BJPS

Brazilian Journal of
Pharmaceutical Sciences
vol. 52, n. 4, oct./dec., 2016
http://dx.doi.org/10.1590/S1984-82502016000400007

Article

Eudragit® FS 30 D polymeric films containing chondroitin sulfate as candidates for use in coating seeking modified delivery of drugs

Camila Borges dos Reis[1], Daniele Schaab Boff[1], Isabela Angeli de Lima[1], Leandro Freire dos Santos[2], Osvaldo Albuquerque Calvalcanti[3], Élcio José Bunhak[1*]

[1]Center of Medical and Pharmaceutical Sciences, Pharmacy course, Universidade Estadual do Oeste do Paraná, Cascavel, Brazil, [2]Department of Pharmacy, Universidade Estadual do Centro-Oeste do Paraná, Guarapuava, Brazil, [3]Department of Pharmacology and Therapeutics, Universidade Estadual de Maringá, Maringá, Brazil

Polymeric films associating different concentrations of Eudragit® FS 30 D (EFS) and chondroitin sulfate (CS) were produced by casting for the development of a new target-specific site material. Formed films kept a final polymer mass of 4% (w/v) in the following proportions: EFS 100:00 CS (control), EFS 95:05 CS, EFS 90:10 CS and EFS 80:20 CS. They were analyzed for physical and chemical characteristics using Fourier transform infrared spectroscopy (FTIR), scanning electron microscopy (SEM) and Raman spectroscopy. Furthermore, they were characterized by their water vapor permeability and degree of hydration at different conditions simulating the gastrointestinal tract. No chemical interactions were observed between CS and EFS, suggesting only a physical interaction between them in the different combinations tested. The results suggest that EFS and CS, when combined, may form films that are candidates for coating processes seeking a modified drug delivery, especially due to the synergism between pH dependency and specific biodegradability properties by the colonic microbiota. EFS 90:10 CS proved to be the most suitable for this purpose considering hydration and permeability characteristics of different associations analyzed.

Uniterms: Polymeric films/smelting process. Eudragit® FS 30 D/modified delivery. Chondroitin Sulfate. Polymeric coating.

INTRODUCTION

The delivery of oral drugs in the colonic region is valuable in the treatment of diseases that affect this intestinal segment. It is possible to achieve a high concentration of drug while reducing side effects. Among the diseases that could benefit from this strategy, Crohn's disease, colon carcinoma, ulcerative colitis and inflammatory bowel disease can be mentioned (Kumar, Chandra, Gautam, 2013). Furthermore, it is believed that the colonic region also provides an improved availability for drugs that are poorly absorbed in the upper gastrointestinal tract, such as peptides and proteins, which has created in recent years a considerable interest for the segmentation of drugs in this intestine portion (Pinto, 2010; Zimová *et al.*, 2012; Kumar, Chandra, Gautam, 2013). One of the existing systems for modified delivery of drugs in specific sites of the gastrointestinal tract (GIT), including the colon (pH 6.4 - 7.0), is based on the exploitation of the local pH. For this, many pH-dependent polymer coatings have been used (Pinto, 2010; Kumar, Chandra, Gautam, 2013; Bunhak, *et al.*, 2015). Eudragit® FS 30 D stands out as one of these polymers, whose dissolution occurs at a pH above 7.0, and thus can be used in a drug delivery system designed for the terminal portion of the GIT (Pinto, 2010; Zimová *et al.*, 2012). The main problem associated with this type of delivery system is GIT pH changes arising from pathophysiological conditions and individual variations, which may influence drug delivery (Bunhak *et al.*, 2015).

Faced with this problem, the use of natural non-cellulosic hydrophilic polysaccharides, particularly when considering the specific biodegradability by the anaerobic microbiota found in distal areas of the GIT, has aroused great interest among researchers (Souto-Maior *et al.*, 2010; Ramasamy *et al.*, 2012). In addition

***Correspondence:** E. J. Bunhak. Universidade Estadual do Oeste do Paraná – UNIOESTE. Rua Universitária, 2069 - 85819-110 – Cascavel - PR, Brazil. E-mail: elciob@gmail.com

to this favorable characteristic, natural polymers are found in abundance, they have low toxicity, low cost and flexibility to chemical modification, making them suitable for use in pharmaceutical coatings (Amrutkar, Gattani, 2009). Chondroitin sulfate is a natural polymer studied as a carrier of drugs in the research and development of new therapeutic systems designed for controlled and site-specific delivery (Bunhak *et al.*, 2015; Amrutkar, Gattani, 2009). It can be found in bones, cartilages and connective tissues. It is comprised of acetylgalactosamine and alternating residues of glucuronic acid (Huang *et al.*, 2010).

Because the GIT colonic region has a wide range of anaerobic microorganisms producing enzymes potentially capable of degrading polysaccharides, including chondroitin sulfate, the use of controlled drug delivery systems activated by the microbial flora in this region is considered as most promising, as they provide a greater specificity in the kinetics of drug delivery (Bunhak *et al.*, 2015; McConnell, Short, Basit, 2008; Rabito *et al.*, 2012).

From the combination of pH dependence and specific biodegradability mechanisms by the colonic microbiota, we seek through this study to characterize the behavior of films of Eudragit® FS 30 D associated with different chondroitin sulfate concentrations hopping the development of a new material for modified drug delivery. Therefore, tests were conducted to assess the degree of hydration and permeability of films in order to ascertain degradation perspectives by the colonic microbiota and drug release, respectively (Cavalcanti *et al.*, 2002; Bunhak *et al.*, 2007a; Melo *et al.*, 2010). Physical and chemical tests were performed involving Fourier transform infrared spectroscopy (FTIR), Scanning Electron Microscopy (SEM) and Raman spectroscopy with the purpose of observing the morphology of the formed films and the possible interactions among polymers in the associations.

MATERIAL AND METHODS

Material

Eudragit® FS 30 D (CAS: 26936-24-3, Evonik, Germany), chondroitin sulphate (Solabia-Brazil), simulated gastric fluid (SGF; pH 1.2) and simulated intestinal fluid (SIF; pH 6.8) prepared according to the 23st ed. Pharmacopoeia of the United States of America (USP 23), Tween® 80 (CAS: 9005-65-6, Aldrich, United States), glyceryl monostearate (CAS: 123-94-4, Fluka, Switzerland), sodium hydroxide (CAS: 1310-73-2, Vetec, Brazil), glacial acetic acid (CAS: 758-12-3, Sinth, Brazil), potassium chloride and potassium phthalate monobasic (CAS: 877-24-7, Nuclear, Brazil). All reactants possess analytical grade and were used without further purification.

METHODS

Preparation of the films

The films were obtained using the conventional method for thermoplastic and thermosetting polymers known as "casting process" (Bunhak *et al.*, 2007b; Braz, Hechenleitner, Cavalcanti, 2007; Santos *et al.*, 2013; Souto-Maior *et al.*, 2010). For the preparation of the films, an aqueous dispersion of the synthetic polymer Eudragit® FS 30 D (EFS) was used associated with the polysaccharide chondroitin sulfate (CS) in the proportions (w/w)%: 100:0 (control), 95:05, 90:10 and 80:20, while maintaining a final polymer concentration at 4% (w/v). EFS was initially stirred with glyceryl monostearate (previously prepared in emulsion at 5% polysorbate 80) at 50 °C for 30 minutes. Subsequently, the CS dissolved in distilled water was slowly added to the dispersion containing EFS, and the mixture was subjected to magnetic stirring. All dispersions were made using a vacuum pump during the homogenization process to avoid air incorporation and the undesirable formation of bubbles in the polymeric mixture. The final dispersion was poured into Teflon® molds in 10 mL samples, which were kept in an incubator at 50 ± 2 °C for 24 hours.

Fourier transform infrared spectroscopy (FTIR)

In order to characterize the synthetic polymer and the polysaccharide and to establish the possible interactions between them in the associations, a spectroscopic analysis in the infrared region was carried out using a Vertex 70v Bruker, performing a reading in the spectral range 4,000-400 cm^{-1} with a resolution of 4 cm^{-1} and 100 scans.

Raman spectroscopy

Raman spectroscopy has been widely used in the characterization of drug delivery systems. By this technique, among other applications, it is possible to understand drug-excipient and drug-polymer interactions in the formulation, characterize the primary coating material and verify the homogeneity of distribution of the active ingredient in a formulated compound pill, using for this purpose Raman chemical imaging (Paudel, Raijada, Rantanen, 2015; Smith *et al.*, 2015). The assay was performed with a micro-Raman SENTERRA spectrometer (Bruker Corporation, Germany). Polymer films were

analyzed at a wavelength of 1,064 nm for the FT-Raman spectroscopy, covering regions between 4,000 and 400 cm^{-1}, and 785 nm for mapping.

Determination of the film swelling index

The determination of the swelling index was made in order to know the degree of hydration of films formed upon contact with the simulation of physiological fluids of the gastrointestinal tract. For this, the films were cut using surgical scissors in 1 cm^2 samples and placed in Petri dishes. The total losses of residual moisture from different film samples were made in an oven at 40 °C for approximately 24 hours. The dried films were weighed and immersed in a medium containing SGF (pH 1.2) and SIF (pH 6.8) at 37±0.5 °C for certain time periods (1, 3, 5, 7, 10, 30, 60, 90, 120 and 180 min). After the pre-determined intervals for film hydration, they were carefully removed from the medium in which they were immersed and the excess of water was removed. Then, they underwent a new weighing. The swelling index was given by the Eq. (1), where *Sw* is the Swelling index, *Ws* and *Wd* is the weight of Swollen and Dry film, respectively (Souto-Maior *et al.,* 2010).

$$S_W\% = \left(\frac{W_s}{W_d} - 1 \right) \times 100 \qquad \text{Eq. (1)}$$

Water vapor transmission

The analysis of water vapor transmission (WVT) was performed according to the method "B" of the ASTM (*American Society for Testing and Materials*) designated as E 96-66. The assay was performed in triplicate for each association: EFS 100:00 CS, EFS 95:05 CS, EFS 90:10 CS and EFS 80:20 CS. Films with a 10 cm^2 area were fixed in permeability cups (Braive Instruments, Liege, Belgium) containing 10 mL of distilled water. This set was weighed and then placed in a desiccator containing dried silica gel, which was kept at room temperature and humidity. After the pre-established times (24, 48, 72, 96 and 120 hours), the set was again weighed and the silica gel was replaced at each interval by a new dehydrated silica gel. The readings of the weight lost by the cups in the respective intervals were recorded and used to calculate water vapor transmission rate according to Eq. (2), where *g* is lost weight, *t* is time in hours during which the weight loss was observed, and *a* represents the area of the film (Souto-Maior *et al.*, 2008).

$$WVT = \frac{g \times 24}{t \times a} \qquad \text{Eq. (2)}$$

Scanning electron microscopy

The morphological characterization of polymeric films was performed using scanning electron microscopy (SEM). Samples of isolated films in the proportions 100:00 and 90:10, both dried and after 60 minutes of immersion in SGF (pH 1.2) and SIF (pH 6.8), were frozen at -18 °C with liquid nitrogen and lyophilized at -55 °C/6 h in a Liotop® L101 lyophilizer (Liobras®). Subsequently, the samples were metallized with gold in a Shimadzu IC-50 equipment and micrograph images of the fractured surfaces of polymer films were obtained with a scanning electron microscope Shimadzu® SS-550 operated at 10 keV (Rabito *et al.*, 2012).

RESULTS AND DISCUSSION

FTIR

EFS 100:00 CS composition films analyzed by FTIR revealed EFS characteristic bands at 1726 cm^{-1} corresponding to the C=O vibrations of carboxylic groups esterified, at 1164 cm^{-1} and 1198 cm^{-1} due to ester vibrations and in the range between 2500-3500 cm^{-1} related to the absorption of OH^- groups. The other combinations containing 5%, 10% and 20% of CS were similar to the control. In the upper spectral region, at 3000 cm^{-1}, there was a greater intensity of bands proportional to the addition of CS, which may be associated with the OH^- group common to both CS and EFS (Evonik, 2015; Amrutkar, Gattani, 2009). When comparing the spectrum of the control film and spectra of films associated with CS, the non-appearance of new bands can be verified, except CS absorption bands. Rabito *et al*. (2012) observed similar results working with Eudragit® FS 30 D and arabinoxylane films: likewise, the appearance of new bands was not noticed. Thus, there were only physical interactions between the compounds (Rabito *et al.*, 2012). FTIR data corroborate Raman spectroscopy data. FTIR spectra of control films and other associations can be seen in Figure 1.

Raman spectroscopy

Chemical images of polymer films provided by Raman spectroscopy showed the homogeneous incorporation of the natural CS polysaccharide to the synthetic polymer EFS. As observed in Figure 2, the increase in the CS concentration resulted in a higher intensity of pink-reddish tint, suggesting a greater incorporation with the EFS-based polymer.

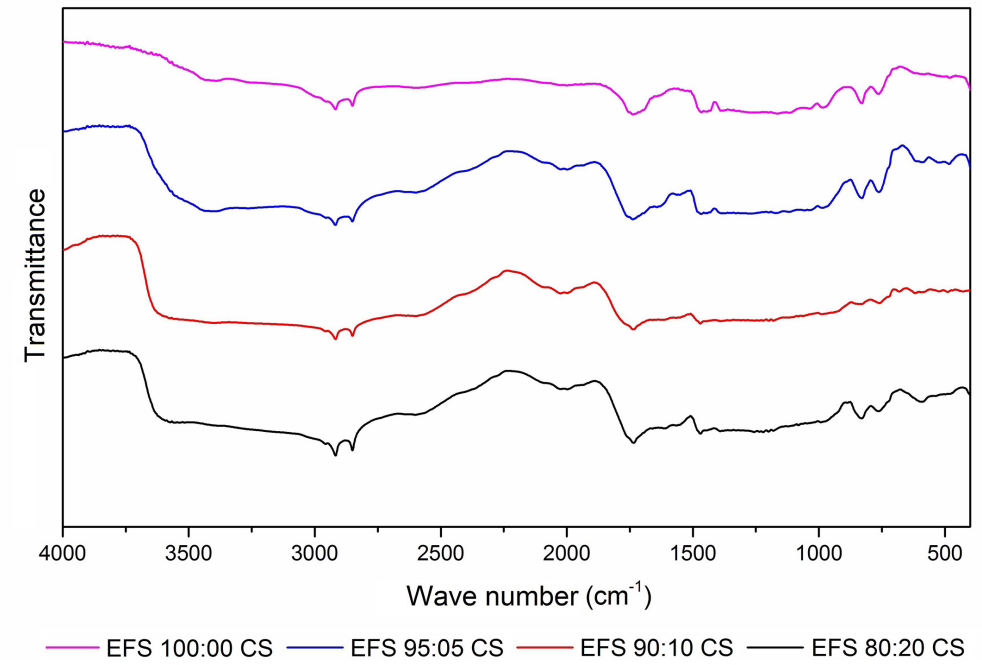

FIGURE 1 - FTIR spectra of EFS 100:00 CS (control) and of the associations containing 5%, 10% and 20% of CS. *EFS* Eudragit® FS 30, *CS* Chondroitin sulfate.

Raman spectra showed a characteristic peak of CS at 1063 cm^{-1} in EFS films associated with CS, which is characteristic of the symmetrical stretching of the grouping SO_3^-, becoming better evidenced with the addition of the polysaccharide (Lim *et al*., 2011; Esmonde-White *et al.*, 2011). The spectrum of a chemical compound provided by Raman spectroscopy has been referred to as the compound fingerprint. This characterization provided by this technique has been very useful in identifying several chemical compounds, including those with a high degree of complexity (Kudelski, 2008). As the spectra of films did not reveal the emergence or extinction of Raman peaks, it is suggested that the addition of the natural polysaccharide did not change the chemical structure of the EFS (Figure 3).

Determination of the film swelling index

The swelling index (Sw%) was the parameter used to assess the degree of hydration of polymer films. As shown in Figure 4, the swelling of the films was proportional to the pH value and the concentration of CS. The influence of pH is due to the pH dependence of the EFS-based polymer, whose dissolution occurs at a pH higher than 7.0, thus justifying the higher Sw% in SIF. Regarding the polysaccharide, the increase in its concentration promoted a higher degree of hydration of films, both immersed in SGF and SIF. However, in both fluids, there was a partial disintegration of films combined with 20% of CS, causing a false decrease in the Sw%, particularly in SGF. Bunhak *et al*. (2007a) found a similar behavior upon studying the influence of CS on the formation of ethyl cellulose films. Disintegration was also observed in films composed of 20% of CS. The author attributed this to the high water solubility of the natural polysaccharide.

The hydration of the films is a factor of interest and concern. In the first case, it is an essential aspect to degradation by colonic microbiota (Bunhak *et al.*, 2007a). In contrast, the increased Sw% may cause premature delivery of the drug (Pinto, 2010). We can thus infer that the EFS 90:10 CS combination film is the candidate most appropriate for the development of coatings seeking a modified drug delivery.

Water vapor transmission (WVT)

The permeability of polymer films was measured by the water vapor transmission rate (Joshi, Petereit, 2013). Figure 5 shows the average values of the WVT rate in function of CS concentration and time. The highest concentration of the polysaccharide promoted an increase in the WVT, which may be associated with the hydrophilicity provided by CS. Souto-Maior *et al.* (2010) observed a similar behavior upon preparing Eudragit® RS 30 D films associated with phosphated pectin and

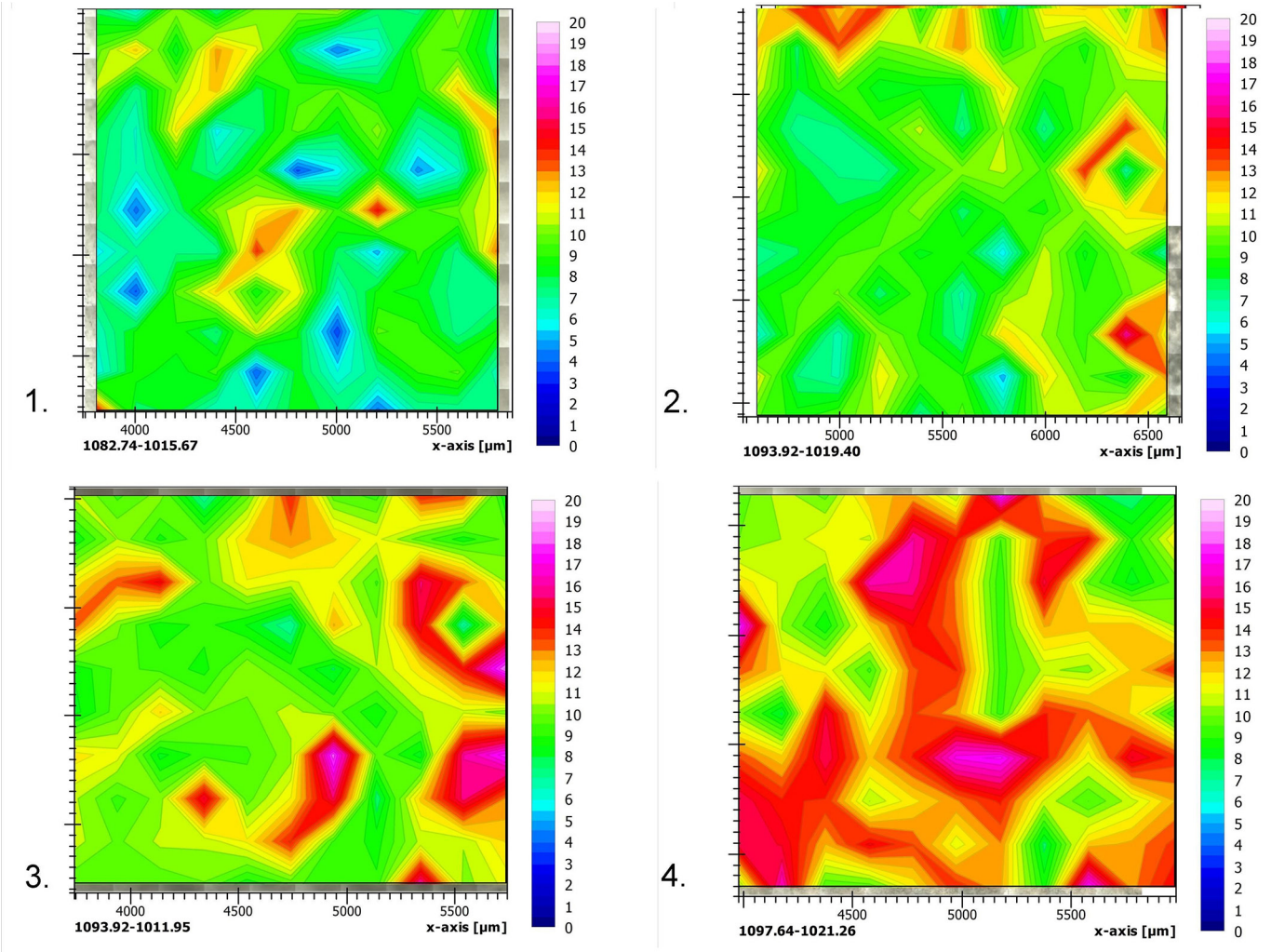

FIGURE 2 - Raman chemical imaging of the EFS 100:00 CS (1.), EFS 95:05 CS (2.), EFS 90:10 CS (3.) and EFS 80:20 CS (4.) composition films. The intensity scale to the right indicates the quantity of chondroitin sulfate, wherein the minimum (zero) indicates absence of the natural polysaccharide.

Bioecolians® prebiotic oligosaccharide. The increase in phosphated pectin caused a greater WVT rate when compared to the control film composed only of Eudragit® RS 30 D. The author attributed this phenomenon to the hydrophilicity related to the addition of the polysaccharide. Bunhak *et al.* (2007b), using Eudragit® RS 30 D associated with CS, also found similar results for permeability, which showed a proportional relation to the increase in polysaccharide concentration.

Scanning electron microscopy

Scanning electron microscope (SEM) was performed only with the control composition (EFS 100:00 CS) and with the composition containing 10% of CS (EFS 90:10 CS), since the latter proved to be the most suitable for the development of coatings aimed at a modified delivery of drugs, according to the results found by the swelling index assay.

From the analysis of micrographs (Figure 6), it was observed that the control film (EFS 100:00 CS) in SGF showed insignificant changes in the polymeric mesh when compared to the dry assay. Compared to the control in SIF, which can be seen in the study by Lima (2013), there is a slight modification of the polymer mesh, which is associated with the pH dependence of EFS. This does not occur when comparing EFS 90:10 CS composition films immersed in SGF and SIF. In SGF, there is a superficial breakdown of the polymer mesh, probably related to an increase in hydrophilicity associated with the presence of CS. However, this breakdown is more significant in SIF because of the pH dependency of EFS combined with the hydrophilic characteristic of the polysaccharide (Lee *et al.*, 2015). Morphological changes recorded with SEM

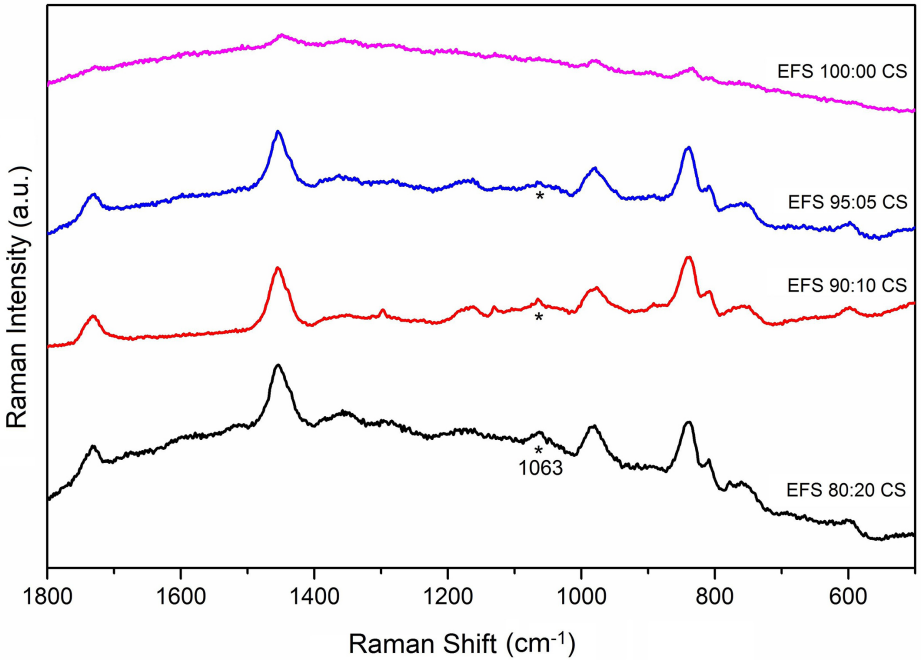

FIGURE 3 - Raman spectra of the EFS 100:00 CS, EFS 95:05 CS, EFS 90:10 CS and EFS 80:20 CS composition films. Characteristic peak of CS at 1063 cm^{-1}, characteristic of symmetric stretching of the grouping SO_3^-.

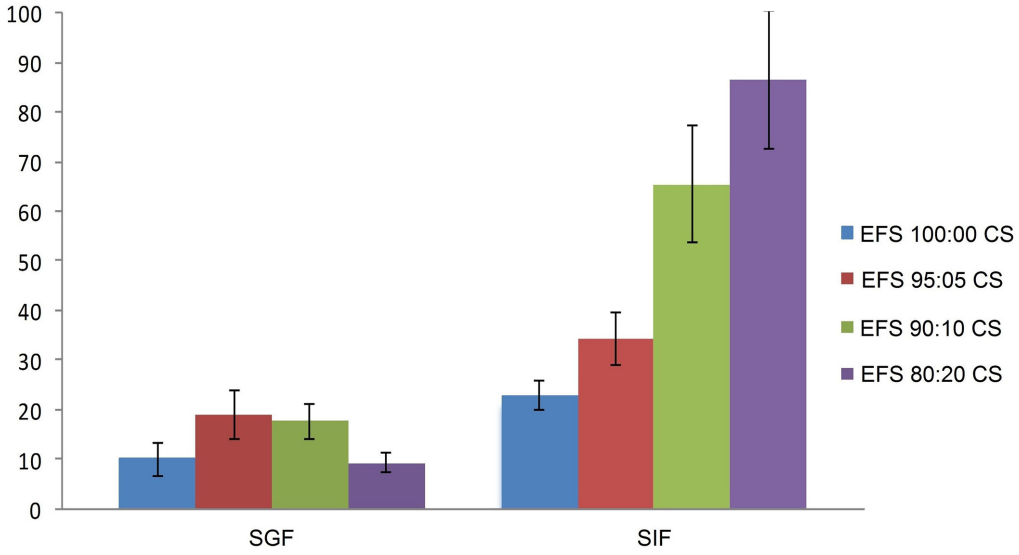

FIGURE 4 - Swelling index (Sw%) of the EFS 100:00 CS, EFS 95:05 CS, EFS 90:10 CS and EFS 80:20 CS composition films in the simulated gastric fluid (SGF; pH 1.2) and simulated intestinal fluid (SIF; pH 6.8), (n=3).

reveal the influence of CS on changing film properties, agreeing with the results obtained in Swelling Index and water vapor transmission studies.

CONCLUSION

The results suggest that the synthetic polymer Eudragit® FS 30 D and the polysaccharide chondroitin sulphate, when combined, may form films that are candidates for use in the coating of modified drug delivery systems especially due to the synergism between pH dependence and specific biodegradability by the colonic microbiota. The analysis using FTIR revealed no occurrence of a chemical interaction between the polymers, since a displacement and/or an appearance of new bands, that would characterize changes in the chemical

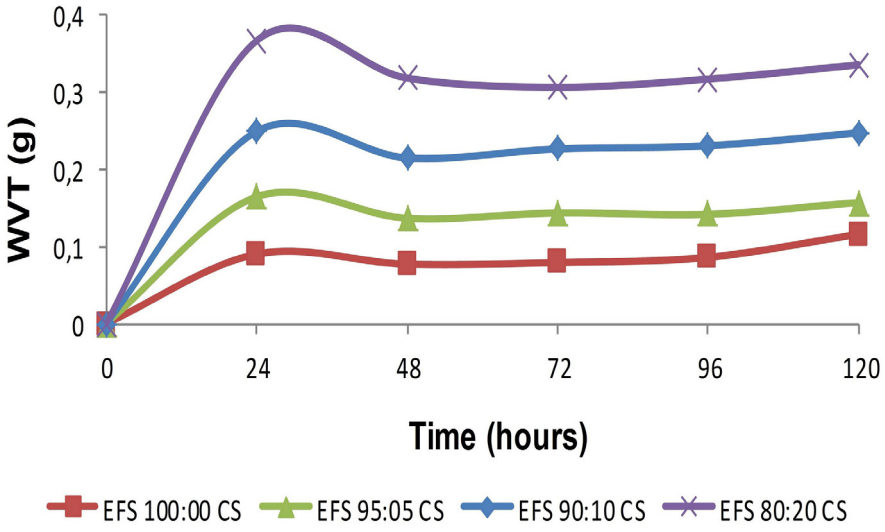

FIGURE 5 - WVT in function of the composition of polymeric films. Eudragit® FS 30 D (EFS) associated with different concentrations of chondroitin sulfate (CS), (n=3).

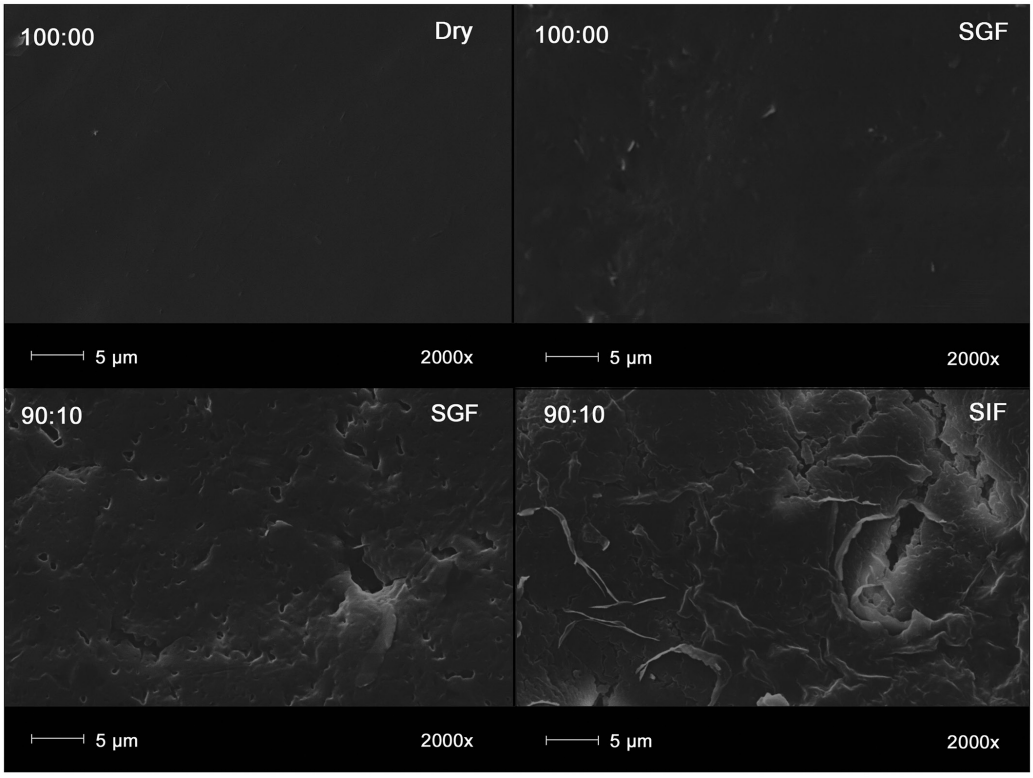

FIGURE 6 - Micrographs of dried isolated films and after swelling for 60 minutes in SGF and SIF: Eudragit® FS 30 D control, Eudragit® FS 30 D associated with chondroitin sulfate at an EFS 90:10 CS concentration.

structure of the components, was not observed. This result corroborates that obtained by Raman spectroscopy, since the emergence or the extinction of Raman peaks was not identified. EFS 90:10 CS composite films were the most suitable for the development of coatings of modified drug delivery systems considering the characteristics of hydration and permeability of the various combinations tested.

ACKNOWLEDGMENTS

The authors thank to companies Almapal (São

Paulo/SP) by donation of the Eudragit® FS 30 D and Solabia (Maringá/PR) by donation of the chondroitin sulfate. Our most sincere thanks to Professor Antonio Medina Neto DFI and COMCAP-UEM the full support for the characterization tests of the polymeric films.

REFERENCES

AMRUTKAR, J.R.; GATTANI, S.G. Chitosan–chondroitin sulfate based matrix tablets for colon specific delivery of indomethacin. *AAPS Pharmscitech*, v.10, n.2, p.670-677, 2009.

BRAZ, R.; HECHENLEITNER, A.A.W.; CAVALCANTI, O.A. Extraction, structural modification and characterization of lotus roots polysaccharides (*Nelumbo nucifera Gaertn*). Excipient with potential application in modified drug delivery systems. *Lat. Am. J. Pharm.*, v.26, n.5, p.706-710, 2007.

BUNHAK, E.J.; MENDES, E.S.; PEREIRA, N.C.; PINEDA, E.A.G.; HECHENLEITNER, A.A.W.; CAVALCANTI, O.A. Análises físico-químicas de biofilmes de sulfato de condroitina modificado. *Quim. Nova*, v.38, n.3, p.316-320, 2015.

BUNHAK, E.J.; MENDES, E.S.; PEREIRA, N.C.; CAVALCANTI, O.A. Influência do sulfato de condroitina na formação de filmes isolados de etilcelulose. Avaliação das características de hidratação e permeabilidade. *Lat. Am. J. Pharm.*, v.26, n.1, p.89-95, 2007a.

BUNHAK, E.J.; MENDES, E.S.; PEREIRA, N.C.; CAVALCANTI, O.A. Influência do sulfato de condroitina na formação de filmes isolados de polimetacrilato: avaliação do índice de intumescimento e permeabilidade ao vapor d'água. *Quim. Nova*, v.30, n.2, p.312-317, 2007b.

CAVALCANTI, O.A.; VAN DEN MOOTER, G.; CARAMICO-SOARES, I.; KINGET, R. Polysaccharides as excipients for colon-specific coatings. Permeability and swelling properties of casted films. *Drug Dev. Ind. Pharm.*, v.28, n.2, p.157-164, 2002.

ESMONDE-WHITE, K.A.; ESMONDE-WHITE, F.W.L.; MORRIS, M.D.; ROESSLER, B.J. Fiber-optic Raman spectroscopy of joint tissues. *Analyst*, v.136, n.8, p.1675-1685, 2011.

EVONIK INDUSTRIES. Eudragit® FS 30 D. Specification and Test Methods. Available at: <http://eudragit.evonik.com/product/eudragit/Documents/evonik-specification-eudragit-fs-30-d.pdf>. Accessed on: Jul. 2015.

HUANG, L.; SUI, W.; WANG, Y.; JIAO, Q. Preparation of chitosan/chondroitin sulfate complex microcapsules and application in controlled release of 5-fluorouracil. *Carbohydr. Polym.,* v.80, n.1, p.168-173, 2010.

JOSHI, S.; PETEREIT, H.-U. Film coatings for taste masking and moisture protection. *Int. J. Pharm.*, v.457, n.2, p.395-406, 2013.

KUDELSKI, A. Analytical applications of Raman spectroscopy. *Talanta*, v.76, n.1, p.1-8, 2008.

KUMAR, R.; CHANDRA, A.; GAUTAM, P.K. Modified approaches for colon specific drug delivery system: a review. *Indian J. Pharm. Biol. Res.*, v.1, n.3, p.67-79, 2013.

LEE, J.Y.; CHUNG, S.J.; CHO, H.J.; KIM, D.D. Bile acid-conjugated chondroitin sulfate A-based nanoparticles for tumor-targeted anticancer drug delivery. *Eur. J. Pharm. Biopharm.,* v.94, p.532-541, 2015.

LIMA, I.A. *Desenvolvimento e caracterização de filmes isolados pululana-polimetacrilato como material potencial à liberação entérica de fármacos*. 2013. 39 f. Dissertação (Mestrado em Ciências Farmacêuticas). Universidade Estadual de Maringá, Paraná, 2013.

LIM, N.S.J.; HAMED, Z.; YEOW, C.H.; CHAN, C.; HUANG, Z. Early detection of biomolecular changes in disrupted porcine cartilage using polarized Raman spectroscopy. *J. Biomed. Opt.*, v.16, n.1, p. 017003-017003-10, 2011.

MCCONNELL, E.L.; SHORT, M.D.; BASIT, A.W. An in vivo comparison of intestinal pH and bacteria as physiological trigger mechanisms for colonic targeting in man. *J. Control. Release*, v.130, n.1, p.154-160, 2008.

MELO, E.J.; ALVES, B.R.V.; FREITAS, A.A.; MUNIZ, E.C.; CAVALCANTI, O.A. Influence of the addition of alpha or gamma-cyclodextrin on the formation of free films in the polymethacrylates eudragit® FS30D. *Lat. Am. J. Pharm.*, v.29, n.6, p.919-926, 2010.

PAUDEL, A.; RAIJADA, D.; RANTANEN, J. Raman spectroscopy in pharmaceutical product design. *Adv. Drug Deliv. Rev.*, v. 89, p.3-20, 2015.

PINTO, J.F. Site-specific drug delivery systems within the gastro-intestinal tract: from the mouth to the colon. *Int. J. Pharm.*, v.395, n.1, p.44-52, 2010.

RABITO, M.F.; REIS, A.V.; TAMBOURGI, E.B.; FREITAS, A.R.; CAVALCANTI, O.A. A pH/enzyme-responsive polymer film consisting of Eudragit® FS 30 D and arabinoxylane as a potential material formulation for colon-specific drug delivery system. *Pharm. Dev. Technol.*, v.17, n.4, p.429-436, 2012.

RAMASAMY, T.; KHANDASAMY, U.S.; SHANMUGAM, S.; RUTTALA, H. Formulation and evaluation of chondroitin sulphate tablets of aceclofenac for colon targeted drug delivery. *Iranian J. Pharm. Res.*, v.11, n.2, p.465-479, 2012.

SANTOS, L. F.; PINEDA, E. A. G.; CELLIGOI, M. A. P. C.; CAVALCANTI, O. A. Levan as a new additive for colon-specific films: A new approach in the use of exopolysaccharides in time-dependent free films (aminoalkyl methacrylate copolymer RS). *Pak. J. Pharm. Sci.*, v.26, n.5, p.943-948, 2013.

SMITH, G.P.S.; MCGOVERIN, C.M.; FRASER, S.J.; GORDON, K.C. Raman imaging of drug delivery systems. *Adv. Drug Deliv. Rev.*, v. 89, p.21-41, 2015.

SOUTO-MAIOR, J.F.A.; REIS, A.V.; PEDREIRO, L.N.; CAVALCANTI, O.A. Avaliação da pectina fosfatada aplicada na formação de filmes isolados. Material candidato a novos sistemas para liberação modificada de fármacos. *Rev. Bras. Ciênc.Farm.,* v.44, n.2, p.203-213, 2008.

SOUTO-MAIOR, J.F.A.; REIS, A.V.; PEDREIRO, L.N.; CAVALCANTI, O.A. Phosphated crosslinked pectin as a potential excipient for specific drug delivery: preparation and physicochemical characterization. *Polym. Int*., v.59, n.1, p.127-135, 2010.

ZIMOVÁ, L.; VETCHÝ, D.; MUSELÍK, J.; STEMBÍREK, J. The development and in vivo evaluation of a colon drug delivery system using human volunteers. *Drug Deliv.*, v.19, n.2, p.81-89, 2012.

Received for publication on 23th February 2016
Accepted for publication on 03rd October 2016