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Combination of time-dependent and pH-dependent polymethacrylates as a single coating formulation for colonic delivery of indomethacin pellets

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Abstract

The objective of this study was to evaluate the combination of pH-dependent and time-dependent polymers as a single coating for design of colon delivery system of indomethacin pellets. Eudragit S100 and Eudragit L100 were used as pH-dependent polymers and Eudragit RS was used as a time-dependent polymer. A statistical full factorial design was used in order to optimize formulations. Factors studied in design were percent of Eudragit RS in combination with Eudragit S and L and coating level. Dissolution studies of pellets in the media with different pH (1.2, 6.5, 6.8 and 7.2) showed that drug release in colon could be controlled by addition of Eudragit RS to the pH-dependent polymers. The lag time prior to drug release was highly affected by coating level. With combination of two factors, i.e. the percent of Eudragit RS and coating level, the optimum formulation was found to be the one containing 20% Eudragit RS, 64% Eudragit S and 16% Eudragit L, and a coating level of 10%. This formulation was reproduced and tested in continuous condition of dissolution, and also separately at pH 7.5. The results of in vitro experiments indicate that the proposed combined time-dependent and pH-dependent polymethacrylate polymer coating may provide a colonic delivery system for indomethacin.

Keywords: Colonic drug delivery; pH-dependent; Sustained release; Indomethacin; Eudragit

1. Introduction

The various pharmaceutical approaches which have been used for targeting drugs to the colon are mainly based on pH-dependent, time-dependent and/or bacterially degradable polymers (Watts and Illum, 1997; Chourasia and Jain, 2003). Among these approaches, pH-dependent systems are simple, but the suitability of them for using alone as a colonic delivery system in different physiological or pathological conditions in gastrointestinal (GI) tract has been doubtful (Ashford et al., 1993). Therefore, it was decided to evaluate the pH-dependent system in combination with time-dependent system in order to alleviate the pH dependency of former system and to ensure drug release under different physiological conditions.

The use of pH-dependent and time-dependent polymers as coating materials for colonic delivery has been reported previously. In those studies sustained release and pH dependent polymers have been applied as a separate coating layers on top of each other and interesting results have been observed (Fukui et al., 2000; Gupta et al., 2001; Qi et al., 2003). There are various studies about the use of mixtures of these two kinds of polymers as a single coating system in design of some sustained release formulations (Fan et al., 2001; Munday, 2003; Zheng and McGinity, 2003). However, there is only one report in design of microspheres for colon delivery of 5-fluorouracil system (Lamprecht et al., 2003) and there is no report regarding the use of combination of these kinds of polymers in design of colonic drug delivery system using conventional coating process.

The objective of the present study was to optimize the formulation consisting of Eudragit RS as a sustained release polymer in combination with Eudragit S and Eudragit L as the pH-dependent polymers (in terms of their ratio and coating level) for coating of indomethacin pellets to achieve colonic delivery of this drug. Indomethacin was selected as a model drug because it has good indication for colonic delivery (Hull et al., 2003; Kapitanovic et al., 2006). The pellets as a multi-unit system were used because of the advantages of multiparticulate systems over

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single unit formulations such as reduced risk of local irritation, increasing bioavailability and non-gastric emptying dependency (Nykanen et al., 2001; Kramar et al., 2003). A statistical full factorial design was also used to optimize the formulation.

2. Materials

Indomethacin (Darupakhsh, Iran), Eudragit L100, Eudragit S100 and Eudragit RS100 (Rohm Pharma, GmbH, Germany), non-pareil seeds (NP Pharm, France), polyvinylpyrrolidone (PVP K30) (Fluka, Switzerland), triethyl citrate (Merck, Germany), talc (Merck, Germany) were obtained from the indicated sources.

3. Methods

3.1. Preparation of drug containing pellets

To prepare drug containing pellets, indomethacin was coated onto non-pareil beads (850–1180 μm) by fluidized bed apparatus (Wurster insert, Werner Glatt, Germany). Firstly, the indomethacin powder was passed through a 90 μm sieve. Then 30% (w/v) aqueous suspension of drug was prepared by dispersing drug in 7% (w/v) PVP K30 solution. The resulted suspension was passed through a 105 μm sieve. Then the suspension was sprayed onto non-pareils seeds using fluidized bed coater. Drug coating conditions are listed in Table 1. The suspension was stirred during coating process. After coating, the pellets were fluidized for extra 5 min and then were kept in an oven for 2 h at 40 °C.

3.2. Content uniformity

Accurately weighed 500 mg of indomethacin loaded pellets were ground by a mortar and pestle and then transferred to a 250 ml volumetric flask containing phosphate buffer pH 7.2. The flasks were shaken in a shaking water bath at 37 °C for 3 h. Followed by filtration the indomethacin absorbance in the solution was determined by spectrophotometry at 318 nm. All assays were carried out in triplicate and the mean value was reported.

3.3. Experimental design

A 3^2 full factorial design was used for optimization of the formulations. The independent variables were the ratio of Eudragit RS100 to Eudragit S100 and Eudragit L100 (in a fixed ratio of 4:1) (X_1), and coating level (X_2). The dependent variables (responses) were lag time (the time required for drug release up to 2%) at pH 6.8 (Y_1), lag time at pH 7.2 (Y_2) and percent of drug release in 10 h at pH 6.8 (Y_3). The independent and dependent variables and the used levels are summarized in Table 2. Also, Table 3 lists the obtained formulations.

Table 1
The conditions used for indomethacin layering and polymer coating onto non-pareils seeds

Process parameter	Indomethacin layering	Polymer coating
Inlet temperature (°C)	60–65	40–45
Outlet temperature (°C)	45–50	30–35
Nozzle diameter (mm)	1.0	1.0
Atomization pressure (bar)	2.0	2.0
Spray rate (g min^{-1})	15	10

Table 2
Independent and dependent variables and the levels used for factorial design

Factors (independent variables)	Levels used			Responses (dependent variables)
	1	0	−1	
X_1 = ratio of Eudragit RS to Eudragit S:Eudragit L (4:1)	40%	20%	0%	Y_1 = lag time at pH 6.8 (min)
				Y_2 = lag time at pH 7.2 (min)
X_2 = coating level	15%	10%	5%	Y_3 = percent drug released in 10 h at pH 6.8

3.4. Polymer coating

Ten percent (w/w) solutions of three polymers Eudragit S, L and RS were prepared in isopropyl alcohol:water (9:1) mixture. The ratios of the polymers were based on factorial design (Table 3). Triethyl citrate was added to the solution as plasticizer (10% (w/w) related to dry polymer). Talc also was added as glidant (5% (w/w) related to dry polymer). The resulted suspension was coated onto 200 g of indomethacin pellets using fluidized bed coater (Wurster insert, Werner Glatt, Germany). Polymer coating conditions are summarized in Table 1. Samples of coated pellets were removed from the apparatus when the coating loads reached 5, 10 and 15% (w/w). At each stage, the pellets were fluidized for extra 5 min and samples were kept in an oven for 2 h at 50 °C.

3.5. Dissolution experiments

Dissolution studies of coated pellets were carried out in a USP XXIII dissolution apparatus I (Pharmatest, PTWS, Germany) at a rotation speed of 100 rpm in 900 mL medium at 37 °C. Accurately weighed pellets containing 80 mg of indomethacin were

Table 3
Composition of experimental formulations

Formulation	Eudragit S (%)	Eudragit L (%)	Eudragit RS (%)	Coating level (%)
1	80	20	0	5
2	80	20	0	10
3	80	20	0	15
4	64	16	20	5
5	64	16	20	10
6	64	16	20	15
7	48	12	40	5
8	48	12	40	10
9	48	12	40	15

transferred to the dissolution medium. At predetermined intervals, the samples were taken from the vessels and passed through a multi-cell system on a UV spectrophotometer (Shimadzu, UV-1204, Japan) and returned to the vessel. Absorbance was read at 318 nm. All assays were repeated 6 times and the mean value was reported.

For simulating conditions of the GI tract, dissolution studies were carried out in media with pH 1.2 (HCl 0.1N), pH 6.5, pH 6.8 and pH 7.2 (phosphate buffer) and samples were tested separately at each medium. Dissolution time was 2 h for medium with pH 1.2 and 10 h for the other media.

The optimum formulation was also tested under continuous dissolution method considering 2, 1 and 2 h for the media with pH 1.2, 6.5 and 6.8, respectively, and the rest of experiment was carried out at the medium with pH 7.2.

3.6. Statistical analysis of data

The mathematical model of the effects of independent variables upon the dependent variables was based on following second order polynomial equation:

$$Y = b_0 + b_1X_1 + b_2X_2 + b_{11}X_1^2 + b_{22}X_2^2 + b_{12}X_1X_2 \qquad (1)$$

The modelling was performed using SPSS software (version 12.0) with a backward, stepwise linear regression technique. Significant terms ($p < 0.05$) were chosen for final equations. Response surface plots resulting from equations were drawn by Statgraphics 5.1 plus.

4. Results and discussion

Fig. 1 shows the results of dissolution test for drug-loaded pellets. Despite the low water solubility of indomethacin, coating of the drug onto non-pareil seeds decreased its dissolution time. This was due to increase in contact surface area between drug particles and dissolution medium when they were applied on non-pareil seeds. As shown in Fig. 1, dissolution profiles for drug-loaded pellets are the same at different media and there is no lag time prior to drug release.

Regarding pH and residence time of different parts of GI tract as the most important factors effective on release of drug from a colonic delivery system, combination of pH- and time-dependent systems were used in order to optimize colon drug delivery of indomethacin. According to our previous study (Akhgari et al., 2005) the best pH-dependent polymeric system based on Eudragit S and L (Eudragit S:Eudragit L in a 4:1 ratio) was used in combination with Eudragit RS in different ratios in order to design a colon delivery system of indomethacin. A statistical full factorial design was used in order to optimize formulations. Independent variables (Table 2) were the percent of Eudragit RS in combination with Eudragit S and L (in 4:1 constant ratio) (X_1) and coating level (X_2). The levels of independent variables were 0, 20 and 40% for X_1 and 5, 10 and 15% for X_2. The Composition of experimental formulations are listed in Table 3.

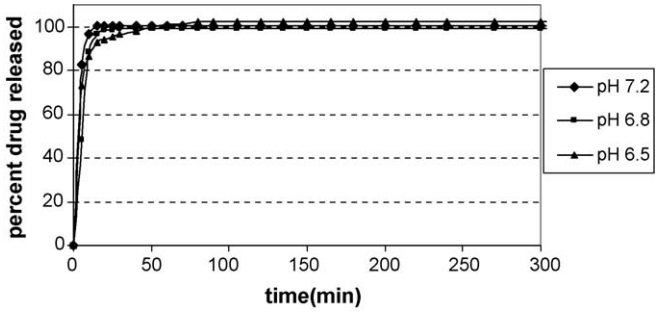

Fig. 1. Dissolution profile of uncoated drug pellets at pH 6.5, pH 6.8 and pH 7.2.

For coated pellets none of formulations released indomethacin at pH 1.2 (data are not shown). This was due to presence of the pH-dependent Eudragit S and Eudragit L in the coating layer and resistance of these polymers to dissolution at media with pH lower than 6.

In accordance with increasing pH of GI tract, dissolution studies were carried out at different media: medium with pH 6.5 as the first part of small intestine, medium with pH 6.8 as the middle part and medium with pH 7.2 as the terminal ileum. The dissolution profiles for coated pellets at different media are depicted in Fig. 2. As shown in Fig. 2, formulations coated with only Eudragit S and L (formulations 1–3) had a slow drug release at pH 6.5. The amount of drug release from these formulations was highly dependent on coating thickness. At pH 6.8, however, drug release was more rapid and at pH 7.2 these formulations released indomethacin without any significant lag time. These results show the high pH-dependent character of Eudragit S and Eudragit L coating. Both polymers have carboxylic groups that are able to ionize at neutral pH. This leads to polymer dissolution at higher pH. Therefore, drug release from formulations coated with only Eudragit S and L would be rapid after dissolution of polymeric coat. In fact, with this system, the delivery of the drug to the terminal part of small intestine could be assured; but drug release would almost be rapid at this place. This phenomenon could affect the effectiveness of such delivery system in situations which formulation stays longer at junction between small intestine and colon. Therefore, addition of a time-dependent polymer to this system could control drug release at pH 7.2 and as a result, the delivery of much more drug to the colon would be guaranteed.

Indomethacin release from formulations containing 20% Eudragit RS in coating layer with 5% coating level (formulation 4) was very slow at pH 6.5 (Fig. 2a). Other formulations containing Eudragit RS (formulations 5–9) did not release drug at this pH and therefore they were not shown in Fig. 2a. At pH 6.8 (Fig. 2b) and pH 7.2 (Fig. 2c) the amount of released drug decreased at all formulations by increasing coating thickness, because of longer drug diffusion pathways and more tortuosity at higher coating levels. Also at these pHs drug release from coated formulations containing Eudragit RS was slower than formulations without this polymer. Moreover, the structure of all pellets containing Eudragit RS remained intact by the end of dissolution tests. This effect was probably due to possible ionic interaction between cationic ammonia groups of Eudragit RS with carboxylic groups of Eudragit S and Eudragit L which pro-

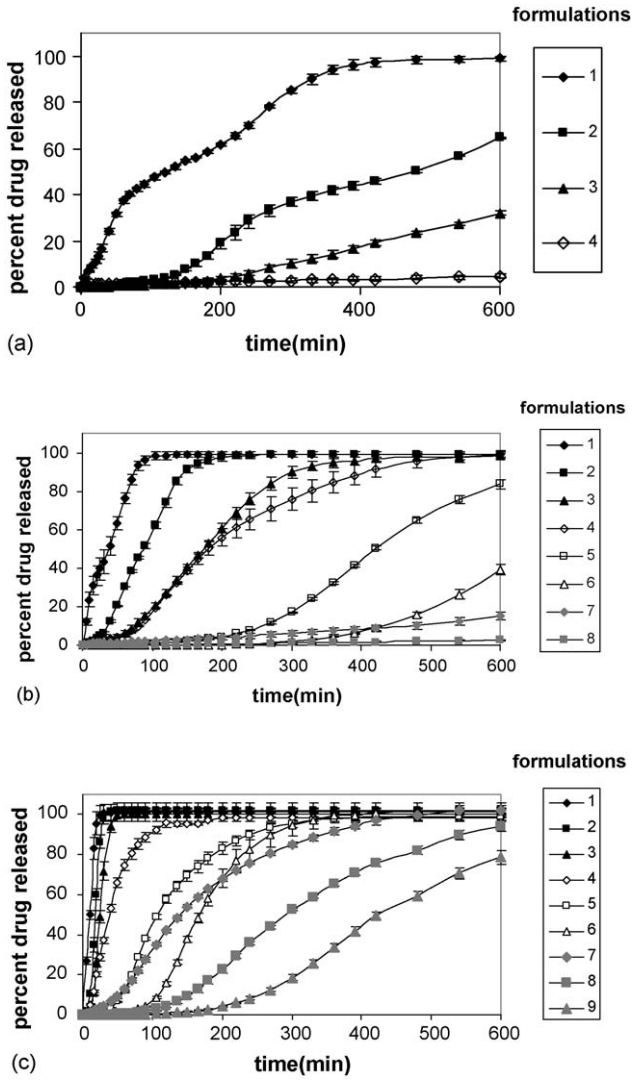

Fig. 2. Dissolution profiles of formulations 1–9 at media with different pH: (a) pH 6.5, (b) pH 6.8 and (c) pH 7.2. Formulations 1–3 contain no Eudragit RS; 4–6 contain 20% Eudragit RS and 7–9 contain 40% Eudragit RS with coating level of (♦) 5%, (■) 10% and (▲) 15%.

tects these anionic groups from rapid ionization and as a result retards dissolution of pH-dependent polymers. The electrostatic interaction between ionic groups of Eudragits and some ionic compounds has been reported earlier (Heun et al., 1998; Takka, 2003; Omari et al., 2004).

To optimize formulations by a factorial design, effective responses (dependent variables) should be defined in order to evaluate the effect of independent variables. Since residence time and pH of different parts of GI tract are important factors for design of a colonic delivery system, they should be considered when selecting effective responses. According to relative constant 3–4 h residence time of objects in the small intestine (Dressman et al., 1998) this period could be divided to 1, 2 and 1 h for the media with pH 6.5, pH 6.8 and pH 7.2 simulating first, middle and final parts of small intestine, respectively. As a result, the best colonic drug delivery system based on coating of polymethacrylates would be a system that could protect drug release in stomach and higher parts of small intestine, deliver it to terminal ileum and finally release drug in a steady state manner at this part. Therefore, lag time at pH 6.8 and lag time at pH 7.2 were considered as responses (Y_1 and Y_2, respectively). The optimum limitation for the response Y_1 was considered minimum 2 h. With this lag time, the optimum formulation would not release drug at first and middle parts of small intestine with pH 6.5 and pH 6.8 for 1 and 2 h, respectively. The best constraint for the response Y_2 was considered maximum 1 h. With this lag time, start of drug release at the end of small intestine with pH 7.2 would be assured. Also, regarding different physiological conditions and situations such as diseases, the pH of small intestine may change from normal values. If in some cases pH of the ileum does not reach to 7.0 (Ashford et al., 1993), the optimum colonic drug delivery system should release drug at pH 6.8 after an optimum lag time. Thus, percent of drug release in 10 h at pH 6.8 (Y_3) was considered as a response with a constraint of minimum 75%. A suitable formulation which could result in this response would be able to release the majority of drug in the colon despite its optimum 2 h lag time for drug release at pH 6.8. As a result, the formulation which produces all of mentioned responses could deliver drug to the colon at different physiological conditions.

Table 4
Analysis of variance (ANOVA) of dependent variables

Source of variation	Sum of squares	Degree of freedom	Mean square	*F*-ratio	*P*-value
Y_1[a]					
Regression	1206811	5	241362.180	87.446	0.000
Residuals	57962.731	21	2760.130		
Total	1264774	26			
Y_2[b]					
Regression	76287.319	3	25429.106	798.420	0.000
Residuals	732.533	23	31.849		
Total	77019.852	26			
Y_3[c]					
Regression	45296.272	2	22648.136	158.391	0.000
Residuals	3431.728	24	142.989		
Total	48728.000	26			

[a] $R^2 = 0.954$.
[b] $R^2 = 0.990$.
[c] $R^2 = 0.930$.

The second order polynomial equations resulted from SPSS software for all of the responses are given below:

$$Y_1 = -186.685 - 7.258X_1 + 43.283X_2 + 0.128X_1^2 - 1.969X_2^2 + 1.179X_1X_2 \quad (2)$$

$$Y_2 = 4.111 - 2.468X_1 + 0.02569X_1^2 + 0.366X_1X_2 \quad (3)$$

$$Y_3 = 113.56 - 0.058X_1^2 - 0.126X_2^2 \quad (4)$$

Analysis of variance (ANOVA) (Table 4) indicated that the assumed regression models were significant and valid for each considered response.

The three-dimensional response surface plots resulted from above equations were drawn to estimate the effects of the independent variables on each response (Fig. 3). As shown in Fig. 3,

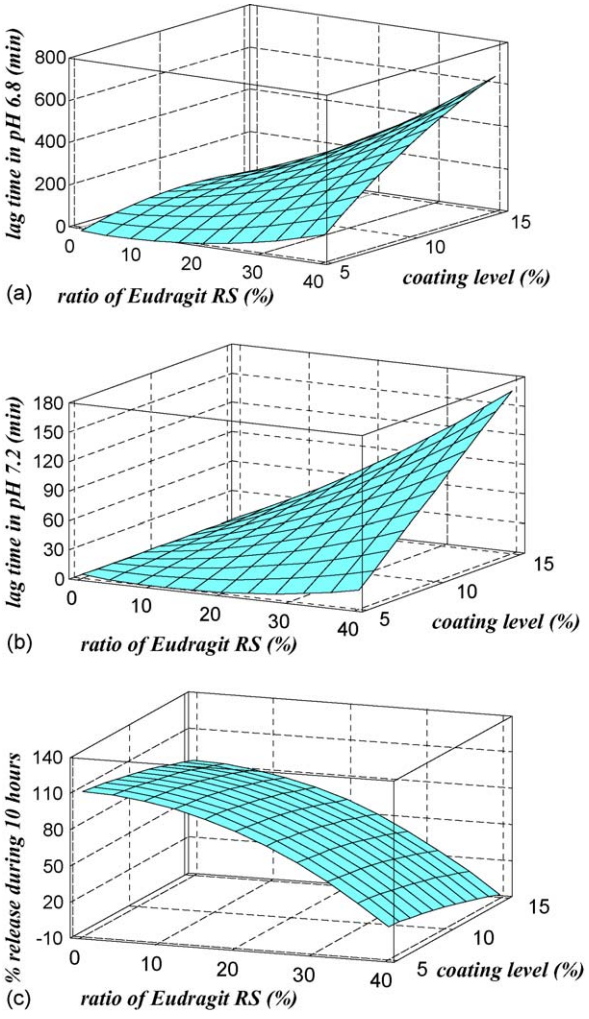

Fig. 3. Surface plots for different responses Y_1 (a), Y_2 (b) and Y_3 (c).

the effect of coating level (X_2) on lag time at pH 6.8 and pH 7.2 is more pronounced than the ratio of Eudragit RS (X_1) (Fig. 3a and b). In contrary, for the percent of drug release in 10 h at pH 6.8 (Y_3 response) the ratio of Eudragit RS is more important factor than coating thickness (Fig. 3c). As a matter of fact, Eudragit RS as a sustained release polymer can modify drug release after a lag time resulted from an appropriate coating level in different media. Indeed, with optimizing two factors coating level and ratio of Eudragit RS:S:L start of drug release at terminal ileum and then slow release of drug from delivery system during its movement to the colon can be assured.

Therefore, according to the best points of desired dependent variables, most suitable area for optimum formulation was found. The best conditions to optimize drug release corresponded to a formulation coated with Eudragit RS:Eudragit S:Eudragit L in ratio of 20:64:16 as coating system and a coating level of 10% (formulation 5). The optimum formulation was reproduced and tested under continuous dissolution test based on GI transit time. Fig. 4a shows the release profile of optimum formulation in media with different pH. This formulation did not release drug at pH 1.2 (for 2 h), pH 6.5 (for 1 h) and pH 6.8 (for 2 h). Pellets started to release their drug after about 45 min

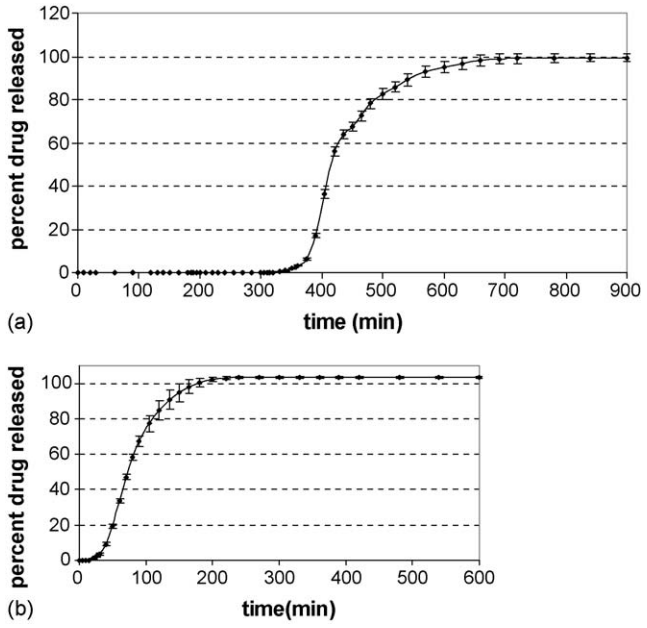

Fig. 4. Dissolution profile of optimum formulation: (a) under continuous dissolution in different media (0–120 min at pH 1.2, 120–180 min at pH 6.5, 180–300 min at pH 6.8 and the rest of experiment at pH 7.2), (b) at pH 7.5.

lag time at pH 7.2. In some cases, pH of terminal ileum may arise because of some diseases (Press et al., 1998); therefore, dissolution test of the optimum formulation was also carried out in medium with pH 7.5. Fig. 4b shows the drug release profile of optimum formulation in this medium. Drug release from pellets at this pH also started after about 30 min lag time and release rate was not rapid. Therefore, this formulation could protect drug from rapid release even if the pH of the end part of small intestine rises and a major load of indomethacin could be delivered to the colon.

5. Conclusion

The results of this study showed that combination of pH-dependent and time-dependent polymers is effective and useful for indomethacin colonic drug delivery. Addition of Eudragit RS as a sustained release polymer to Eudragit L and Eudragit S in the coating formulation could modify drug release after a suitable lag time in different media. The resulted optimum formulation was the one coated with Eudragit RS:Eudragit S:Eudragit L in the ratio of 20:64:16 with the coating level of 10%. The in vitro studies showed that this formulation has suitable release profile and it can be a good candidate for delivery of indomethacin to the colon at different physiological pH conditions.

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