AAPS PharmSciTech 2002; 3 (3) article 22 (http://www.aapspharmsci.org).

Double-Layered Mucoadhesive Tablets Containing Nystatin

Submitted: March 11, 2002; Accepted: July 24, 2002

Juan Manuel Llabot[1], Ruben Hilario Manzo[1] and Daniel Alberto Allemandi[1]

[1]Departamento de Farmacia, Facultad de Ciencias Químicas, Universidad Nacional de Córdoba, Córdoba, Argentina

ABSTRACT The objective of this work was to design a mucoadhesive tablet with a potential use in the treatment of oral candidosis. A 2-layered tablet containing nystatin was formulated. Lactose CD (direct compression), carbomer (CB), and hydroxypropylmethylcellulose (HPMC) were used as excipients. Tablets were obtained through direct compression. Properties such as in vitro mucoadhesion, water uptake, front movements, and drug release were evaluated. The immediate release layer was made of lactose CD (100 mg) and nystatin (30 mg). The CB:HPMC 9:1 mixture showed the best mucoadhesion properties and was selected as excipient for the mucoadhesive polymeric layer (200 mg). The incorporation of nystatin (33.3 mg) in this layer affected the water uptake, which, in turn, modified the erosion front behavior. Nystatin showed a first-order release. The polymeric layer presented an anomalous kinetic (n = 0.82) when this layer was individually evaluated. The mucoadhesive tablet formulated in this work releases nystatin quickly from the lactose layer and then in a sustained way, during approximately 6 hours, from the polymeric layer. The mixture CB:HPMC 9:1 showed good in vitro mucoadhesion. A swelling-diffusion process modulates the release of nystatin from this layer. A non-Fickian (anomalous) kinetic was observed.

KEYWORDS: mucoadhesive tablets, nystatin, oral candidosis.

INTRODUCTION The buccoadhesive drug delivery systems have been developed basically to increase the retention of drug in the oral cavity and/or to keep a sustained release of drug towards the medium from where it is constantly removed [1,2]. These characteristics make this kind of drug delivery system very useful for the treatment of buccal diseases among which oral candidosis is one of the most important [3].

The clinical treatment of this pathology using conventional pharmaceutical dosage forms—such as solutions, gels, suspensions, and mouthwashes—is usually not very effective, mainly because drugs are quickly removed from the oral cavity. In order to solve this problem, the design of different buccoadhesive pharmaceutical dosage forms containing nystatin [4], miconazole [5], and fungicidal agents [6] has been reported. Similar systems have also been proposed to treat other buccal affections such as periodontitis [7,8] or to supply the buccal environment with fluor supplement [9].

The strategy for designing buccoadhesives is based principally on the utilization of polymers with suitable physicochemical properties, such as polyacrylic acid (carbomer [CB]) and cellulose derivatives (hydroxypropylmethylcellulose [HPMC]) [1,10-12].

This work deals with the design of a double-layered buccoadhesive tablet containing nystatin. For that purpose, in vitro mucoadhesive properties of the CB:HPMC mixtures, the water uptake, and the swelling-diffusion processes were evaluated. The relation between these properties and the in vitro nystatin release rate were analyzed.

MATERIALS AND METHODS

Materials

Tablet formulation. Double-layered tablets were prepared by direct compression. A physical blend of the polymers was mixed with mortar and pestle for 15 minutes. Then the mixture was compressed in a single-punch (13-mm) eccentric press (Delfabro HPH 15, San Francisco, Córdoba) under 1500 kg/cm ^{2} for 5 seconds, resulting in a 2-mm-thick tablet.

Correspondence to:
Daniel Alberto Allemandi
Facsimile: 54 351 4334127
E-mail: dalemand@yahoo.com

Table 1. Double-Layered Nystatin Tablet Composition*

Immediate Release Layer		Sustained Release Layer	
Components	**Amount, mg**	**Components**	**Amount, mg**
Lactose CD	100	CB:HPMC 9:1	200
Nystatin	30	Nystatin	33.3

*CB indicates carbomer; HPMC, hydroxypropylmethylcellulose.

Water uptake. The liquid uptake kinetic of the tablets was evaluated by using a modified version of the apparatus described by Nguyen-Xuan et al [13]. Distilled water was used.

Mucoadhesion test. The in vitro mucoadhesion was measured in terms of the force needed to pull out a tablet from a gelatin gel layer (30% wt/wt), simulating oral mucose, with an adapted Jolly Balance (Facultad de Astronomía, Matemáticas y Física, Córdoba, Argentina). The tablets were fixed to a support with cyanoacrylate adhesive and then suspended from a spring. They were lowered until they just contacted the surface of the gelatin, with 0.1 mL of distillated water between the tablet and the gelatin gel. A 20-g force was applied to the tablets for 30 seconds. Then the platform was raised at 0.3 cm/s until the tablet was separated from the gelatin. This point represents the adhesive bond strength between these elements. This value is expressed in N/cm^2. For each mixture, the assay was performed for 5 different tablets and averaged.

In vitro drug release. These experiments were carried out using a US Pharmacopoeia (USP) Nº II dissolution apparatus (Hanson SR II 6 Flask Dissolution Test Station Hanson Research Corporation, Chatsworth, CA) at 37ºC and 75 rpm with distilled water as a medium (900 mL). The tablet was fixed with a cyanoacrylate adhesive to a metallic disk placed at the bottom of the vessel containing distilled water. The samples were withdrawn, filtered, and measured at 306 nm with a UV-Vis spectrophotometer (Shimadzu UV 160-A, Shimadzu Corporation, Kyoto, Japan).

Movement front determinations. Swelling studies of tablets were performed by clamping each matrix between 2 transparent glasses and introducing them into the vessel with distilled water at 37ºC without agitation, as described by Colombo et al [14]. The front position was determined through the measurement of the distance of the fronts relative to the radius of the tablet at time = 0. A micrometric gauge and a magnifying glass were used to carry out the determination, which was done in triplicate.

RESULTS AND DISCUSSION

Design Rationality

An ideal pharmaceutical dosage form for buccal affection treatments would be able to (1) release drug immediately to produce a prompt pharmacological action, (2) remain in the oral cavity, and (3) provide a sustained release of enough drug over an extended period of time.

Taking into account such requirements, the design of a mucoadhesive double-layered tablet was addressed. The selected composition is detailed in **Table 1**.

The amount of nystatin in the formula was established according to its clinical use [15] and the doses usually contained in some brand drug products [16].

Lactose CD was selected as diluent for its high aqueous solubility, its flavoring characteristics, and its physical-mechanical properties, which make it suitable for direct compression [17,18]. The lactose CD layer containing nystatin was designed to promptly release the drug to obtain the attack dose.

The second layer, composed of CB:HPMC mixtures, produces the mucoadhesion and modulates the nystatin release. The polymer ratio was determined by measuring in vitro mucoadhesion. CB:HPMC 9:1 proved to be the most convenient mixture (see below).

Tablet Mucoadhesion and Water Uptake

In the mucoadhesion process, several well-defined events have been identified. The interaction between polymer and mucosa is carried out through (1) close polymer-mucosa contact, where adsorption occurs as a result of a reduction in the surface free energy as 2 surfaces are lost and a new interface is formed, and (2) a consolidation step, where several physical-chemical interactions occur to extend mucoadhesion.

Several polymers and hydrophilic macromolecules containing groups able to form hydrogen bonds have showed good adhesion properties that seem to be enhanced by the incorporation of amine and carboxylic groups [1]. This water-activated process produces the polymer swelling and improves the consolidation step that increases the mobility of molecules and facilitates the interpenetration with the mucus layer [1]. Therefore, the polymer swelling is a property related to the mucoadhesion of the system. In vitro mucoadhesion and water uptake were assayed in order to evaluate such properties in CB:HPMC mixtures. Likewise, the effect of nystatin incorporation on these properties was evaluated. The results are shown in **Figures 1**, **2**, and **3**.

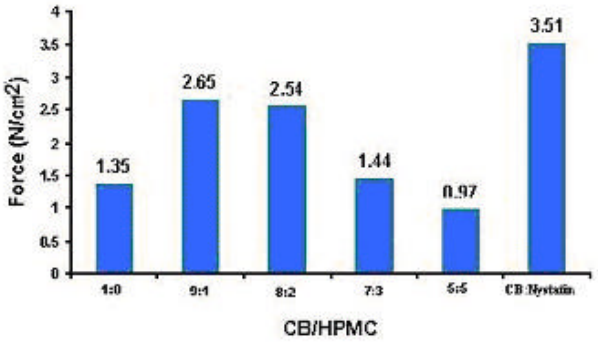

Figure 1. Mucoadhesion of compressed CB:HPMC mixtures in gelatin gel (30% wt/wt).

Polymer mixtures containing high percentages of CB (>80%) showed the best in vitro mucoadhesion (**Figure 1**). As a result, the CB:HPMC 9:1 mixtures were selected for the formulation of the polymeric layer. The incorporation of nystatin in the polymer blend had an unexpected outcome: a considerable increase in mucoadhesion was observed. It is possible that the presence of nystatin may be responsible for the increase in the osmotic pressure of the mixture, which, in turn, would produce a rise in the water uptake, facilitating the interaction between the polymers and the mucus. Likewise, it was observed that polymer mixtures that showed higher adhesion also presented a higher water uptake (**Figures 2** and **3**).

However, the CB:HPMC 9:1 mixture, which presents the best mucoadhesion, absorbs slightly lower amounts of water than CB:HPMC 8:2 (**Figure 2**). Another point that should be considered is the water uptake observed after 1 hour. After that time, the drug dissolved in the gel layer

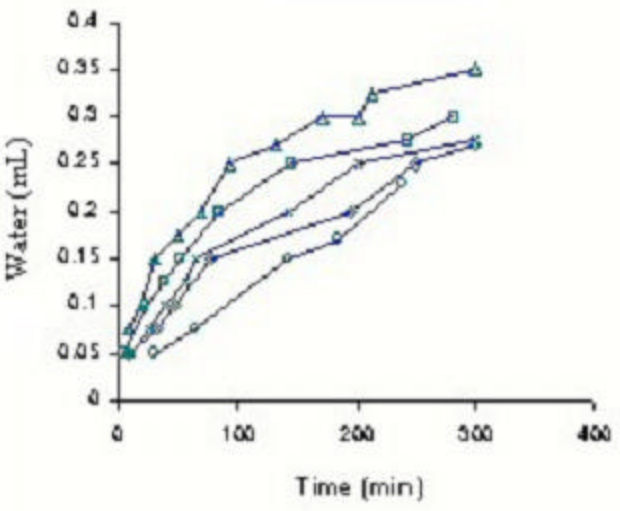

Figure 2. Water uptake of compressed CB:HPMC mixtures of different CB:HPMC ratios and pure CB (○), 5:5 (◇), 7:3 (X), 9:1 (□), and 8:2 (△).

would be enough to produce an increase in the water uptake, changing the erosion front kinetics (see next section).

Drug Diffusion in Mucoadhesive Layer

Since hydrophilic polymers like CB and HPMC can easily take in water and can swell because of structural relaxation, it is possible to modulate the drug release, which depends on the interaction between water, polymer, and drug. As a result, an outer gel layer is formed, producing in the matrix physical changes that

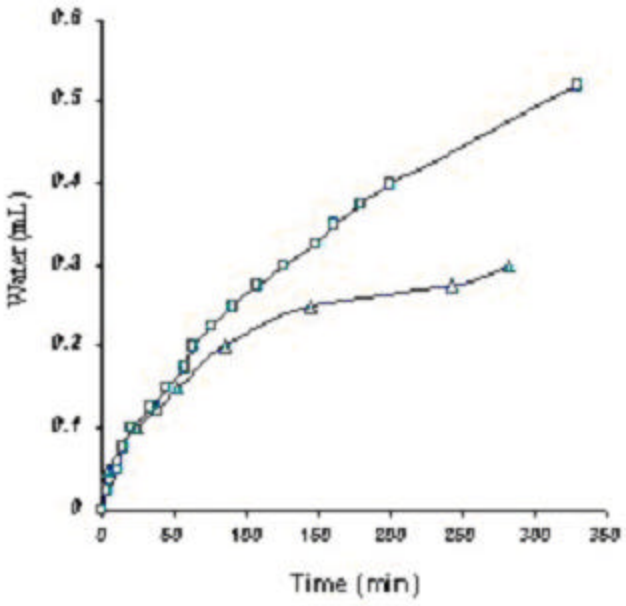

Figure 3. Water uptake of matrix layer with nystatin (□) and without nystatin (△).

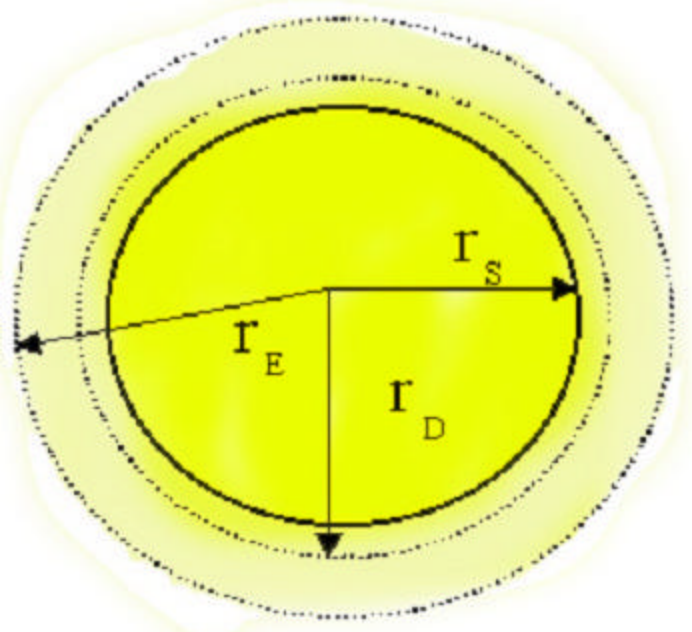

Figure 4. Schematic representation of the front positions.

can be observed through the behavior of different fronts as the process develops [14]. These fronts have been identified (**Figure 4**) as (1) swelling front (ratio = r_S), delimitating the boundary between the glassy polymer and its rubbery gel state, (2) diffusion front (ratio = r_D), indicating the boundary between the undissolved (solid) and the dissolved drug in the gel layer, and (3) erosion front (ratio = r_E), delimiting the boundary between the matrix and the dissolution medium.

The evaluation of the movement of these fronts permits us to determine 3 important parameters for the swelling-diffusion process: (1) the water uptake rate, which is related to the swelling front position, (2) the drug dissolution rate, which is reflected in the diffusion front position, and (3) the erosion rate of the matrix, which corresponds to the erosion front.

In this work, front movements for the CB:HPMC 9:1 mucoadhesive layer containing nystatin were analyzed; the results are shown in **Figure 5**.

In this case, the color of nystatin makes the observation of the front movements easier. Nystatin is a yellow drug in solid state; when it is dissolved in water, an intense yellow color appears. As **Figure 5** shows, the erosion front moved outward while the swelling and diffusion fronts moved inward. Owing to the immediate swelling of the polymers, a rapid initial increase in the erosion front was observed; then that increase became constant. In the same way, the diffusion and swelling fronts showed a rapid initial change; after approximately an hour, an almost linear decrease as they moved inward was observed.

It is important to analyze not just the behavior of the fronts but the relation between them. The distance between the diffusion and erosion front defines the dissolved drug gel layer, which is involved in controlling the drug release process [19]. **Figure 6** depicts the behavior of this gel layer over time. The increase in the thickness of this layer is not constant; it increases quickly early on and then slows down. It is very interesting to note that this pattern does not coincide with the constant release of the nystatin from the matrix. If these did coincide, the polymer swelling and relaxation would be secondary mechanisms in drug release, while drug dissolution and diffusion would be the main controller of the process.

Besides, a significant difference between the swelling and the diffusion front ($r_S - r_D$) is observed because of the low aqueous solubility of nystatin. This difference, as is well known, is inversely proportional to the drug solubility. For that reason, matrices containing aqueous soluble drugs have been reported to have smaller differences [14,19].

Finally, the erosion process would not be significant in the modulation of nystatin release from the matrix CB:HPMC 9:1 because of the high percentage of CB, owed to this polymer is not soluble and very rigid in gel state because of its chemical crosslinking.

Nystatin Release From Tablets

In vitro nystatin release from the double-layered tablet and from each separate layer were assayed. The comparison of these results led to qualitative conclusions, since assay conditions change when the tablet is split into its layers. The results are shown in **Figure 7**.

The immediate release lactose-nystatin layer releases over 50% of the drug in 1 hour. It exhibits a profile of delivered drug concentration versus time of Noyes-Whitney type, suggesting that the dissolution step controls the process. Regarding the mucoadhesive layer, a slower rate of drug release is observed; about 80% of drug is delivered in 6 hours with a practically constant rate (zero order).

Last, when the drug release profile from the double-layered tablet is analyzed, it is clear that the immediate release layer produces the main contribution in the first 2 hours and that the mucoadhesive layer is the main contributor to the sustained release observed in the last 4 hours (**Figure 7**). The polymeric layer release mechanism can also be evaluated from its diffusional exponent (*n*) [20]. A value of $n = 0.82$ was calculated

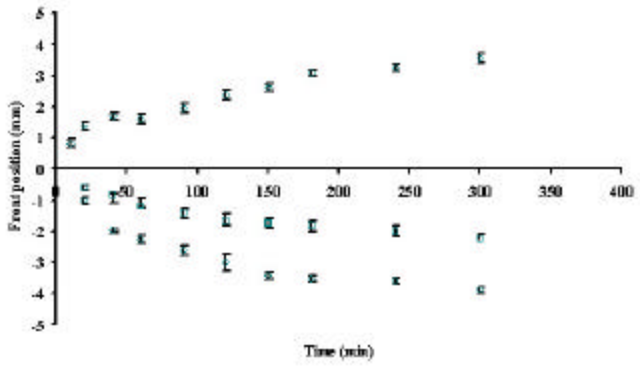

Figure 5. Front movement of matrix layer containing nystatin: swelling front (○), erosion front (◇), and diffusion front (□).

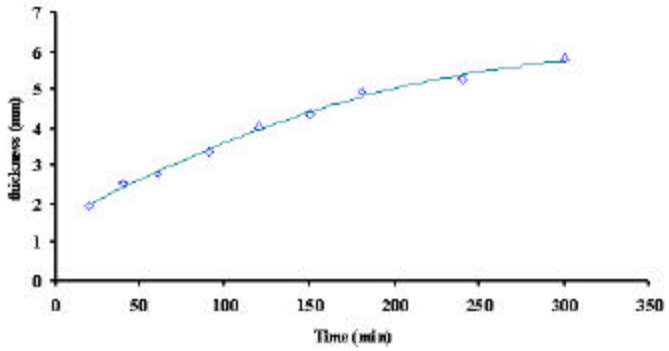

Figure 6. Thickness of dissolved gel layer (r_E - r_D) versus time.

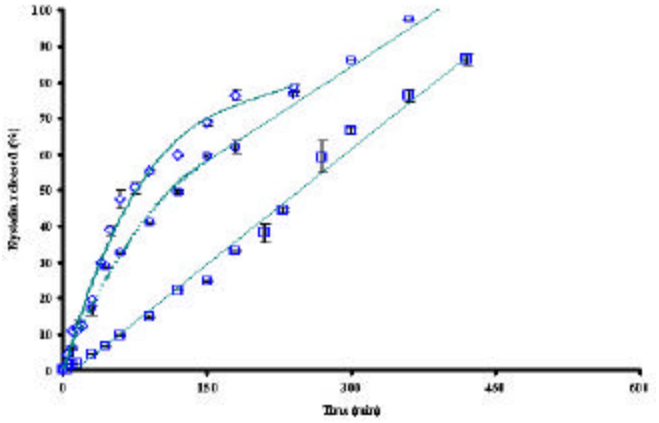

Figure 7. Nystatin dissolution profile in distilled water at 37°C, 75 rpm (USP N° II dissolution apparatus): CB:HPMC 9:1 layer (□), lactose layer (◇) and double-layer tablet (○).

for the release of nystatin, indicating an anomalous (non-Fickian) kinetic and suggesting that the release mechanism is modulated by the drug diffusion and the polymer relaxation. Similar kinetic results have been reported for CB:HPMC matrices containing metronidazole [21] and morphine sulphate [22].

In in vitro assays, the buccal environment was artificially reproduced and drug liberation was inhibited in 1 face of the layer. This restriction could affect the kinetic of nystatin release. It has been reported that in these kinds of systems, when of 1 of the faces of the mucoadhesive layers is inhibited, as may occur when the tablet is adhered to the mucose, the delivery kinetics shift to a zero order [23]. However, this article does not evaluate this phenomenon.

CONCLUSION The mucoadhesive tablet formulated in this work releases nystatin quickly from the lactose layer and then in a sustained way for approximately 6 hours from the polymeric layer. The mixture CB:HPMC 9:1 showed good in vitro mucoadhesion. This property was enhanced with the incorporation of nystatin in the matrix. A swelling-diffusion process modulated nystatin release from this layer. A non-Fickian (anomalous) kinetic was observed. After 1 hour, the mucoadhesive layer water uptake was increased due to the incorporation of the drug. A similar behavior was observed in the erosion front movement. Double-layered mucoadhesive tablets containing nystatin showed good in vitro biopharmaceutical performance, and their potential usefulness for oral candidosis treatment will be evaluated later.

ACKNOWLEDGEMENTS The authors thank Agencia Córdoba Ciencia SE and Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET) for financial support.

REFERENCES

1. Machida Y, Nagai T. Bioadhesive preparation as topical dosage Forms. In: Mathiowitz E, Chickering III D E, Lehr C M, eds. Bioadhesive drug delivery systems. New York, NY: Marcel Dekker Inc; 1999;98:646-647.

2. Weatherell JA, Robinson C, Rathbone MJ. The flow of saliva and its influence on the movement, deposition and removal of drugs administred to the oral cavity. In: Rathbone MJ ed. Oral mucosal drug delivery. New York, NY: Marcel Dekker Inc; 1996;74:157.

3. Carr D, Corbett CE, Koo PJS. Mycotic and parasitic infections. In: Herfindal ET, Gourley DR eds. Textbook of therapeutic: drug and disease management, 6th ed. Baltimore MD: Williams & Wilkins: 1996;1432.

4. Millns B, Martin MV. Nystatin pastilles and suspension in the treatment of oral candidosis. Brit Dent J. 1996;181(6):209-211.

5. Bouckaert S, Schautteet H, Lefebvre RA, Remon JP, Van Clooster R. Comparison of salivary miconazole concentrations after administration of a bioadhesive slow-release buccal tablet and oral gel. Eur J Clin Pharmacol. 1992;43:137-140.

6. Codd JE, Deasy PB. Synergistic antifungal interaction between miconazole nitrate and chlorhexidine acetate. Int J Pharm. 1998;173:3-11.

7. Bromberg LE, Buxton DK, Friden PM. Novel periodontal drug delivery system for treatment of periodontitis. J Control Release. 2001;71:251-259.

8. Bromberg LE, Braman VM, Rothstein DM, et al. Sustained release of silver from periodontal wafers for treatment of periodontitis. J Control Release. 2000;68:63-72.

9. Bottemberg P, Cleymaet R, DeMuynck C, et al. Development and testing of bioadhesive, fluoride-containing slow-release tablets for oral use. J Pharm Pharmacol. 1991;43:457-464.

10. Bottemberg P, Herman J, Coomans D, et al. Bioadhesion of fluoride-containing slow-release tablets on porcine oral mucosa in vitro. STP Pharma. 1989;5(12):863-866.

11. Ponchel G, Touchard F, Duchene D, Pepas NA. Bioadhesive analysis of controlled release systems, I: fracture and interpenetration analysis in poly(acrylic acid)-containing systems. J Control Release. 1987;5:129-141.

12. Nagai T. Topical mucosal adhesive dosage forms. Med Res Rev. 1986;6(2):227-242.

13. Nguyen-Xuan T, Towart R, Terras A, Jacques Y, Buri P, Gurny R. Mucoadhesive semi-solid formulations for intraoral use containing sucralfate. Eur J Pharm Biopharm. 1996;43(2):133-137.

14. Colombo P, Bettini R, Massimo G, Catellani PL, Santi P, Peppas NA. Drug diffusion front movement is important in drug release control from swellable matrix tablets. J Pharm Sci. 1995;84(8):991-997.

15. Bennett JE, Fármacos antimicrobianos-Fármacos antimicóticos In: Hardman JG, Limbird LE, Molinoff PB, Ruddon RW, Goodman Gilman A. Las Bases Farmacológicas de la Terapéutica 9 ed., México DC, Mexico: McGraw-Hill Interamericana; 1996(II), 49, 1261.

16. MYCOSTATIN (nystatin lozenges, USP) Pastilles. Available at: http://www.bms.com/medicines/data/index.html.

17. Goodhart FW. Lactose. In: Wade P. Weller, eds. Handbook of pharmaceutical excipients, 2th ed. Washington DC: American Pharmaceutical Association; 1994; 252.

18. Shangraw RF. Compressed tablets by direct compression. In: Lieberman HA, Lachman L, Schwartz JB, eds. Pharmaceutical Dosage Form: Tablets. Vol 1. New York, NY: Marcel Dekker; 1990:195.

19. Colombo P, Bettini R, Santi P, Peppas NA. Swellable matrices for controlled drug delivery: gel-layer behavior, mechanisms and optimal performance. Pharm Sci Technol Today. 2000;3(6):198-204.

20. Kim H, Fassihi R. Application of a binary polymer system in drug release rate modulation, I: characterization of release mechanism. J Pharm Sci. 1997;86(3):316-322.

21. Ponchel G, Touchard F, Wouessidjewe D, Duchene D, Peppas NA. Bioadhesive analysis of controlled release systems, III: bioadhesive and release behavior of metronidazole containing poly(acrylic acid)-hydroxypropyl methylcellulose systems. Int J Pharm. 1987;38:65-70.

22. Anlar S, Capan Y, Guven O, Gogus A, Dalkara T, Hincal AA. Formulation and in vitro-in vivo evaluation of buccoadhesive morphine sulfate tablets. Pharm Res. 1994;11(2):231-236.

23. Conte U, Maggi L, Colombo P, La Manna A. Multi-layered hydrophilic matrices as constant release devices (Geomatrix Systems). J Control Release. 1993;26:39-47.