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Formulation and evaluation of colon specific drug delivery system

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Abstract

The aim of present research work was to develop colon targeted controlled drug delivery system of mesalamine using slight modification in CODES^{TM} technology which protects drug during its passage through the stomach and about first six meters of small intestine. Drug and polymers were directly compressed in tablet press. Mesalamine tablet were prepared in two groups A and B respectively. Group A consisted of combination of carbopol with HPMC and in group B only carbopol was present. Tablets were spray coated with lactulose USP solution, followed with acid-soluble coating material, Eudragit E-100 (10% w/w), water-soluble HPMC (10%w/w), enteric-coating material, Eudragit L-100 (10% w/w). Release of mesalamine from the coated tablets of different batches was studied in 0.1 N HCl (pH of 1.2) for first hour and (PBS) phosphate buffers solution (pH 6.8) for next four hour after that the same tablet was studied for release profile in PBS (pH 5.0). Release profile showed that drug release was possible only in PBS (pH 5.0) i.e. colonic medium. The above test result showed that the enteric and cationic coatings were able to bring the core tablet in colon.

Key words: CODES^{TM} technology, mesalamine, coating polymer, Eudragit, colon targeted

Introduction

The colon is not a suitable site for drug absorption as the small intestine, because the water content in the colon is much lower and the colonic surface area for drug absorption is narrow in comparison with of small intestine (Edwards 1997, Vyas and Khar 2002). However, the colon is a preferable site for the absorption of protein drugs, because the hydrolytic enzyme activities of the colon are lower than that of small intestine (Langguth et al. 1997, Rubinstein et al. 1997). Therefore, many researchers have focused on the colon as a potential delivery site for peptide and protein drugs. Many colon-specific drug delivery systems have been investigated, not only to treat the colonic diseases, but also to improve the bioavailability of such drugs (Saffran et al. 1986). Several approaches utilizing the GI-transit time of various formulation (Steed et al. 1997, Fukui et al. 2000) and change in pH (Gazzania et al. 1994), bacterial concentration (Yamaoka et al. 2000), and pressure (Muraoka et al. 1998) in the GI-tract have been reported to achieve colon-specific drug delivery. An ideal colon-specific drug delivery system should prevent drug

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release in the stomach and small intestine; it has an abrupt onset of drug release upon entry into the colon. This requires a triggering mechanism built in the delivery system responsive to the physiological changes particular to the colon. However, the physiological similarity between distal small intestine and proximal colon presents very limited options in selecting an appropriate drug release triggering mechanism.

Mesalamine also known as mesalazine (USAN) or 5-Aminosalicylic acid, is an anti-inflammatory drug used for treatment of inflammation of the digestive tract (Crohn's disease), mild to moderate ulcerative colitis and IBD (Inflammatory Bowel Disease).

In the present study, we have studied that slight modification in CODES^{TM} technology provide controlled release nature of mesalamine in colon. The therapeutic advantage of controlling and targeting mesalamine was to increase local tissue concentration of the drug hence lesser amount of drug is required to exert therapeutic effect. The aim of the present study was to establish a new concept, which exploits pH control by bacterial degradation of lactulose in order to develop slight modification in CODES ^{TM} and to reduce the dose of mesalamine tablet.

Material and Method

Material

Mesalamine was kindly provided as a gift sample by Sarex Overseas, India. Lactulose was prepared from the marketed preparation livoiluk manufactured by Panaecea Biotech, India. Hydroxypropyl methylcellulose procured from Central Drug House, India. Eudragit L-100 and Eudragit E-100 was provided as a gift sample by Evonik India private Ltd. Carbopol 934PNF was provided as a gift sample by Noveon Pvt. Ltd. Castor oil was procured from Central Drug House, India. Methanol and Potassium dihydrogen phosphate was procured from Qualigens Fine Chemicals, India. Sodium hydroxide pellets, n-Octanol and Hydrochloric acid were procured from Central Drug (P) Ltd., India.

Preparation method of mesalamine tablets

The core tablet was composed of drug and polymer. These were mixed according to prescribed ratio and mixture was directly compressed in tablet punching machine. Then spray coating of lactulose USP solution over the tablet was carried out. The resulted coated tablet was further coated with 10% (w/w) Eudragit E-100 in methanol as acid-soluble coating material. The amount of coating was 17.3 mg per tablet core. Second, the tablets were coated with water-soluble coating material, HPMC as an under coating layer. A coating solution was prepared by dissolving 10% (w/w) HPMC in water. The amount of coating was 3.5 mg per tablet core. Finally, the tablets were coated with 10% (w/w) Eudragit L100 and 2% (w/w) castor oil in methanol as enteric coating material. The amount of coating was 19.1 mg per tablet core (Table1) (Jamini and Reva 2007).

Characterization of mesalamine tablets (Lachman et al. 1991)

For determining the uniformity of weight, twenty tablets were weighed individually, calculating average weight and comparing the individual tablet weight to the average. The same procedure was repeated after acid soluble coating and enteric coating. Hardness of core tablet was

determined by Monsanto hardness tester. Friability of core tablet was determined by Roche friabilator. Thickness of the tablet was determined by screw gage. The determination of thickness was carried out after each coating. Diameter of the core tablet was determined with the help of Vernier Calliper's. The same instrument was used for the determination of acid soluble coated tablet and enteric coated tablet.

Table 1. Composition of mesalamine tablets

Ingredients (mg per tablet)	Formulation Codes									
	Group-A						Group-B			
	CT1	CT2	CT3	CT4	CT5	CT6	R1	R2	R3	R4
Mesalamine	250	250	250	250	250	250	250	250	250	250
Carbopol	500	500	750	250	-----	250	25	37.5	50	62.5
Sodium CMC	500	750	500	250	250	----	---	----	---	----
Lactulose	100	100	100	100	100	100	100	100	100	100
EudragitE-100	17.3	17.3	17.3	17.3	17.3	17.3	17.3	17.3	17.333	17.3
HPMC	3.5	3.5	3.5	3.5	3.5	3.5	3.5	3.5	3.5	3.5
EudragitL-100	15.9	15.9	15.9	15.9	15.9	15.9	15.9	15.9	15.9	15.9
Castor oil	3.2	3.2	3.2	3.2	3.2	3.2	3.2	3.2	3.2	3.2

Drug content (Jamini and Reva 2007).

The uniformity of drug content in each formulation was determined by triturating 20 tablets and powder equivalent to average weight was added to 100 mL of 5.0 pH PBS followed by stirring for 30 min. The solution was filtered through whattman filter paper, diluted suitably and absorbance of resultant solution was measured using double beam UV spectrophotometer (Systronic 2202, India) at maximum wavelength of 230 nm.

In vitro drug release

The drug release rate from mesalamine tablets were determined by using USP type II dissolution apparatus for batch A tablets and USP type I dissolution apparatus for batch B (Jinhe et al. 2002). Tablet was placed inside the dissolution apparatus. The dissolution test was performed in 900 mL 0.1 N HCl for first two h and then the tablet was placed in 900 mL PBS pH 6.8 for few hours. Finally the tablet was placed in 900 mL PBS pH 5.0. The dissolution was carried out at 50 rpm and 37°C temperature was maintained. At specified time intervals, 1 mL aliquots was withdrawn, filtered, diluted with the same medium and assayed at 230nm in case of 0.1N HCl, 214.4 nm in case of PBS pH 6.8 and 309.2 nm in case of PBS pH 5.0 for mesalamine using a UV double-beam spectrophotometer (Shimadzu UV-1700, Japan). Samples with drawn were replaced with equal volume of the same dissolution medium.

Statistical analysis

In this study, the results are given as mean ± SD. Student's t-test and one-way analysis of variance (ANOVA) were applied to find out the significant difference in drug release from different batches by using GRAPH PAD software programme considered statistically significant difference was at $p < 0.05$.

Kinetics of drug release (Khan and Jiabi 1998, Cosfa and Lobo 2001).

There are various types of equations which shows different release rate (Eqs. 1-5). The zero-order rate (Eq. 1) describes systems where drug release is independent of its concentration and this is applicable to the dosage forms like transdermal system, coated forms, osmotic system as well as matrix tablets with low soluble drugs. The first-order equation (Eq. 2) describes systems in which the release is dependent on its concentration (generally seen for water-soluble drugs in porous matrix). The Higuchi model describes the release of the drug from an insoluble matrix to be linearly related to the square root of time and is based on Fickian diffusion (Eq. 3). The Hixson-Crowell cube root law (Eq. 4) describes the release of drug from systems where it depends on the change in surface area and diameter of the particles or tablets with time and mainly applies in the case of systems that dissolute or erode over time. In order to authenticate the release model, dissolution data can further be analyzed by Peppas and Korsmeyer equation (Eq. 5).

$$Q_t = k_n t \qquad \rightarrow (1)$$

$$\ln Q_t = \ln Q_0 - k_1 t \qquad \rightarrow (2)$$

$$Q_t = k_H^{1/2} \qquad \rightarrow (3)$$

$$Q_0^{1/3} - Q_t^{1/3} = k_{HC}\, t \qquad \rightarrow (4)$$

$$M_t / M_\infty = k\, t^n \qquad \rightarrow (5)$$

Where Q_t is the amount of drug released at time t; Q_0 is the initial amount of the drug in the formulation; k_0, k_1, k_H, and k_{HC} are release rate constants for zero-order, first-order, Higuchi model and Hixson-Crowell rate equations. In Eq. 5, M_t is the amount of drug released at time t, and M_∞ is the amount released at time ∞; k is the kinetic constant, and n is the diffusion coefficient.

Stability studies (Jain et al. 2004)

Selected formulations were observed for morphology and percentage residual drug content for 30 days at the interval of 10 days. The storage temperatures were 4±1°C, 25±1°C, 50±1°C and relative humidity was 75±5 % in all cases.

Results and Discussion

The core tablets were prepared by direct compression technique as the polymers used in the core tablet had good binding property. Physicochemical properties are given Table 2. Lactulose coating along with Eudragit E-100 helped in achieving the site specificity. Hydroxypropyl methylcellulose coating was provided in order to avoid possible interactions between two oppositely charged polymers, Eudragit E-100 and Eudragit L-100. Eudragit L-100 provided enteric coating. Six formulations were prepared in group A (CT1 - CT6) and four formulations were prepared in group B (R1 - R4) simultaneously. The weight of core tablet of different batches of group A varies between 634mg to 1636 mg and 414 to 450 mg for different formulations of Group B. The variation in the weight was within the range of ±5% complying with the pharmacopoeal specifications. The hardness of core tablet varies with in the range of 8.0 to 9.1 kg/cm^2, indicating satisfactory mechanical strength. The friability was below 1% for

all formulations, which reveals good mechanical strength. The drug contents varied between 99.1% - 98.2% in different formulations showing content uniformity in the prepared tablet (Table 3 and 4).

Table 2. Physicochemical characterization of core tablets of mesalamine

Batch Code	Uniformity of weight[a]	Thickness[b]	Diameter[c]	Friability[d]	Hardness[e]
CT1	1248 ± 2.44	2.5 ± 0.0	14.0 ±0.05	0.65 ± 0.05	8.25 ± 0.180
CT2	1494 ± 3.74	3.15 ± 0.05	14.015 ±0.05	.60 ± 0.04	9.1 ± 0.1
CT3	1493 ± 2.44	3.15 ± 0.05	14.0 ± 0.05	.65 ± 0.05	9.1 ± 0.0
CT4	748 ± 2.44	1.35 ± 0.05	12.01 ±0.05	.75 ± 0.05	8.0 ± 0.0
CT5	493 ± 2.44	1.25 ±0.05	12.0 ± 0.5	.60 ± 0.04	8.1 ± 0.0
CT6	493 ± 2.44	1.25 ± 0.05	12.0 ± 0.5	.65 ± 0.03	8.25 ± 0.05
R 1	260 ± 11.66	1.05 ± 0.05	12.01 ± 0.05	.68 ±0.02	8.3 ± 0.0
R 2	285.4 ± 0.49	1.055 ± 0.05	12.01 ± 0.05	.69 ± 0.02	8.15 ± 0.05
R 3	293 ± 2.44	1.15 ± 0.05	12.0 ± 0.05	.55 ± 0.04	8.4 ± 0.0
R 4	310.4 ± 0.8	1.25 ± 0.05	12.01 ± 0.05	.59 ± 0.02	8.15 ± 0.05

a mean ± sd, n=20; b mean± sd, n=3; c mean ± sd, n=3; d mean± sd, n=10; e mean ±sd, n=3

Table 3. Physicochemical Characterization of acid soluble coated tablets of mesalamine

Batch Code	Uniformity of weight[a]	Thickness[b]	Diameter[b]
CT1	1363.0±2.44	2.80 ± 0.0	14.1 ± 0.05
CT2	1612.0±2.44	3.65 ± 0.05	14.115 ± 0.05
CT3	1613.0 ±2.44	3.45 ± 0.05	14.01 ± 0.05
CT4	865.4±0.49	1.85 ± 0.05	12.05 ± 0.05
CT5	614.0±2.00	1.70 ± 0.0	12.05 ± 0.05
CT6	614.0±2.00	1.80 ± 0.05	12.04 ± 0.05
R 1	388.0±2.44	1.50 ± 0.0	12.04 ± 0.05
R 2	402.0±2.44	1.55 ±0.5	12.05 ± 0.00
R 3	415.0±0.00	1.75 ± 0.05	12.04 ± 0.05
R 4	425.2±0.89	1.75 ± 0.05	12.05 ± 0.05

a mean ± sd, n=20; b mean± sd n=3

Table 4. Physicochemical characterization of coated tablets of mesalamine

Batch Code	Uniformity of weight[a]	Thickness[b]	% Drug content
CT1	1372.0±11.22	3.725 ± 0.083	98.5 ± 0.05
CT2	1631.0±2.44	4.25 ± 0.05	98.6 ± 0.05
CT3	1636.0±1.36	4.25 ± 0.05	97.5 ± 0.05
CT4	885.4±0.94	2.45 ± 0.05	98.9 ± 0.05
CT5	634.0±2.0	2.35 ±0.05	99.1 ± 0.05
CT6	634.0±2.0	2.25 ± 0.05	98.2 ± 0.05
R 1	414.0±2.0	2.05 ± 0.05	98.3 ± 0.05
R 2	423.2±2.64	2.055 ± 0.05	98.5 ± 0.05
R 3	434.2±2.13	2.15 ± 0.05	98.4 ± 0.05
R 4	450.8±0.96	2.25 ± 0.05	98.6 ± 0.05

a mean ± sd, n=20; b mean± sd n=3

In the CODES™ technology, the functionality of enteric coating was to maintain the integrity of the system in stomach, while the cationic acid-soluble coating intends to minimize the drug release in the small intestine. In this study, the dissolution media of pH 1.2 and 6.8 were used to simulate the pH conditions of stomach and intestine, while pH 5.0 buffer was used to represent the environment after the system entered the ascending colon, where lactulose was degraded into organic acids by colon bacteria. During a dissolution run, the duration of 4 hours in pH 6.8 buffers was chosen for simulating the average transit time of a solid dosage form in the small intestine. Release of Mesalamine from formulations of different batches of pH 1.2 for first h and in buffers of pH 6.8 for next four hour was found to be nil which shows that the enteric and cationic coatings appeared sufficient to prevent premature drug release in stomach and small intestine. But when the coated tablet was kept in PBS pH 5.0, the release was observed. The above test result shows that the CODES™ is able to bring the core tablet in colon (Katsuma et al. 2002, Yang et al. 2003, Yang 2008).

In case of group A the value of regression coefficient for batch CT1 and CT2 is 0.9978 indicating zero order release. Formulations CT1 and CT2 were achieving the objectives. It means that when drug, carbopol and sodium CMC were used in a combination of 1:2:2 or 1:2:3 then the drug release was at control rate while in case of group B the value of regression coefficient for formulation R3 was found to be 0.9935 indicating zero order release. It means that when the concentration of carbopol was 15% the formulation provided zero order release. From the above findings it was concluded that the concentration of polymer affect the release of drug from formulation. (Table 5)

Table 5. Kinetics- *in vitro* mesalamine release from mesalamine tablets

Batch code	Zero order		First order		Matrix		Peppas		Hixon Crowell	
	R	k	R	k	r	K	R	k	R	k
CT 1	0.9978	3.9179	0.8787	0.0844	0.8844	12.9921	0.9896	3.7065	0.9415	0.0202
CT 2	0.9978	3.9342	0.8800	0.0847	0.8870	13.0659	0.9902	3.7907	0.9429	-0.0203
CT 3	0.9964	3.8983	0.8721	0.0840	0.8698	12.8206	0.9937	2.8011	0.9359	-0.0202
CT 4	0.9154	4.6716	0.9012	0.1050	0.9709	16.4031	0.9857	14.5804	0.9660	-0.0246
CT 5	0.8241	15.4273	0.9190	0.3652	0.9729	35.3458	0.9347	43.0534	0.9377	-0.0863
CT 6	0.9577	15.8161	0.9523	0.3103	0.9874	33.1441	0.9875	29.3166	0.9797	-0.0799
R 1	0.9528	4.0961	0.9803	0.0673	0.9213	14.0591	0.9634	7.0526	0.9847	-0.0186
R 2	0.9809	3.7162	0.9679	0.0598	0.9089	12.5687	0.9766	5.7290	0.9855	-0.0167
R 3	0.9935	3.5198	0.9633	0.0555	0.9116	11.8585	0.9888	4.5119	0.9869	-0.0156
R 4	0.9877	3.0459	0.9824	0.0430	0.9021	10.2388	0.9692	3.2881	0.9913	-0.0126

After 30 days the drug content was found to be varied within the range of 98.5% to 98.1% for CT1 and 98.4% to 98.2 for R3 and no change in morphology was observed. It reveals good stability of the formulations (Table 6).

Comparison of release profile of different batches of group A

In case of CT1 and CT2, the amount of carbopol varies from 500-750 mg. The release of drug from CT-1 and CT-2 after 24 h was 93.37% and 93.74% respectively indicating that there is no significant difference between release profile of CT1 and CT2 (Fig. 1).

Table 6. Stability studies for batch CT1 and R3

Stability	4±1°C						25±1°C						50±1°C					
	Days						Days						Days					
	10		20		30		10		20		30		10		20		30	
	CT1	R3	CT1	R3	CT1	R3	CT1	R3	CT1	R3	CT1	R3	CT1	R3	CT1	R3	CT1	R3
% Residual drug content	98.5	98.4	98.4	98.3	98.4	98.4	98.5	98.4	98.5	98.4	98.4	98.3	98.2	98.3	98.2	98.3	98.1	98.2
Morphology	No Change		No Change		No Change		No Change		No Change		No Change		No Change		No Change		No Change	

In case of CT3 the amount of carbopol as well as sodium CMC was reduced to 250 mg and release of drug after 24th hour. was 91.70% showing that sodium CMC did not show significant effect on the release while the concentration of carbopol influence the release of drug from formulation. In batch CT5 and CT6 the release at 6thhour was 93.719% and 89.74% respectively and tablet disappeared in seventh hour depicts that individually carbopol and sodium CMC will not control the release rate of drug, but in combination they may control the release rate of drug. Probably integrity of tablet was maintained by sodium CMC by forming the gel covering over the core tablet and carbopol act as a rate controlling agent (Fig. 2 and 3).

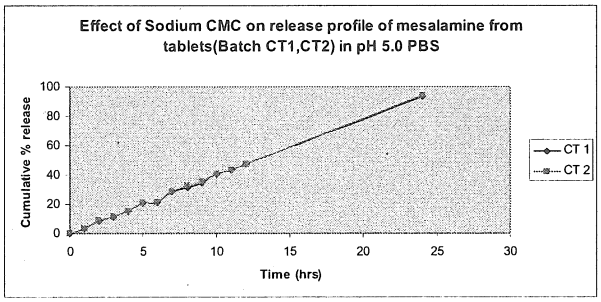

Figure 1. Effect of sodium CMC on release profile of mesalamine from tablets (Batch CT1,CT2) in pH 5.0 PBS

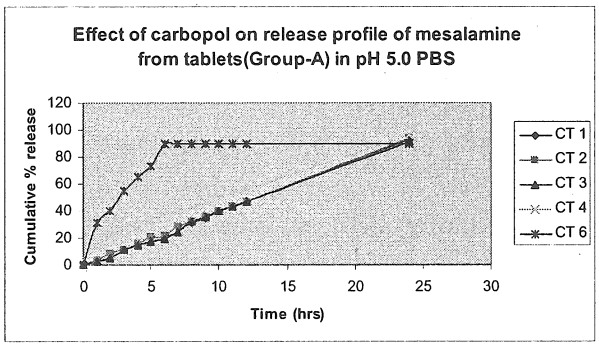

Figure 2. Effect of sodium CMC on release profile of mesalamine from tablets (Group-A) in pH 5.0 PBS

Comparison of release profile of different batches of group B

In case of R1, R2, R3, R4 as the amount of carbopol increases from 25mg to 62.5mg the release of drug decreases from 81.9 % to 68.0 % simultaneously.

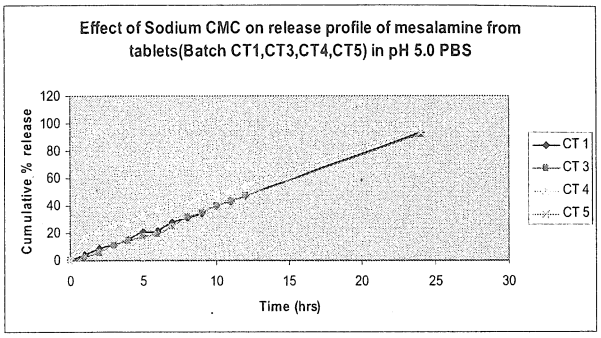

Figure 3: Effect of carbopol on release profile of mesalamine from tablets (batch-CT1, CT3, CT4, CT5) in pH 5.0 PBS

It depicts that as concentration of carbopol was increased release rate of drug from formulation was decreased. It is due to the increase in entrapment efficiency of polymer (Fig. 4).

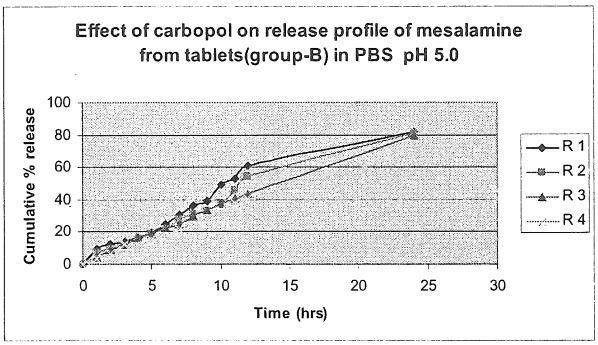

Figure 4. Effect of carbopol on release profile of mesalamine from tablets (Group-B) in pH 5.0 PBS

Conclusion

The objective of present study was to prepare and evaluate colon specific drug delivery system(s) for treatment of ulcerative colitis. The drug used was mesalamine. Mesalamine has been shown to be an effective agent for the treatment of ulcerative colitis. Effect of different formulation variables i.e. amount of carrier (polymers) were studied on release profile and other characteristics. The core tablet was prepared using different amount of carbopol and sodium carboxy methyl cellulose. The site specificity was achieved with the help of CODES^{TM} technology with some modification. *In vitro* drug studies were performed in 0.1N hydrochloric acid, 6.2 and 5.0 pH phosphate buffer. Different drug release kinetics models were applied for selected batches. Stability studies showed, there was not any significant change in residual drug content and morphology for the tablets.

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