Table No.2)

### Percentage yield

The percentage yield of different batches was determined by weighing the microspheres after drying. The percentage yields of different formulation of TM-alginate microspheres were in range of 61.58-65.58 %. (Table No.2)

### Drug entrapment efficiency

The drug entrapment efficiency of different batches of microspheres was determined. The entrapment efficiency was in the range of 64.97-70.12%, as shown in Table No.2. Drug entrapment efficiency was increased when the crosslinking agent increases. 10% of CaCl<sub>2</sub> shows the maximum drug entrapment as compared with the 5% of CaCl<sub>2</sub>. The drug entrapment efficiencies were increased with decreasing sodium alginate blend ratios and increasing cross-linking concentrations. This may be due to the high degree of cross-linking.

**Table 2: Particle size, percentage yield and entrapment efficiency of different batches of TM-alginate microsphere.**

Formulations	Parameter	Mean particle size (µm)	Percentage yield (%)	Drug Entrapment efficiency (%)
		Mean ± SD (n=3)		
F1		713± 1.78	64.89± 0.60	68.66 ± 0.95
F2		822.33± .36	64.36± 0.75	66.88 ± 1.08
F3		823.66±1.86	62.28 ± 0.95	65.84 ± 0.73
F4		758.33±.51	62.60 ± 0.80	69.07 ± 1.14

### In-vitro drug release study

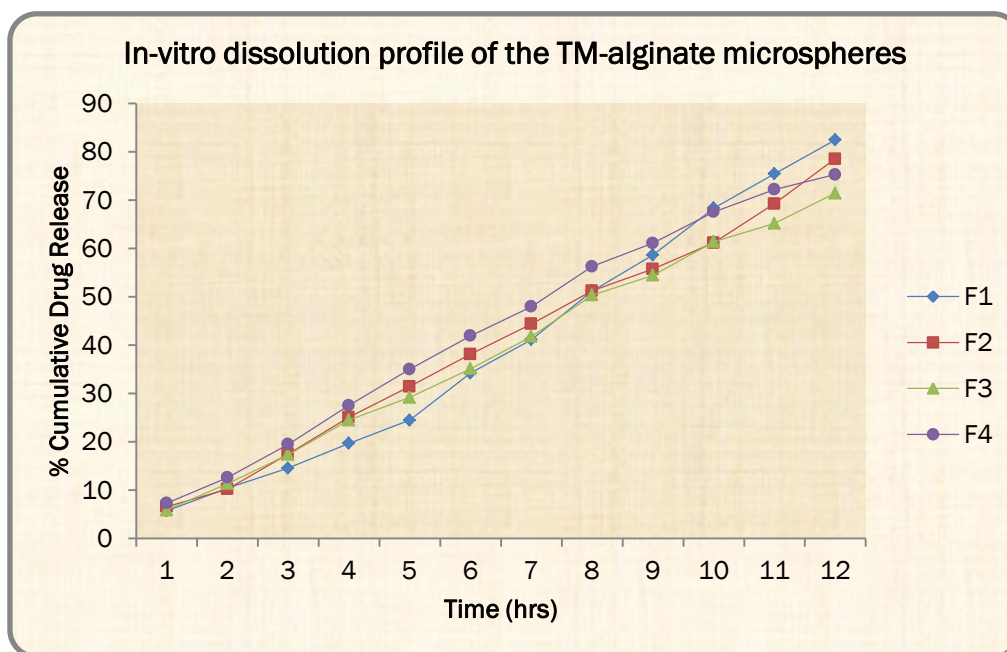
The *in-vitro* drug release study carried out of TM-alginate microspheres for first 2 hrs in 0.1N HCL and subsequently 10 hrs for pH 7.4 phosphate buffers to all F1-F4 formulated batches at 370 ± 0.50 C,

50 rpm. The release of Tegaserod from sodium alginate microsphere was  $10.42 \pm 0.24$  (F1),  $10.22 \pm 0.22$  (F2)  $11.36 \pm 0.24$  (F3) and  $12.62 \pm 0.37$  (F4) at 0.1N HCL after 2 hrs, while  $82.48 \pm 0.68$  (F1),  $78.53 \pm 0.25$  (F2),  $71.44 \pm 0.18$  (F3) and  $75.28 \pm 0.67$  (F4) at pH 7.4 phosphate buffer after 12 hrs. F1 shows the higher release of drug hence it was better formulation. These results of *in-vitro* drug release shown in Table No.3 and graphical representation shown in Fig.1.

The release of Tegaserod from sodium alginate microspheres at gastric pH was comparatively slower than intestinal pH. This was due to the shrinkage of alginate at acidic pH (as alginate is pH sensitive), which might slower the drug release from sodium alginate microspheres. The reason of the higher drug release was due to lower the concentration of cross linking agent.

**Table 3: *In-vitro* drug release profile of TM-alginate microspheres formulation**

Time (hrs)	Cumulative Drug Release (%) Mean $\pm$ SD (n=3)			
	F1	F2	F3	F4
1	$5.73 \pm 0.13$	$6.60 \pm 0.50$	$5.89 \pm 0.21$	$7.33 \pm 0.22$
2	$10.42 \pm 0.24$	$10.22 \pm 0.22$	$11.36 \pm 0.24$	$12.62 \pm 0.37$
3	$14.55 \pm 0.43$	$17.52 \pm 0.53$	$17.38 \pm 0.52$	$19.49 \pm 0.41$
4	$19.68 \pm 0.29$	$25.09 \pm 0.56$	$24.48 \pm 1.10$	$27.55 \pm 0.31$
5	$24.50 \pm 0.43$	$31.46 \pm 0.56$	$29.20 \pm 0.53$	$35.02 \pm 0.46$
6	$34.24 \pm 0.30$	$38.13 \pm 0.48$	$35.14 \pm 0.50$	$41.95 \pm 0.65$
7	$41.16 \pm 0.25$	$44.39 \pm 0.25$	$41.82 \pm 0.51$	$47.96 \pm 0.77$
8	$51.21 \pm 0.28$	$51.26 \pm 0.51$	$50.36 \pm 0.15$	$56.27 \pm 0.77$
9	$58.66 \pm 0.34$	$55.74 \pm 0.56$	$54.46 \pm 0.30$	$61.11 \pm 0.81$
10	$68.36 \pm 0.30$	$61.18 \pm 0.85$	$61.40 \pm 0.42$	$67.56 \pm 0.86$
11	$75.45 \pm 0.42$	$69.27 \pm 0.96$	$65.17 \pm 0.17$	$72.22 \pm 0.56$
12	$82.48 \pm 0.68$	$78.53 \pm 0.25$	$71.44 \pm 0.18$	$75.28 \pm 0.67$



**Figure 1: *In-vitro* drug release profile of TM-alginate microspheres formulation**

## CONCLUSION

In our present study we have successfully formulated sodium alginate microspheres containing tegaserod by crosslinking sodium alginate with calcium ions. From the data, the proposed TM-alginate microspheres could show better control of drug release for targeted drug delivery, which might improve patient compliance and reduce gastric side effects. Developed formulations are relatively economical and

easy to be manufactured by conventional pharmaceutical coating technique and are a promising candidate for specific drug delivery to the colonic region.

#### REFERENCES

1. Drug Bank of Tegaserod [cited 2009 Nov 27]. Available from: [http://www. DrugBank Showing Tegaserod \(DB01079\)](http://www.DrugBank.org).
2. Camilleri M. Review article: Tegaserod. *Aliment Pharmacol Ther.* 2001;15: 277-89.
3. Buchheit K, et al. The serotonin 5HT<sub>4</sub> receptor: Design of a new class of agonists and receptor map of the agonist recognition site. *J Med Chem.* 1995; 38: 2326-2330.
4. Grider JR, Foxx-Orenstein AE. A selective 5-HT<sub>4</sub>-receptor agonist stimulates transmitter release and activates the intestinal peristaltic reflex. *Gastroenterol.* 1996; 110: A1075.
5. Bregni C, Degrossi J, García R, Lamas MC, Firenstein R and D'aquino M. Alginate microspheres of *Bacillus subtilis*: Microesferas de alginato con *Bacillus subtilis*. *Ars Pharmaceutica.* 2000; 41(3): 245-248.
6. Martin A, Swarbrik G. 1991, *Physical pharmacy, physical chemical principal of pharmaceutical sci.* 3<sup>rd</sup> ed. Varghese publication house, 518-519, 630.
7. Cooper J, Gunn C. *Tutorial Pharmacy.* 6<sup>th</sup> ed. CBS Publishers and distributors, New Delhi; 2000. p. 225.
8. Banker SG, Anderson RN. Tablets. In, Lachman L, Liberman AH, Kanig LJ. , 1991, *Text book of the theory and practice of industrial pharmacy*, 3<sup>rd</sup> Ed<sup>n</sup>, Mumbai, Varghese Publication House; 317-324.
9. Aulton ME. 2002, *Pharmaceutics: The science of dosage form design*, 2<sup>nd</sup> Ed<sup>n</sup>, Churchill Livingstone, New York, 290-291.
10. Sudhamani T. Reddy KN, Kumar RV, Revathi R, Ganesan V. Preparation and evaluation of ethyl cellulose microspheres of ibuprofen for sustained drug delivery, *Int J Pharm Res Dev.* 2010;2:119-125.
11. United States of Pharmacopoeia, 2004, Washington, Board of Trustee Publisher, 1226.