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Probiotic Assisted Colon Specific Delivery of Ciprofloxacin

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Abstract

Matrix tablets of Ciprofloxacin were prepared using guar gum as the matrix forming polymer and incorporating Sporlac and Vivaflora probiotics in varying quantity to assist the targeted release of the drug in the colonic microflora. The angle of repose was found to be in the range of 26°32' to 28°52'. The bulk density value ranged from 0.360 to 0.45 g/cm^3 while the tapped density value ranged from 0.50 to 0.54 g/cm^3. The granules prepared in the present work were found to have the Carr's Index between 15.68 to 18.51 % and the Hausner's ratio was found to be ranging from 1.186 to 1.227. The thickness of all the tablets was found to be 6 mm while the hardness ranged from 5.2-5.3 kg/cm^2. The friability and weight variation were found to be less than 0.3% and the drug content in each tablet was found to be 98 - 99%. The matrix tablets were also investigated for the in vitro drug release in absence and presence of rat caecal content and it was found that the release of drug from the tablets was affected by the presence of probiotics. The rat caecal content was able to increase the drug release from all the formulation. The drug release was highest from the formulation MT4 which exhibited 98.78% and 99.84 %cumulative release in absence and presence of rat caecal content respectively.

Keywords: Ciprofloxacin, probiotic, colon, microflora, matrix tablet, release

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Introduction

Over the last two decades, the interest of pharmaceutical scientists in development of colon targeted drug delivery systems has tremendously increased (Goindi et al., 2011). Targeting drugs to the colon has been proven to be reasonably helpful in a variety of disorders, and the colon has been found to be a prospective site for local as well as systemic administration of drugs (Libo et al., 2002). Colonic delivery offers several preferential benefits as the site of drug delivery (Ahmad et al., 2011; Ahuja et al., 2010).

Polysaccharides arriving from small intestine are the main source of nourishment for microflora in the colon. The ability of colon microflora to degrade polysaccharides such as pectin, guar gum, chitosan, etc. forms the basis of formulation development for colon-specific drug delivery. The colonic microflora must be present in sufficient number to digest the carrier (like guar gum) to ensure the release of the drug at colon. Probiotic supplements (*Bifidobacteria* spp and Lactobacilli) are known to improve resistance to gut infections by inhibiting the growth of harmful bacteria, to reduce cholesterol levels, improve the immune response and produce vitamins.

Various probiotic-containing preparations like Sporlac (*Lactobacillus*) are available in the market in spore form. Incorporation of probiotics would provide the health benefits on one hand, while ensure drug release at colon in any condition on the other.

The objective of the present work was to combine the colon specific potential of probiotics and guar gum for designing matrix tablet based delivery system of ciprofloxacin for colon targeting.

Material and Methods

The preformulation studies of ciprofloxacin were performed for determination of its purity, solubility and compatibility with the polymer.

In vitro digestion of guar gum by probiotics (Ghosh et al., 2010)

Slurry of guar gum (1% w/v) was prepared by dispersing 2 g of guar gum in 200 mL distilled water. To the slurry was added the contents of one sachet of Sporlac (1 gm) and one capsule of Vivaflora separately and the mixture were incubated at 37°C in incubator for a period of 24 h. At various time interval the change in pH and viscosity was measured for each dispersion using calibrated pH meter (Labtronics) and Brookfield viscometer, respectively. Guar gum dispersion (1% w/v) was used as the control sample for the study.

Formulation of matrix tablet of Ciprofloxacin using guar gum

The matrix tablets of guar gum were prepared by wet granulation method (Ghosh et al., 2010). Lactose, guar gum, talc and magnesium stearate were sifted separately through sieve number 60 to obtain particles of uniform size. Weighed quantity of Ciprofloxacin was sifted through sieve number 100.

Ciprofloxacin, lactose and guar gum was mixed together and blended with addition of water (q.s) for granulation. The wet mass was passes through sieve number 14 and the granules were allowed to dry at 50°C in a tray drier for 2 h. The dried granules were passed through sieve number 16 to obtain a mixture of granules and fines. Magnesium stearate and talc were added to the granules and blended in a double cone blender for 5 min. The lubricated granules were compressed using tablet punching machine. Table 1 presents the composition of Ciprofloxacin tablets. The probiotics were added in two portions: half prior to granulation and the other half prior to final blending of the mixture.

Table 1 Composition of matrix tablets

	Quantity of each ingredient per tablet in mg				
Ingredient	**MT1**	**MT2**	**MT3**	**MT4**	**MT5**
Ciprofloxacin	100	100	100	100	100
Guar Gum	100	200	200	200	200
Lactose	285	185	105	85	65
Magnesium Stearate	5	5	5	5	5
Talc	10	10	10	10	10
Sporlac	-	-	40	50	60
Vivaflora	-	-	40	50	60
Weight of each tablet	**500**	**500**	**500**	**500**	**500**

Rheological Properties of the granules (Jalonya et al., 2018)

Angle of repose, Carr's Index, Bulk density, Tapped density and Hausner's ration were determined to assess the flow ability of the prepared granules.

Evaluation of the matrix tablets

The tablets were evaluated for thickness, weight variation, hardness, friability, drug content and *in vitro* release (in presence and absence of rat caecal content).

Drug content

Twenty tablets from each formulation were weighed to determine the average weight. These tablets were crushed in a mortar then the amount of powder equivalent to 25 mg of drug was transferred in 25 mL of 0.1N HCl. 10ml from this stock solution was withdrawn and diluted up to 100 mL with 0.1N HCl. 2 mL of this stock solution was pipetted out and diluted to 10 mL to obtain concentration of 20 µg/mL. Absorbance of the resulting solution was measured at 276 nm using UV spectrophotometer and the concentration was determined using the calibration curve. The drug content was calculated by applying the dilution factor.

In vitro release study

In vitro dissolution studies for all matrix tablet formulations were performed by using USP dissolution test apparatus (Apparatus 1, Basket type, 37°C) at 100 rpm using phosphate buffer pH7.4 as the dissolution medium (200 mL). The tablet was placed in the basket and the dissolution study was carried out for 12 h. At predetermined intervals, 1 mL of the dissolution media was pipetted out and its volume was made up to 10 mL using PBS pH 7.4. Absorbance of the solution was recorded at 276 nm

using UV visible spectrophotometer. 1 mL of fresh medium was added to the dissolution flask after each withdrawal.

In vitro release studies in presence of rat caecal content (Radhika et al., 2009)

To evaluate the susceptibility of guar gum being acted upon by the microbial flora of colon, the release study was carried out in presence rat caecal content.

Preparation of rat caecal content

All the animal works were approved by the Institutional Animal Ethical Committee (IAEC) of the institute. The caecal contents were obtained from Wistar rats weighing 150-200 g, maintained on a normal diet were administered with 4 mL of 1% w/v of dispersion of Guar gum in water for 7 consecutive days. Thirty minutes before starting drug release studies, 3 rats were killed by spinal traction, after which abdomens were opened, dissected, and immediately transferred to pH 6.8 phosphate buffer previously bubbled with CO_2. The caecal bags were then opened; their contents were individually weighed, homogenized, and then suspended in pH 6.8 phosphate buffer to give the desired concentration of 4% w/v of caecal content.

In vitro dissolution studies

The drug release for all the formulations was carried out with 200 mL of phosphate buffer pH 7.4 with rat caecal content (4%W/V). At predetermined intervals, 1 mL of the dissolution media was pipetted out and its volume was made up to 10 mL using 0.1N HCl. Absorbance of the solution was recorded at 276 nm using UV visible spectrophotometer. 1 mL of fresh medium was added to the dissolution flask after each withdrawal. The experiment was continued for 24 h.

Results and Discussion

Matrix tablets containing Ciprofloxacin were prepared by wet granulation method using guar gum as the colon targeting polymer and varying concentrations of probiotics Sporlac and Vivaflora to assist in colon targeting.

The ciprofloxacin sample was pale yellow in color, has no odor, displayed a melting point of 254-257°C. It was soluble in water, 0.1N HCl, and DMF and slightly soluble in methanol.

In vitro digestion of guar gum by probiotics

The *in vitro* digestion study of guar gum in presence of probiotics was performed to assess the effect of intestinal microbial flora of guar gum. The pH and viscosity of guar gum 1% w/v solution was used as the marker for degradation of guar gum. The results obtained are presented in Table 2.

Table 2 Effect of probiotics on pH and viscosity of guar gum

Time (h)	Guar Gum		Guar Gum + Sporlac		Guar Gum + Vivaflora	
	pH	Viscosity (cps)	pH	Viscosity (cps)	pH	Viscosity (cps)
0	6.97	2560	6.98	2560	6.97	2560
1	6.97	2560	6.97	2560	6.98	2560
2	6.98	2560	6.97	2560	6.97	2560

4	6.98	2560	6.98	2560	6.98	2560
6	6.98	2560	6.98	2560	6.98	2560
8	6.97	2560	6.97	2560	6.98	2560
10	6.99	2560	6.81	2380	6.73	2400
14	6.98	2560	5.93	2200	5.96	2250
18	6.97	2560	5.77	1800	5.73	1830
24	6.97	2560	5.40	670	5.52	740

Rheological properties of the prepared granules

Angle of repose of granules was determined by fixed funnel method. The angle of repose was found to be in the range of 26°32' to 28°52'. The apparent bulk density of the granules was measured with help of measuring cylinder. The bulk density value ranged from 0.360 to 0.45 g/cm^3. The tapped density was determined using tapping method. The tapped density value of various formulations of the granules was found to be in the range from 0.50 to 0.54 g/cm^3. The Carr's Index and Hausner's Ratio was calculated using the bulk and tapped density (Table 3).

The microbial flora of intestine in reported to cause degradation of guar gum thereby decreasing its viscosity due to depolymerization of the fatty acid chains of guar gum (Tomlin et al., 1986). The reduction in viscosity of guar gum was witnessed after 8 h of incubation with the microbial spores suggesting that the spores took almost 8 h to get activated into the vegetative form.

Table 3 Rheological parameters of various granular blends

Batch Code	Angle of repose	Bulk Density	Tapped Density	Hausner's Ratio	Carr's Index
MT1	26°32'	0.43	0.51	1.18605	15.6863
MT2	26°97'	0.42	0.5	1.19048	16
MT3	27°48'	0.44	0.54	1.22727	18.5185
MT4	28°52'	0.46	0.55	1.19565	16.3636
MT5	27°37'	0.43	0.52	1.2093	17.3077

Evaluation of matrix tablets of Ciprofloxacin

All the formulated matrix tablets were evaluated for weight variation, hardness, thickness, drug content and friability (Table 4).

Table 4 Characteristics of the matrix tablet formulations

Batch Code	Thickness (cm)	Hardness (Kg/cm^2)	Weight variation (%)	Friability (%)	Drug Content (%)
MT1	0.6	5.2	0.250	±0.33	98.3
MT2	0.6	5.2	0.250	±0.32	98.7
MT3	0.6	5.3	0.250	±0.31	98.7
MT4	0.6	5.2	0.250	±0.29	99.2
MT5	0.6	5.3	0.250	±0.28	98.5

In vitro drug release study

The *in vitro* release study was done for all the formulations to assess the time duration up to which the drug is released by the matrix tablets and to prove a sustained release from the matrix. The % cumulative release was plotted against time to obtain the release kinetics equation for the formulations (Figure 1).

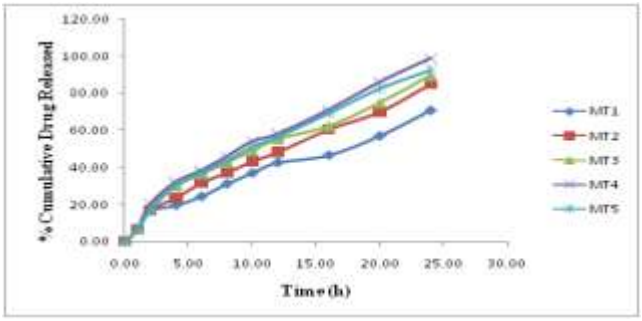

Figure 1 Cumulative percent of Ciprofloxacin released from matrix tablets in absence of rat caecal content

The release profile in presence of rat caecal content is represented in Figure 2.

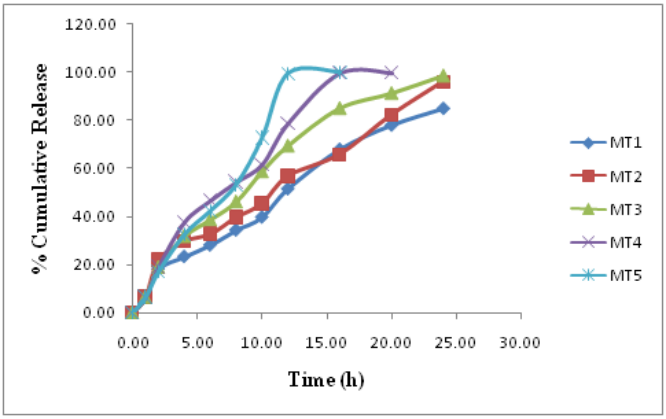

Figure 2 Cumulative percent of Ciprofloxacin released from matrix tablets in presence of rat caecal content

It was found that in absence of the caecal medium, the release of drug from formulation containing 20% guar gum (MT1) was only 70% while all other formulations released 80-99% drug over a period of 24 h. It was also observed that increasing the concentration of probiotic was able to increase the drug release from the formulations suggesting the assistive role of the probiotic in degradation of guar gum in the colonic microflora.

All the formulations exhibited increased drug release in presence of rat caecal content. It was found that the effect of the probiotic on degradation of guar gum was time dependent and an significant increase in drug release was observed after 8 h of dissolution study. This was in consonant with the digestion study which suggested that the microflora requires around 8 h to interact with guar gum thereby causing its depolymerization and enhancing the drug release.

Conclusion

In the present study, colon specific matrix tablets loaded with Ciprofloxacin were prepared using wet granulation method employing guar gum as the polymeric matrix and probiotics as the targeting aids. The results obtained showed that this methodology was able to produce colonic release of the majority of drug even in the absence of colonic microflora and also produced sustained release of drug from the formulations. Consequently, it can be concluded that the matrix tablets produced using the probiotic assisted procedure is an excellent delivery system that has good release behavior for actively releasing drug

in the colon, and therefore, this system would provide a safe and effective strategy for treatment of ulcers of the stomach or other diseases of the gut.

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References

Ahmed IS, Ayres JW (2011). Comparison of in vitro and in vivo performance of a colonic delivery system. International Journal of Pharmaceutics, 409 (1-2), 169-177

Ahuja V, Tandon RK (2010). Inflammatory bowel disease in the Asia–Pacific area: A comparison with developed countries and regional differences**.** Journal of Digestive Diseases, 11, 134-147

Ghosh PK, Gupta VB, Gondoliya B, Rathore MS. (2010). Probiotic-assisted colon-specific delivery of diclofenac sodium from guar gum matrix tablets: In vitro evaluation. Asian Journal of Pharmaceutics, 4(4): 173-178

Goindi S, Mann K, Aggarwal N (2011). Gastro-retentive floating beads of curcumin β-cyclodextrin complex to treat stomach tumors. Alternative Medicine Studies, 1 (e12), 49-53

Libo Y, Chu JS, Fix JA (2002). Colon-specific drug delivery: new approaches and in vitro/in vivo evaluation. International Journal of Pharmaceutics, 235, 1-15

Radhika PR, Pal TK, Sivakumar T (2009). Formulation and evaluation of sustained release matrix tablets of glipizide. Iranian Journal of Pharmaceutical Sciences, 5(4), 205-214

Tomlin J, Read NW, Edwards CA, Duerden BI (1986). The degradation of guar gum by a faecal incubation system. British Journal of Nutrition, 55, 481-486.

Jalonya R, Soni P, Malviya K, Omray LK (2018). Formulation and evaluation of bilayer tablets of diltiazem HCl. Journal of Pharmacology and Biomedicine, 2(3), 189-198