

DEVELOPMENT OF A PH AND TIME CONTROLLED RELEASE COLON DELIVERY SYSTEM OBTAINED BY COMPRESSION-COATING AND FILM-COATING

DANA HALES, RAREȘ IULIU IOVANOV, MARCELA ACHIM, SORIN E. LEUCUȚA, DANA-MARIA MUNTEAN*, LAURIAN VLASE, IOAN TOMUȚĂ

"Iuliu Hațieganu" University of Medicine and Pharmacy Cluj-Napoca, Faculty of Pharmacy, Department of Pharmaceutical Technology and Biopharmacy, 41 Victor Babeș, 400012, Cluj-Napoca, România

*corresponding author: dana@tbs.ubbcluj.ro

Manuscript received: October 2014

Abstract

The purpose of this study was to develop a colonic delivery system with pH and time-dependent release. Ketoprofen cores, mini-tablet type, were prepared; then, they were coated by compression with different powder mixtures containing 25% - 35% hydroxypropylmethylcellulose (HPMC) K4M and K100M, and embedded by film coating with two types of polymers with pH-dependent solubility (Eudragit® FS 30D and Eudragit® L 30D-55). *In vitro* release studies were carried out in pH gradient: 1.2, 6.8, and 7.4. The polymers with pH-dependent solubility, located in the outer film layer, prevented the drug release in the gastric environment: Eudragit® L 30D-55 and Eudragit® FS 30D dissolved in the simulated intestinal fluid, at pH 6.8 and at pH 7.4, respectively. The polymer with time-dependent solubility, HPMC, situated in the subsequent layer of the film coating, allowed water penetration and subsequent swelling of the tablet core that gradually released the active drug. The obtained results showed that the ratio and properties of the two types of Eudragit polymers are capable to initiate the delivery of the active drug at the alkaline pH (after more than 5 h), the type and percentage of the HPMC polymer prolonging by swelling this process (until about 8 h). This study proved that compression-coated tablets, coated with enteric polymers are a promising technology for colon specific delivery of drugs.

Rezumat

Obiectivul acestui studiu a fost dezvoltarea unui sistem farmaceutic pentru transportul și cedarea colonică, dependente de pH și de timp. S-au preparat nuclee medicamentoase cu ketoprofen, de tip minicompriat; acestea au fost apoi acoperite prin comprimare cu amestecuri de pulberi conținând 25%-35% hidroxipropilmetilceluloză (HPMC) K4M și K100M, apoi filmate cu două tipuri de polimeri cu solubilitate dependentă de pH (Eudragit® FS 30D și Eudragit® L 30D-55). Studiile de cedare *in vitro* au fost realizate în gradient de pH: 1,2, 6,8, 7,4. Polimerii cu solubilitate dependentă de pH, situați în stratul filmului exterior, au prevenit eliberarea ketoprofenului în mediul gastric: Eudragit® L 30D-55 și Eudragit® FS 30D s-au dizolvat în fluidul intestinal simulat, la pH 6,8 și respectiv, la pH 7,4. Polimerul cu solubilitate dependentă de timp, HPMC, situat în stratul următor filmului enterosolubil, a permis pătrunderea apei și umflarea ulterioară a acestui strat care a determinat eliberarea treptată a substanței medicamentoase. Rezultatele obținute au arătat că procente și caracteristicile celor două tipuri de polimeri Eudragit sunt capabile să inițieze cedarea substanței medicamentoase la pH alcalin (după mai mult de 5 ore), tipul și procentul de polimer HPMC prelungind acest proces prin umflarea sa (până la aproximativ 8 ore). Acest studiu dovedește faptul că sistemul de tip comprimat acoperit cu polimeri enterosolubili constituie un sistem farmaceutic promițător pentru eliberarea substanțelor medicamentoase la nivelul colonului.

Keywords: compression-coated tablets, colonic delivery, pH-dependent release, time-dependent release

Introduction

The most common colonic dosage forms rely on physiological features of the colon and include pH controlled release systems, time controlled release systems, enzymatic controlled release systems and pressure controlled release systems [1, 13, 17, 24-26]. The pH controlled release systems for colonic drug delivery are based on the pH gradient along the gastrointestinal tract: the values increase from about 1 to 2 in the stomach, to 6.5 in the proximal small bowel, reaching a peak of about 7.5 in the terminal ileum, and finally decreasing to about 6.4

in the colon [19, 20]. Therefore, these types of systems use polymers or polymer blends that are insoluble at low pH values, but are soluble in the distal gastrointestinal tract, where the pH reaches values close to neutral [21]. However, a disadvantage of these systems is the possibility of releasing a considerable amount of active substance in the small intestine, before the system reaches the colon due to the oscillating pH between the terminal regions of the small bowel and the ascending colon [1, 20]. The time controlled release systems are based on the fact that the transit time in

the colon is extended (up to 20-30 h), even if it is variable [21]. A disadvantage of this type of system is the insensitivity to variations in transit time of upper gastrointestinal tract [12, 13, 20].

Therefore, in order to overcome the disadvantages of the two systems and to provide targeted colonic release, the release mechanisms and delivery systems, respectively, should be combined. The association of the two systems was studied before [2, 4, 10, 15], but the following drawbacks have been noticed in the majority of the studies: if the active pharmaceutical ingredient release starts after a convenient period of time (minimum 5 h), the release will be incomplete, whereas in the case of formulations that allow a complete release, the beginning of release occurs after 2 or 3 hours, in the small intestine, the system no longer having the characteristic of targeted colonic release.

This study aimed to develop a ketoprofen colonic drug delivery system, pH and time dependent, capable of releasing the entire amount of ketoprofen after a minimum period of five hours. Ketoprofen, an aryl propionic acid derivative used as a model drug in this study, is an anti-rheumatic drug with well-known anti-inflammatory, antipyretic and analgesic properties [29], but the aim of the study was to develop a system in which we can change at any time the active pharmaceutical ingredient (API) and its concentration. The colon drug delivery system was designed to contain a ketoprofen mini-tablet core, included by compression-coating in a layer based on a matrix-forming polymer (HPMC), the compression-coating tablet being coated with an enteric outer film based on Eudragit® FS 30D or Eudragit® L 30D-55, methacrylic polymers.

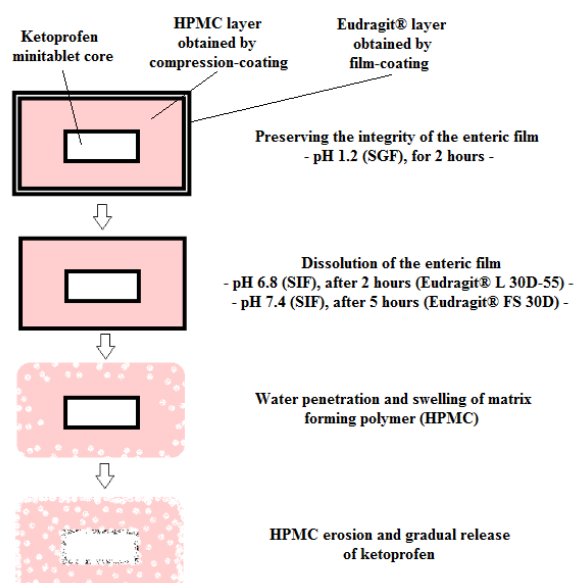


Figure 1.

The steps of the colon release system transformation along the gastro-intestinal tract

The enteric polymers were intended to delay ketoprofen release by being insoluble in the acid pH of the stomach, dissolving only after reaching the intestine, at pH 6.8 (in the case of Eudragit® L 30D-55) or at pH 7.4 (in the case of Eudragit® FS 30D [7, 8]. After enterosoluble polymers' dissolution, the hydrophilic polymer (HPMC) was intended to delay even more the release of ketoprofen: the hypromellose was supposed to progressively absorb water and swell for a lag time of at least 3 hours (the average transit of the small intestine); after that, ketoprofen release was supposed to occur gradually, as the pores of the HPMC matrix allowed dissolution and release of ketoprofen [14] (Figure 1). The influences of polymers' types and ratios on ketoprofen release from the colonic delivery system were evaluated in order to decide which characteristics are more desirable for such a system.

Materials and Methods

Apparatus

Tablet press (Korsch EK-0, Germany); analytical balance (Ohaus, Switzerland); tablet hardness test apparatus Dr. Schleuniger (Dr. Schleuniger, Germany); tablet disintegration test apparatus ZT 2 (Erweka, Germany); tablet friability test apparatus TA (Erweka, Germany); dissolution test apparatus I (rotating basket) PharmaTest PT-DT7 (PharmaTest, Germany); HPLC 1100 system (Agilent, Germany).

Materials

Ketoprofen (SIMS, Italy), was used as model drug. Lactose monohydrate for direct compression - Ludipress (BASF, Germany); microcrystalline cellulose (MCC) PH 102 (Pharma, Germany); hydroxypropylmethylcellulose (HPMC) - Methocel K100M and Methocel K4M (Colorcon, UK); silicon dioxide - Aerosil 200 (Degussa, Germany); magnesium stearate (Merck, Germany); Eudragit® FS 30D and Eudragit® L 30D-55 (Evonik, Germany). All other chemicals were of analytical grade.

Methods

Preparation of ketoprofen core mini-tablets. The mini-tablets were prepared using the direct compression technique. The formula is presented in Table I.

Table I
Qualitative and quantitative composition of minitab.lets

Constituent	mg / mini-tablet	%
Ketoprofen	1.00	5.00
Lactose monohydrate DC	13.00	65.00
MCC PH 102	5.70	28.50
Silicon dioxide	0.10	0.50
Magnesium stearate	0.20	1.00
Total	20.00	100.00

The compression was achieved using an eccentric tablet press (Korsch EK0, Germany), equipped with a 3 mm diameter concave punch, using a compression force which resulted in obtaining minitabets with the hardness of 24-30 N. The mass of the tablets was 20 mg and the concentration of the active pharmaceutical ingredient was 1 mg ketoprofen/tablet. The pharmacotechnical properties – uniformity of mass, disintegration time, friability, hardness, uniformity of content and dissolution – were evaluated for the prepared mini-tablets. The mini-tablets corresponded to 7th Edition of the European Pharmacopoeia's (Ph. Eur.) requirements [5, 6, 22, 23]. *Compression-coating with time-dependent layers.* Ketoprofen core mini-tablets were compression-

coated with coating powder mixtures containing a matrix-forming polymer. In this respect, six coating powder mixtures were prepared, the differences between formulations consisting in the type of the matrix-forming polymer (HPMC K100M and HPMC K4M) and its percentage: 25%, 30% and 35%, resulting six different formulations, F1-F6 (Table II). The same eccentric tablet press (Korsch EK0, Germany) was used, equipped with a 7 mm diameter concave punch. The mass of the tablets was 240 mg. The prepared ketoprofen compression-coated tablets were characterized in terms of uniformity of mass, friability and hardness and corresponded to Ph. Eur. requirements.

Table II

Qualitative and quantitative composition of powder mixtures for compression-coating

Constituent	Code					
	F1	F2	F3	F4	F5	F6
	Constituent percentage (%)					
Lactose monohydrate DC	71.50	66.50	61.50	71.50	66.50	61.50
HPMC K100M	25.00	30.00	35.00	-	-	-
HPMC K4M	-	-	-	25.00	30.00	35.00
Silicon dioxide	0.50	0.50	0.50	0.50	0.50	0.50
Magnesium stearate	1.00	1.00	1.00	1.00	1.00	1.00
Opadry II Orange	2.00	2.00	2.00	2.00	2.00	2.00
Total	100.00	100.00	100.00	100.00	100.00	100.00

Film-coating with pH-dependent layers. Each of the six formulations of compression-coated tablets was subsequently coated in a coating pan with two different enterosoluble polymers: Eudragit® FS 30D

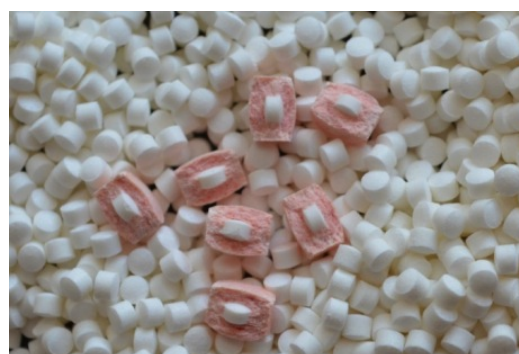
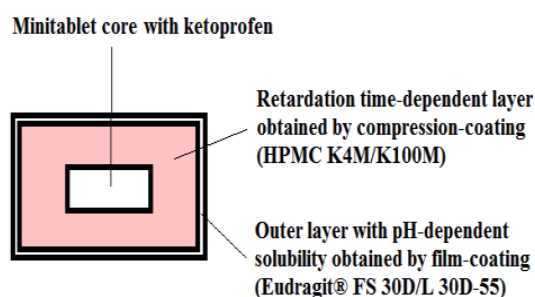
and Eudragit® L 30D-55, resulting in 12 formulations (Table III), the structure of the colonic delivery system being specified in Figure 2.

Table III

Formulations resulted after film-coating

Enterosoluble polymer	Code					
	F1	F2	F3	F4	F5	F6
Eudragit® FS 30D	F1 + EFS	F2 + EFS	F3 + EFS	F4 + EFS	F5 + EFS	F6 + EFS
Eudragit® L 30D-55	F1 + EL55	F2 + EL55	F3 + EL55	F4 + EL55	F5 + EL55	F6 + EL55

EFS = Eudragit® FS 30D; EL55 = Eudragit® L 30D-55

**Figure 2.**

The structure of the colonic delivery system

Table IV shows the coating formulas for the compression-coated tablets and Table V presents the working conditions of the coating process. After the coating, the tablets were maintained in a

stability chamber (Binder, Germany) for 15 h, at 40°C and 20% relative humidity in order to stabilize the polymer film (curing).

Table IV
Film coating formulas

Constituent		Constituent quantity (g)	
		Eudragit® FS 30D coating	Eudragit® L 30D-55 coating
		%	%
Core	Compression-coated tablets	90.5	87.5
Film layer	Eudragit® FS	9.0	-
	Eudragit® L 30D-55	-	9.0
	Triethyl citrate	0.5	1.0
	Talc	-	2.5

Table V
Working conditions

Parameter	Value
Batch size (g)	230
Nozzle diameter (mm)	0.8
Atomization pressure (atm)	2.6 - 2.8
Nozzle flow rate (g/min)	7
Inlet air temperature (°C)	35 - 40
Outlet air temperature (°C)	28 - 34
Pre-heating of the tablets (min)	10
Spraying duration (min)	40
Post-coating drying in coating pan (min)	10

In vitro drug release studies. The ability of the prepared tablets to keep the drug from being released in the stomach and small intestine and allow its release in the colon was assessed by performing drug release studies in fluids simulating the pH in the gastric and intestinal environment [4, 16, 19, 28, 30]. All buffers were prepared according to the US Pharmacopoeia [27]. For the dissolution test, the Pharma Test PT-DT7, apparatus I (rotating basket) (Pharma Test, Germany) was used at a rotation speed of 50 rpm and a temperature of 37.0 ± 0.5 °C. The release study was performed in 500 ml HCl 0.1M pH 1.2 for 2 hours, then the tablets were dried and weighed to determine the weight gain (maximum 5%), while changing the media inside the vessels. The HCl 0.1 M pH 1.2 was replaced with 500 mL phosphate buffer pH 6.8 (0.05 M) and the studies were continued for another 3 hours. Finally, 820 μ L solution of NaOH 10M was added to adjust the pH to 7.4 and the studies were continued until the end of the 24 h. Samples of 5 mL were withdrawn at 0.5, 1, 2, 3, 4, 5, 6, 8, 10, 12, 16, 20 and 24 hours' time intervals and replaced with an equal volume of fresh medium. The dissolution studies were performed in triplicate for each formulation.

The content of ketoprofen in the withdrawn samples was analysed by HPLC, using a validated quantification method according to the International conference on harmonisation (ICH) guidelines [11], by determining the fundamental validation parameters: linearity, accuracy and precision (data not shown). The chromatographic conditions, adapted from a previously published method [18], were: Agilent

HPLC system equipped with a UV/Vis detector; stationary phase: Phenomenex Gemini C18 column (100 mm x 3 mm, 3 μ m); mobile phase: acetonitrile:phosphoric acid solution 0.1% (60:40), isocratic elution; flow: 1ml/min; temperature: 22°C; wavelength: 260 nm (UV); pressure: 222 bars; retention time: 1.05 min; injection volume: 10 μ L.

Results and Discussion

Ketoprofen minitables' characterization. All characteristics found (Table VI) proved that the prepared mini-tablets are suitable to be used for the next step, compression-coated tablets preparation: the mass and drug content were within the limits established by Ph. Eur.; the disintegration time occurred in a few seconds, the release of ketoprofen occurring immediately after this step (> 90% after 15 min); and the friability and hardness were satisfactory enough to ensure mini-tablets' integrity during the compression-coating process.

Table VI
Pharmacotechnical properties of ketoprofen mini-tablets

Average mass and uniformity of mass (mg $\pm$ S.D.)	19.50 $\pm$ 0.51
Disintegration time (seconds $\pm$ S.D.)	30.00 $\pm$ 0.01
Friability (%)	0.215
Hardness (Newton $\pm$ S.D.)	25.9 $\pm$ 1.66
Uniformity of content (mg $\pm$ S.D.)	0.775 $\pm$ 0.05
Dissolution test (%)	> 90% in 15 min

S.D. – standard deviation

Table VII
Average mass, friability and hardness of compression-coated tablets

Code	Average mass (mg $\pm$ S.D.)	Friability (%)	Hardness (Newton $\pm$ S.D.)
F1	0.240 $\pm$ 0.002	0.139	405.20 $\pm$ 16.07
F2	0.239 $\pm$ 0.004	0.108	414.90 $\pm$ 20.77
F3	0.239 $\pm$ 0.002	0.139	407.50 $\pm$ 14.07
F4	0.239 $\pm$ 0.002	0.156	353.60 $\pm$ 26.91
F5	0.240 $\pm$ 0.002	0.124	361.80 $\pm$ 26.98
F6	0.239 $\pm$ 0.003	0.155	377.40 $\pm$ 18.48

S.D. – standard deviation

Compression-coated tablets' characterization. The obtained data for the six formulations concerning the uniformity of mass, friability and hardness are listed in Table VII. For all formulations, tested

parameters were within the limits established by Ph. Eur 7th edition.

The influence of HPMC's type on the release. The hydrophilic polymer's (HPMC) role was to delay drug release after the dissolution of enteric polymer for minimum 3 h (the average transit time of small

bowel). For all 12 formulations, ketoprofen release started after 8 h, therefore the HPMC matrix was able to prevent an immediate release and to allow ketoprofen release only after a certain lag time necessary for polymer swelling and erosion.

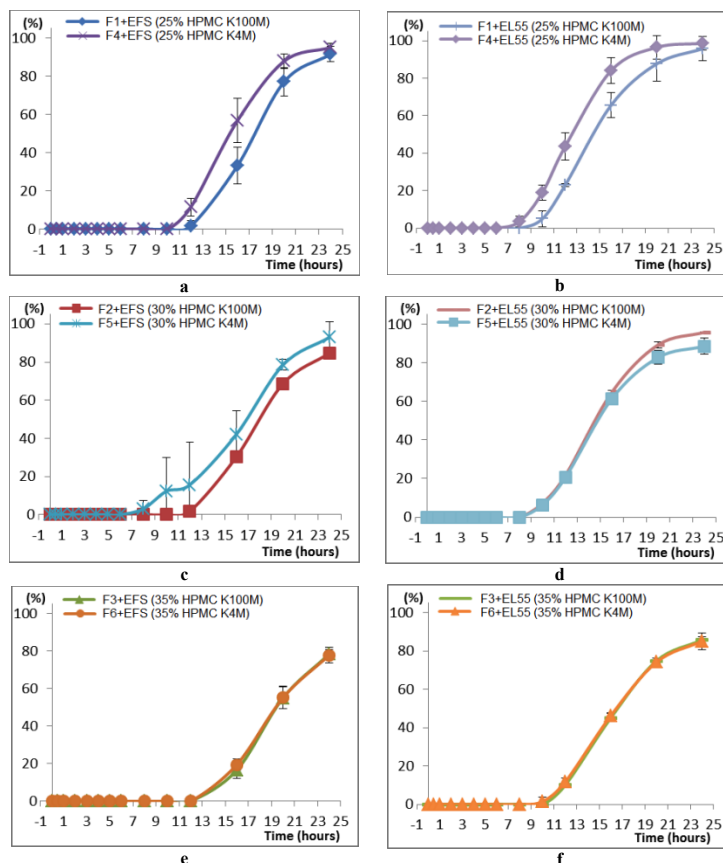


Figure 3.

The influence of HPMC type on ketoprofen release

Graphs show formulations prepared with: a - 25% HPMC and Eudragit® FS 30D; b - 25% HPMC and Eudragit® L 30D-55; c - 30% HPMC and Eudragit® FS 30D; d - 30% HPMC and Eudragit® L 30D-55; e - 35% HPMC and Eudragit® FS 30D; f - 35% HPMC and Eudragit® L 30D-55. Data are shown as mean $\pm$ S.D. (n = 3)

Regarding the type of polymer – HPMC K100M (F1, F2) compared to HPMC K4M (F4, F5) – the ketoprofen release was lower and the lag time was longer for the formulations prepared with HPMC K100M – F1, F2 – (Figure 3a-d) because a higher viscosity of the hydrophilic matrix decreased the release rate of ketoprofen. However, at high concentrations of HPMC (35%), the difference was small, F3 and F6 (Figure 3e-f) presenting similar release profiles. Therefore, it may be stated that at low concentrations of HPMC, there was an influence of HPMC type on release, but as the concentration increased, the type of HPMC was less important. Moreover, it can be observed that the variability of the release between the samples of the same formulation was higher for a low viscosity of HPMC (K4M) than for a greater viscosity (K100M), and also for a low amount of HPMC (25%) than for a

high amount (35%). This phenomenon is well-known, therefore, in the modified release systems it is recommended a minimum of 30% HPMC as matrix forming polymer [3].

The influence of HPMC concentration on the release. To evaluate HPMC's concentration on ketoprofen release, F1 and F4 formulations (25% HPMC) were compared to F2, F5 (30% HPMC) and F3, F6 (35% HPMC) (Figure 4) and the results showed that the percentage of ketoprofen released decreased with the increase of HPMC concentration, even if formulations F1 + EL55 and F2 + EL55 presented a similar release, despite the 5% difference in HPMC K100M amount. Also, even if it was less obvious, F1 + EFS and F2 + EFS were rather similar, which leads us to the conclusion that using a high concentration of HPMC along with a high viscosity type of HPMC decreased the release rate [3, 24].

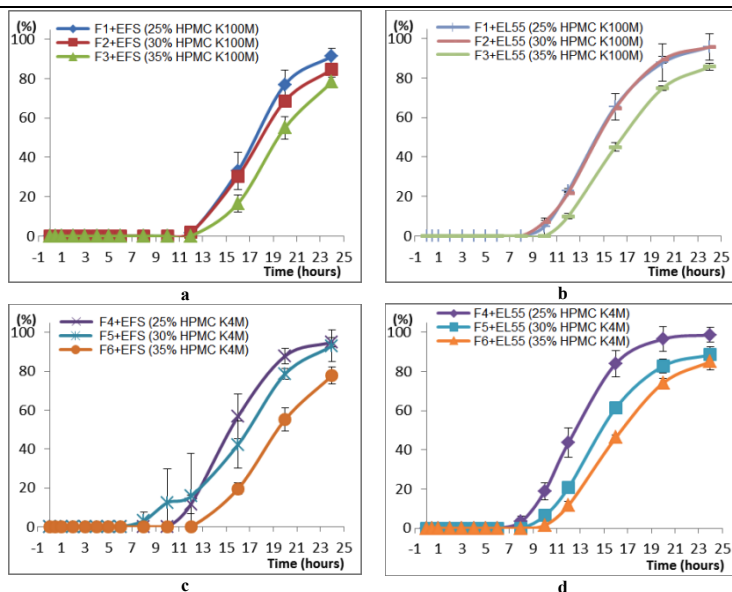


Figure 4.

The influence of HPMC's concentration on ketoprofen release

Graphs show formulations prepared with: a - HPMC K100M and Eudragit[®] FS 30D; b - HPMC K100M and Eudragit[®] L 30D-55; c - HPMC K4M and Eudragit[®] FS 30D; d - HPMC K4M and Eudragit[®] L 30D-55. Data are shown as mean $\pm$ S.D. (n = 3)

The explanation for the decrease in drug release by increasing HPMC concentration might be the fact that HPMC is a matrix-forming polymer that has the ability to swell in the aqueous medium of the gastrointestinal tract, forming a gel layer; the higher the concentration is, the more resistant the gel layer is to withstand erosion and subsequent diffusion of active pharmaceutical ingredient.

Hashem et al. consider that increasing the percentage of HPMC (from 10% to 20%) will increase the drug release due to a more porous structure, but increasing the percentage over a certain value (30%) will lead to reducing the drug release due to a greater viscosity and a reduction of the free water volume [9]. On the contrary, the results obtained by Yehia et al. [28] were similar to the ones obtained in this study, namely a proportional relationship between HPMC concentration and ketoprofen release. However, in comparison with the results presented in this paper, Yehia et al. obtained a maximum total release after 24 h of only 43.55% for 10% HPMC and the lowest percentage released in this study was $77.78 \pm 4.27\%$ for 35% HPMC K4M.

The influence of enteric polymers on release. Figure 5 presents the influence of the enteric polymers on ketoprofen release.

Eudragit[®] FS 30D and Eudragit[®] L 30D-55 are anionic methacrylic copolymers, Eudragit[®] FS 30D having the ratio of the free carboxyl groups to the ester groups of approximately 1:10 [7], while Eudragit[®] L 30D-55 ratio being of approximately 1:1 [8]. A higher percentage of ester groups than carboxylic groups requires a higher pH – a stronger alkaline solution – for hydrolysis and subsequent

release of drug, so Eudragit[®] FS 30D with a higher percentage of ester groups will require a higher pH to dissolve than Eudragit[®] L 30D-55.

Both Eudragit[®] FS 30D and Eudragit[®] L 30D-55 were able to prevent the drug release in the simulated gastric fluid: ketoprofen was not detected in the first two hours. Due to the fact that Eudragit[®] L 30D-55 is soluble at pH > 5.5 (in the upper gastrointestinal tract), ketoprofen could have been released after the polymer dissolution, but the HPMC layer delayed additionally the drug release, so that the first ketoprofen concentration was detected after 8 hours, for the formulation F4 + EL55. Eudragit[®] FS 30D, on the other hand, is soluble at pH > 7 (in the lower gastrointestinal tract), therefore prevented ketoprofen release until adjusting the pH from 6.8 to 7.4 and the first ketoprofen concentration was detected also after 8 hour, for F5 + EFS. The difference between the two enteric polymers is illustrated in Figure 5, where it can be observed that for all 12 formulations, the release rate is delayed for Eudragit[®] FS 30D, compared to Eudragit[®] L 30D-55.

Ibekwe et al. [10] showed in their research that drug release was very rapid for Eudragit[®] FS 30D, after 30 min in pH 7.4 phosphate buffer following a 2 h exposure to acid, the release being complete after approximately 1.5 h. Therefore, probably Eudragit[®] FS 30D alone would not be enough to target the colon, but Eudragit[®] FS 30D associated with HPMC would probably determine a sufficient delay to reach the colon.

As for choosing one of the two enteric polymers, the results showed that protecting compression-coated tablets in acidic environment (Eudragit[®] L

30D-55) would probably be enough in the case of this colonic delivery system, because the lag time needed for swelling and gelation of HPMC matrix would be sufficient to leave the small bowel without losing the drug. The protection in the simulated stomach and small intestine media (Eudragit® FS 30D) delayed too much the release, these formulations conducting to incomplete release of ketoprofen: in case of Eudragit® FS 30D associated with the highest percentage of HPMC (35% HPMC K100M and K4M), the maximum released

percentages were $78.37 \pm 2.43\%$ (F3 + EFS) and $77.78 \pm 4.27\%$ (F6 + EFS).

F4 formulation (25% HPMC K4M) coated with Eudragit® L 30D-55 (F4 + EL55) presented the highest release rate, as expected: $98.70 \pm 3.71\%$. As this formulation prevented ketoprofen release in the simulated gastric and small intestine environment, but also allowed achieving an almost 100% ketoprofen release, we may consider that this formulation fits best the desired characteristics.

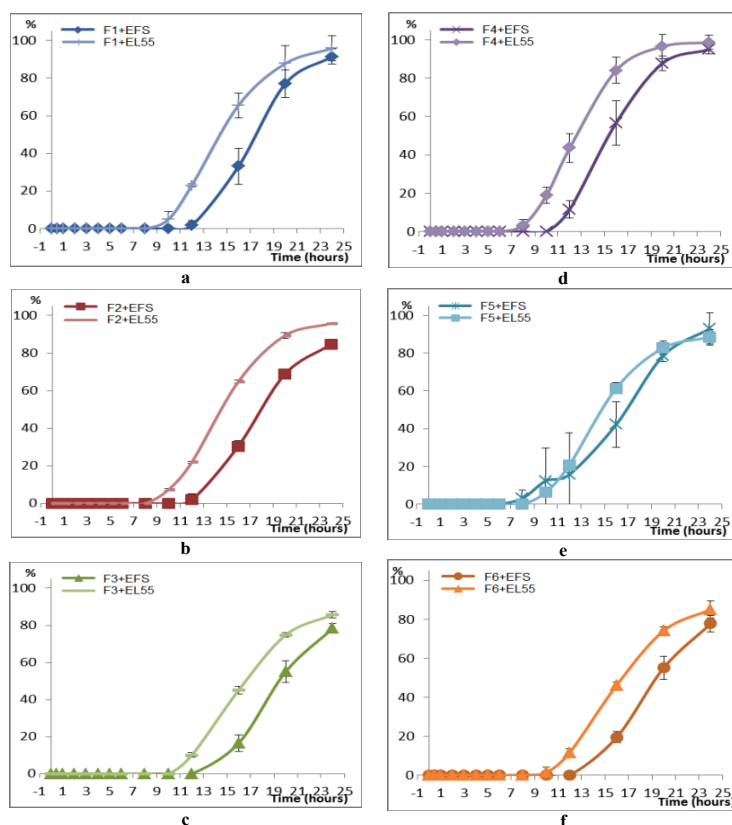


Figure 5.

The influence of the type of film-coating polymer on ketoprofen release

a – Ketoprofen released (%) from F1 formulations; b – Ketoprofen released (%) from F2 formulations; c – Ketoprofen released (%) from F3 formulations; d – Ketoprofen released (%) from F4 formulations; e – Ketoprofen released (%) from F5 formulations; f – Ketoprofen released (%) from F6 formulations. Data are shown as mean $\pm$ S.D. (n = 3)

Conclusions

This study aimed at obtaining compression-coated tablets capable of specific and complete delivery of active drugs to the colon, after a minimum period of time of 5 hours.

The influence of the type and concentration of HPMC in the compression-coating layer on the lag time and the influence of enterosoluble polymer type on the release of ketoprofen have been studied. The obtained results showed that the release of ketoprofen decreased with the increase of the viscosity of HPMC: the percentage of ketoprofen released was lower for the tablets prepared with HPMC K100M than for the tablets prepared with

HPMC K4M. The same was observed when HPMC concentration was increased: the percentage of ketoprofen released was higher for the tablets prepared with 25% HPMC compared to the ones prepared with HPMC 30% and 35%. Regarding the influence of enteric polymers, Eudragit® L 55 was found to cause a faster release of ketoprofen compared to Eudragit® FS, but in both cases the release started after a minimum period of time of 8 hours.

The formulation prepared with the type of HPMC with the lowest viscosity (HPMC K4M) and added in the smallest amount (25%), coated with the enteric polymer that dissolved first along the gastrointestinal tract, Eudragit® L 30D-55 - could be considered the best, because the percentage of

ketoprofen released was the highest (98.70 ± 3.71%) and the onset time of release (8 h) exceeded the initial objective of 5 h and was not too long, as for other formulations. Therefore, this formulation could meet the appropriate requirements to be a good candidate for *in vivo* studies.

References

- Anuj S., Jain K.A., Colon targeted drug delivery using different approaches. *Int. J. Pharm. Stu. Res.*, 2010; 1: 60-66.
- Auriemma G., Mencherini T., Russo P., Stigliani M., Aquino R.P., Del Gaudio P., Prilling for the development of multi-particulate colon drug delivery systems: Pectin vs. pectin–alginate beads. *Carbohydr. Polym.*, 2013; 92: 367-373.
- *** Colorcon. Using METHOCEL Cellulose Ethers for Controlled Release of Drugs in Hydrophilic Matrix Systems. Published July 2000.
- Crcarevska M.S., Dodov M.G., Goracinova K., Chitosan coated Ca–alginate microparticles loaded with budesonide for delivery to the inflamed colonic mucosa. *Eur. J. Pharm. Biopharm.*, 2008; 68: 565-578.
- *** European Pharmacopoeia (Ph.Eur.) 7.2th edition, 2012. General Chapter 2.9.6. Uniformity of Content of Single-Dose Preparations.
- *** European Pharmacopoeia (Ph.Eur.) 7.2th edition, 2012. General chapter 2.9.7. Friability of uncoated tablets.
- *** Evonik Industries. Technical Information, Eudragit® FS 30 D, Specification and Test Methods. December 2012.
- *** Evonik Industries. Technical Information, Eudragit® L 30 D-55, Specification and Test Methods. May 2014.
- Hashem F.M., Shaker D.S., Nasr M., Saad I.E., Ragaey R., Guar gum and hydroxy propyl methylcellulose compressed coated tablets for colonic drug delivery: *in vitro* and *in vivo* evaluation in healthy human volunteers. *Drug Discov. Ther.*, 2011; 5(2): 90-95.
- Ibekwe V.C., Fadda H.M., Parsons G.E., Basit A.W., A comparative *in vitro* assessment of the drug release performance of pH-responsive polymers for ileo-colonic delivery. *Int. J. Pharm.*, 2006; 308: 52-60.
- *** International conference on harmonisation of technical requirements for registration of pharmaceuticals for human use. ICH harmonised tripartite guideline. Validation of analytical procedures: text and methodology Q2(R1).
- Kshirsagar S.J., Bhalekar M.R., Umap R.R., *In vitro In vivo* comparison of two pH sensitive Eudragit polymers for colon specific drug delivery. *J. Pharm. Sci. & Res.*, 2009; 1(4): 61-70.
- Kushwaha P., Fareed S., Nanda S., Promising approaches to target drug delivery to colon. *Int. J. Ph. Sci.*, 2010; 2(3): 669-679.
- Lee V.H.L., Yang J.J., Oral Drug Delivery. In: Hillery AM, Lloyd AW, Swarbrick J. Drug Delivery and Targeting. For Pharmacists and Pharmaceutical Scientists. Taylor & Francis e-Library, London, 2005: 131-167.
- Maroni A., Del Curto M.D., Serratonì M., Zema L., Foppoli A., Feasibility, stability and release performance of a time-dependent insulin delivery system intended for oral colon release. *Eur. J. Pharm. Biopharm.*, 2009; 72: 246-251.
- Meissner Y., Ubrich N., El Ghazouani F., Maincent P., Lamprecht A., Low molecular weight heparin loaded pH-sensitive microparticles. *Int. J. Pharm.*, 2007; 335: 147-153.
- Metha T.J., Patel A.D., Patel M.R., Patel N.M., Need of colon specific drug delivery system: review on primary and novel approaches. *IJPRD*, 2011; 3(1): 134-153.
- Özlü C., Basan H., Şatana E., Ertaş N., Göger N.G., Quantitative determination of ketoprofen in gels and ampules by using flow-injection UV spectrophotometry and HPLC. *J. Pharm. Biomed. Anal.*, 2005; 39: 606-611.
- Philip A.K., Philip B., Colon Targeted Drug Delivery Systems: A Review on Primary and Novel Approaches. *Oman Med. J.*, 2010; 25: 70-78.
- Rajguru V.V., Gaikwad P.D., Bankar V.H., Pawar S.P., An overview on colonic drug delivery system. *Int. J. Pharm. Sci. Rev. Res.*, 2011; 6: 197-204.
- Singh B.N., Modified-Release Solid Formulations for Colonic Delivery. *Recent Pat. Drug Deliv. Formul.*, 2007; 1: 53-63.
- Stoltenberg I., Breitreutz J., Orally disintegrating mini-tablets (ODMTs) – A novel solid oral dosage form for paediatric use. *Eur. J. Pharm. Biopharm.*, 2011; 78: 462-469.
- Tissen C., Woertz K., Breitreutz J., Kleinebudde P., Development of mini-tablets with 1 mm and 2 mm diameter. *Int. J. Pharm.*, 2011; 416: 164-170.
- Sîrbu C., Tomuță I., Achim M., Rus L.L., Vonică L., Dinte E., Quantitative characterization of powder blends for tablets with indapamide by near-infrared spectroscopy and chemometry. *Farmacia*. 2014; 62(1): 48-57.
- Alecă C., Tomuță I., Dudaș D., Leucuța S., Mircioiu C., Optimization of metoprolol tartrate modified release matrix tablets formulation using Surelease as binder for granulation. *Farmacia*. 2013; 61(6): 1117-1130.
- Tomuță I., Vlase L., Popa A., Leucuța S.E., *In Vitro - in Vivo* evaluation of a novel drug delivery system for colonic targeting. *Farmacia*, 2010; 58(3): 368-377.
- *** US Pharmacopoeia (USP) 35th edition, 2012. Reagents. Solutions. Buffer solutions.
- Yehia S.A., Elshafeey A.H., Sayed I., Shehata A.H., Optimization of Budesonide Compression-Coated Tablets for Colonic Delivery. *AAPS PharmSciTech.*, 2009; 10(1): 147-157.
- Yiyun C., Tongwen X., Rongqinag F., Polyamidoamine dendrimers used as solubility enhancers of ketoprofen. *Eur. J. Med. Chem.*, 2005; 40: 1390-1393.
- Zhao X.L., Li K.X., Zhao X.F., Pang D.H., Chen D.W., Study on Colon-Specific 5-FU pH-Enzyme Di-Dependent Chitosan Microspheres. *Chem. Pharm. Bull.*, 2008; 56(7): 963-968.