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Research paper

New oral solid dosage form for furosemide oral administration

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ABSTRACT

Furosemide (FURO) is a drug labeled in class IV of the Biopharmaceutics Classification System (BCS) as it is both poor soluble and poor permeable.

The aim of this work was to improve FURO biopharmaceutical properties by its formulation in a new solid oral dosage form. It consists in the realization of the composite MgAl-HTlc-FURO, obtained by FURO intercalation into the inorganic matrix hydrotalcite (MgAl-HTlc), and its successive formulation in tablets intended to be swallowed whole and to disintegrate rapidly in the stomach. These formulations were prepared by direct compression of a simple powder mixture constituted by MgAl-HTlc-FURO, a super disintegrant (Explotab®, Polyplasdone®XL, Polyplasdone®XL-10, Polyplasdone®INF 10 or L-HPC®LH-21) and a filler.

The prepared formulations were submitted to disintegration time tests, and only those displaying the lowest disintegration time in gastric medium were submitted to in vitro release studies. Drug dissolution profiles from MgAl-HTlc-FURO tablets were compared with those containing crystalline FURO alone or physically mixed to MgAl-HTlc instead of MgAl-HTlc-FURO. The results revealed that tablets containing MgAl-HTlc-FURO give the best dissolution profile and that L-HPC®LH-21 is able to promote the highest drug release in gastric medium, resulting in the most suitable super disintegrant in comparison with the other tested.

1. Introduction

The rate and the extent of drug dissolution and its absorption depend on the characteristics of the active pharmaceutical ingredient (API) as well as the dosage form properties. For this reason, the role of the formulation at the drug delivery site should not be ignored. Since an orally administered drug must be in solution, in order to be absorbed in the GI tract and to reach the systemic circulation, the dosage form plays an important role in conditioning the absorption rate.

Solid oral dosage (SOD) forms represent the main choice among the variety of oral devices for most APIs because they are easy to prepare, conveniently stored, easily transported, often are more stable than their liquid counterparts, and demonstrate a high patient acceptability [1]. For these reasons, SODs are produced so widely by pharmaceutical industries covering the 84% of USA and Europe pharmaceutical markets [2].

The absorption of many drugs, administered as SOD forms, depends on a succession of processes: (i) formulation disintegration, (ii) drug dissolution in the GI fluids, (iii) drug absorption across cell membranes into the systemic circulation and, ultimately, to its action site. In this context, it was demonstrated that API biopharmaceutical properties (solubility/dissolution rate and permeability) are very important as they influence drug bioavailability at biophase [3].

A large number of drugs, both on the market and in development, show problems of low efficacy responsible for therapy failure, mainly due to their unsatisfactory biopharmaceutical properties [4]. This means that when an API is orally administered as solid formulation, it is subject to incomplete dissolution, and/or low ability to permeate the membranes separating the absorption site from the systemic circulation, and sometimes to metabolic instability as the drug moves from the absorption site to the systemic circulation.

Furosemide (FURO), representative of this molecule category, is labeled in class IV of the Biopharmaceutics Classification System (BCS) because of its low water solubility (5–20 μg/ml) and low permeability [5]. FURO is preferentially absorbed in the stomach and upper intestine [6], sites in which it has the lowest solubility because of its weakly acidic nature (p*K*a = 3.8).

Over the past three decades, there has been a rapid growth in searching new drug delivery systems aimed to improve the biopharmaceutical properties of orally administered drugs [7]. Recently,

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some types of inorganic materials such as silica derivatives [8,9], carbon materials [10], and layered double hydroxides (hydrotalcite, HTlc) [11–13] have been employed as hosts for poor soluble and/or poor permeable drugs [14,15]. The growing interest toward the materials formed by host inorganic matrices and drug guests has attracted considerable attention as they represent an interesting alternative to organic matrices, which show problems as (i) limited chemical and mechanical stability, (ii) susceptibility to microbiological contamination (for water based matrices) [7], and (iii) low drug loading.

In a previous study, the effect of the layered double hydroxide inorganic material hydrotalcite (HTlc) on FURO dissolution was evaluated. The drug was intercalated into MgAl-HTlc obtaining MgAl-HTlc-FURO, a composite in which FURO (representing the intercalated phase) is homogeneously dispersed, in molecular form, between the HTlc interlamellar spaces (representing the external phase). It must be highlighted that, once intercalated, FURO is not organized as crystals; thus in this new solid state form, its release is enhanced once in contact with the dissolution medium [16]. This is a significant starting point in the development of a new SOD form for FURO oral administration. With regard to this objective, an important aspect that must be taken into account is the choice of the most appropriate dosage form for the composite oral administration. The use of MgAl-HTlc-FURO as simple powder (i.e., unit dose powders or into capsules) in fact, must be avoided as HTlc, because of its physico-chemical properties, is a compactable material [15–17] able to form easily aggregates when is subject to light forces (such as during the capsules filling) as well as during the manufacture processes and under storage conditions too. The formation of aggregates is responsible for the change of the surface area exposed to the dissolution medium, bringing to a reduced and unpredictable drug release, with consequent oral bioavailability impairment.

Taking into account these considerations, a SOD formulation, suitable for MgAl-HTlc-FURO oral administration, was planned. As the purpose was to promote FURO release in the stomach, its preferential absorption site, immediate release tablets, able to disperse finely the intercalation product in gastric fluid, were designed, developed, and studied.

Among the methods available for tablet manufacturing, direct compression (DC) is the most advantageous because it requires less equipment and space, low labor costs, less processing time, and low energy consumption, and moreover, DC is fastest and simplest for tablet manufacturing and protects the drug from heat and moisture [18]. Although DC technique seems quite simple, the selection of appropriate excipients and their amounts in the formulation are crucial steps for the success of tablet formulation. HTlc is a material that finds a wide application in different fields [12,19], showing also a high versatility for DC [15,17,20]. In addition, it has good glidant and lubricant properties, characteristics that make it suitable for the realization of SODs by a simple composition [21]. On the basis of these considerations, the simplest composition that could be used for tablet preparation is represented by the physical blend of MgAl-HTlc-FURO with a filler. However, these formulations do not satisfy the requirement to promote an immediate dispersion of the intercalation product in the gastric fluids as they are unable to disintegrate rapidly. In this context, in order to obtain the rapid MgAl-HTlc-FURO dispersion into gastric fluids, it is useful to introduce a disintegrant in tablet composition. In fact, this is a substance, or a mixture of substances, introduced in the formulation in order to facilitate its breakup and/or disintegration, after administration, with the aim to obtain a rapid drug dissolution [22]. The mechanisms responsible for tablet disintegration are mainly due to the great affinity for water that promotes tablet swelling with following rapid breaking. The disintegration is frequently considered a prerequisite for drug dissolution; however, potential disadvantages can be associated in the use of disintegrant in tablet formulation as (i) high concentration needed for optimum disintegrating efficiency, (ii) poor disintegration, (iii) susceptibility to high compression forces which decrease the efficiency, (iv) poor tableting properties, and (v) decreased disintegrating efficiency in hydrophobic formulations [23]. In recent years, a new disintegrant class, known as super disintegrants, emerged. These excipients have the advantage to improve disintegrant efficiency resulting in lower amounts, typically 2–4% wt./wt., in comparison with traditional disintegrants [22]. The development of immediate release tablets using super disintegrant is interesting in particular for sparingly water soluble drugs for which, when formulated as SOD, the dissolution can be delayed because of poor wettability and/or slow liquid penetration into tablet matrix. Examples of super disintegrants are the following: crospovidones, sodium starch glycolate, cross-linked celluloses, cross-linked polymer, and a cross-linked starch [22]; they are excipients able to be directly compressed without need of solvents.

The objective of this research work was to formulate tablets containing MgAl-HTlc-FURO and a super disintegrants, intended to be swallowed whole and to disintegrate rapidly in the stomach in order to improve FURO release in this site.

2. Materials and methods

2.1. Materials

Sodium starch glycolate (Explotab®) was kindly provided from JRS Pharma (New York, USA); Polyplasdone®INF-10, Polyplasdone®XL, and Polyplasdone®XL-10 were furnished from ISP Chemicals Inc. (Calvert City, USA); hydroxypropyl cellulose low-substituted L-HPC®LH-21 was provided from Shin-Etsu Chemical Co., Ltd. (Tokyo, Japan); lactose (Farmacopea Ufficiale Italiana, XII Edizione (F.U. XII Ed.) and talc were furnished by Comifar (Perugia, Italy); microcrystalline silica was furnished from Farvima (Firenze, Italy). The intercalation product MgAl-HTlc-FURO has been prepared following the procedure described in the previous work [16]. Commercially available furosemide tablets (Lasix® 25 mg) were purchased in pharmacy. Deionized water was obtained by reverse osmosis process with a MilliQ system (Millipore, Roma, Italy). Other reagents and solvents were of reagent grade and were used without further purification. Simulated gastric fluid was prepared, according to F.U. (XII Ed.), by adding 100 μl of HCl 10 M to 400 ml of a NaCl (2%) water solution and adjusting the resulting volume to 1000 ml with deionized water. The phosphate buffer solution of pH 6.8 was prepared according to European Pharmacopoeia (Ph. Eur. VI Ed.), mixing 51 ml of a $K_2H_2PO_4$ water solution (27.2 g/l) to 49 ml of a Na_2HPO_4 water solution (71.6 g/l).

2.2. Determination of MgAl-HTlc flow properties

HTlc (nitrate form) flow properties have been determined by compressibility index and Hausner ratio calculation (F.U. XII Ed.) after previous determination of the apparent volume before and after powder settling (ERWEKA SVM 101/201). The measurements were taken following the procedure reported in F.U. (XII Ed.). A weighted amount of sample was introduced in a graduated dried cylinder (250 ml) in order to occupy a volume between 150 and 250 ml. The volume measured before settling is the bulk volume (V_0). The sample was submitted to a successive tapping (10, 500 and 1250), and the corresponding volumes were measured (V_{10}, V_{500}, and V_{1250}); V_{1250} represents the settled volume (Vf). The measurements were taken in triplicate, each result represents an average of three measurements and the error was expressed as standard

deviation (SD). The compressibility index was calculated following Eq. (1) while the Hausner ratio was calculated by Eq. (2):

$$\text{Compressibility index} = \frac{(V_0 - Vf)}{V_0} * 100 \quad (1)$$

$$\text{Hausner ratio} = \frac{V_0}{Vf} \quad (2)$$

2.3. Manufacturing of tablets

Three different kinds of tablet were prepared by DC, displaying the following compositions:

(1) a homogeneous physical blend of MgAl-HTlc-FURO, a super disintegrant and a filler;
(2) a homogeneous physical blend of crystalline FURO (not intercalated), a super disintegrant and a filler;
(3) a homogeneous physical blend of crystalline FURO (not intercalated) physically mixed to MgAl-HTlc, a super disintegrant and a filler.

Each tablet contained 25 mg of drug and had a final weight of 100 mg as the market product Lasix®. The blends were gently prepared by using pestle and mortar and then compressed by a 6 mm diameter die on an single-punch (steel AISI 300 series) through a manual hydraulic press (PerkinElmer, England), by a compression force of 1×10^3 kg for a total time of 5 s. Tablet thickness was measured ($n = 10$), by a micrometer (Borletti, Milano, Italy).

2.4. Differential Scanning Calorimetry (DSC) analyses

DSC analyses were performed using an automatic thermal analyzer (Mettler Toledo DSC821e) and indium standard for temperature calibrations. Holed aluminum pans were employed in the experiments for all samples, and an empty pan, prepared in the same way, was used as a reference. Samples of 3–6 mg were weighted directly into the aluminum pans, and the thermal analyses of samples were conducted, at a heating rate of 5 °C/min, from 25 to 250 °C.

2.5. Physical characterization: crushing strength

The crushing strength was measured, according to F.U. (XII Ed.) using a hardness tester (instrumented uniaxial press ERWEKA TBH 220). Data are reported as an average of 10 measurements.

2.6. Physical characterization: friability

Friability was determined according to F.U. (XII Ed.) by submitting 65 previously weighed tablets to falling shocks for 4 min in a friabilator (Erweka TA 200), set at 25 rev/min. After 4 min, the tablets were reweighed and the percentage friability was calculated.

2.7. Disintegration time test

The disintegration time was evaluated modifying the disintegration test for dispersible tablets (F.U. XII Ed.) with the only exception that, instead of water, gastric fluid was used as medium. A beaker was filled with 100 ml of simulated gastric fluid of pH 3.0 thermostated at 37 °C ± 0.5. The tablet was carefully put in the beaker, and the time for tablet complete disintegration into fine particles was measured. Each datum represents the average of three measurements ± SD.

2.8. In vitro release studies

FURO in vitro release from tablets was carried out by using the flow-through diffusion cell (F.U. XII Ed.). The test was performed at 37 °C ± 0.5 pumping at 15 ± 1 ml/min a fluid at pH 3.0 ± 0.1 for 2 h, changed with a fluid at pH 6.8 ± 0.1 (USP XX) pumped for further 2 h in order to simulate the formulation passage from the stomach to the first part of intestine. The dosage form containing 25 mg of drug (the same amount of the corresponding market Lasix®) was placed in the dissolution chamber. The samples, collected at predetermined times, were filtered through a cellulose membrane (Filter paper Whatman 41, 20–25 μm, Whatman GmbH, Dassel, Germany), and drug concentration was determined by using a UV–Vis spectrophotometer (Agilent mod. 8453). Drug calibration curves were previously prepared in acidic fluid at pH 3.0 (λ_{max} = 274.0 nm, r = 0.9996) and in phosphate buffer at pH 6.8 (λ_{max} = 276.0 nm, r = 0.9990).

3. Results and discussions

3.1. Tablet preparation and characterization

SOD forms for MgAl-HTlc-FURO oral administration were designed taking into account that they must be able to give a fine dispersion of the intercalation product in the gastric medium in order to promote the rapid drug release in the stomach, site in which FURO is preferentially absorbed [6]. The composite MgAl-HTlc-FURO, whose exact formula is $[Mg_{0.63}Al_{0.37}(OH)_2]$ $(FURO)_{0.27}$ $(OH)_{0.1}\cdot 0.56\ H_2O$, was prepared, as previously described [16], by ion exchange of FURO anions with the precursor MgAl-HTlc in nitrate form, reaching the final drug loading of 55.60% [16].

At the beginning, simple tablets containing only MgAl-HTlc-FURO (45 mg, corresponding to 25 mg of free FURO) and a filler (to reach the final weight of 100 mg) were prepared by DC. However, the resulting formulations had limited disintegration ability as they showed a high hardness mean 91 N determined by crushing strength studies, as reported method section, and long disintegration times (data not reported). Therefore, with the aim to improve tablet disintegration time, new formulations were planned by introducing a disintegrant in tablet composition. These were prepared by DC of a very simple blend constituted by (i) MgAl-HTlc-FURO, (ii) a super disintegrant, and a (iii) filler (to reach the final weight of 100 mg as the market product Lasix®) additionally, similar tablets, containing the crystalline FURO (not intercalated), alone or physically mixed to MgAl-HTlc in place of the intercalation product, were prepared as controls.

The selection of suitable tablet ingredients is one of the crucial steps that could make a success in DC processing because, although DC simplicity and cost-effectiveness, the powder properties can greatly influence blend compaction behavior and tablet performances. DC success might be guaranteed by using ingredients with good flowability and compressibility, low or no moisture sensitivity, low lubricant sensitivity, and good machinability even in high-speed tableting machinery with reduced dwell times [24].

As the ingredients flow properties play a key role in DC successful as well as final tablet characteristics, it was important to know those relative to MgAl-HTlc, in particular its compaction attitude, indirectly assessed in previous studies [15,17,20]. Thus, MgAl-HTlc (nitrate form) flow properties were determined calculating the compressibility index and the Hausner ratio as F.U. XII Ed. prescribes. In order to calculate these two parameters, the powder apparent volume values before and after settling were measured (F.U. XII Ed., see method section). Data obtained were elaborated in Eqs. (1) and (2) obtaining a compressibility index of 19.9 ± 0.854 and a Hausner ratio of 1.24 ± 0.015, data then compared with the values reported in

the flowability scale of F.U.XII Ed. HTlc compressibility index and Hausner ratio resulted included in the range of values, identifying the materials with fair flow character. As suitable tablet for MgAl-HTlc-FURO oral administration must generate a fine dispersion of the intercalation product in the acidic medium, in the choice of the best filler and super disintegrant, two aspects must be considered, (i) powder blends manageability and machinability and (ii) tablets disintegration time.

Among the variety of directly compressible super disintegrants available, sodium starch glycolate (Explotab®), Polyplasdone®INF-10, Polyplasdone®XL, Polyplasdone®XL-10, and hydroxypropyl cellulose low-substituted L-HPC®LH-21 were chosen, examined, and submitted to some compression attempts.

Sodium starch glycolate (Explotab®) is a cross-linked low-substituted carboxymethyl ether of poly-α-glucopyranose (Fig. 1), in which about 15% of the glucose units are carboxymethylated; it is obtained by the suitable treatment of potato starch and possess a particle size average in the range of 35–55 μm [25]. Polyplasdones® (crospovidones) are synthetic, non-ionic, insoluble, cross-linked homopolymers of N-vinyl-2-pyrrolidone [26], characterized by porous particles. Tablet disintegration is connected to its typical particle morphology (Fig. 2) that allows the fast water adsorption generating a rapid volume expansion and hydrostatic pressure responsible for tablet breaking. It is possible to distinguish three different chemical grades: Polyplasdone®XL, Polyplasdone®XL-10, and Polyplasdone®INF 10 characterized by different particle size (100–130 μm; 30–50 μm, and 5–10 μm respectively) (Fig. 2) [26]. L-HPC®LH-21 is a low-substituted hydroxypropyl cellulose, insoluble in water, non-ionic, chemically inert (no interactions with APIs), and low hygroscopic which particles show a moderate fibrous morphology (Fig. 3) and a size ranging 40 μm. L-HPC®LH-21 is a direct compressible material with high disintegrant capacity, due to its swelling action when put in contact with water.

As filler for final tablets preparation, the most common excipients microcrystalline silica, talc, and lactose were selected and examined. Microcrystalline cellulose (MCC) is a purified and partially depolymerised cellulose prepared by treating α-cellulose, obtained as a pulp from fibrous plant material with mineral acids. MCC is widely used as a filler and binder for wet granulation, DC and as a filler for hard gelatin capsules. It has low chemical reactivity combined to excellent compactability at low pressures [27,28]. However, during tablet preparation, the powder blends containing MCC showed low flow properties resulting in a very low manageability additionally, the final tablet resulted irregular due to picking problems, and for this reason, MCC was discarded from the study. These observations are supported from some studies highlighting that, although the large use of MCC in tablet preparation, it has low bulk density, high lubricant sensitivity, poor flow characteristics, and high sensitivity to moisture during the compression procedure [29,30]. Talc is a purified, hydrated, magnesium silicate widely used in oral dosage formulations as lubricant and diluent [31]. The physical blends prepared by using talc were homogeneous and easily manageable; however, some problems have been

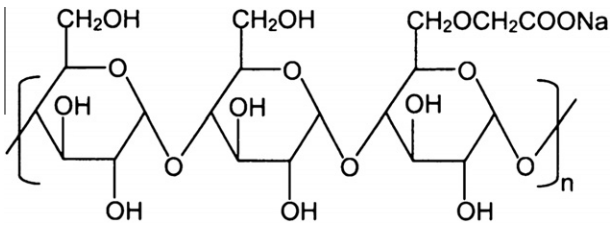

Fig. 1. Sodium starch glycolate chemical structure.

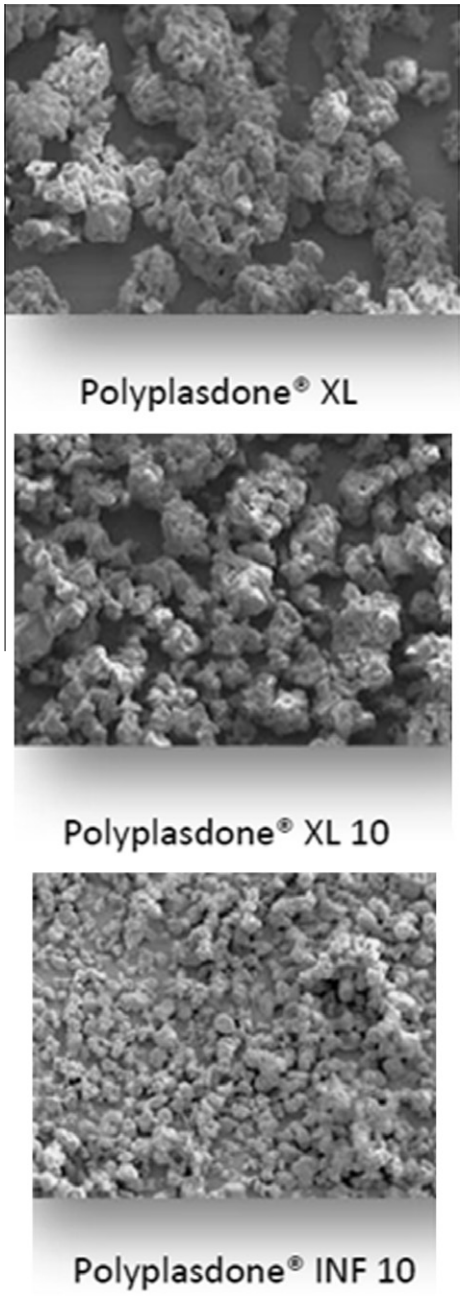

Fig. 2. Particles morphology of the three different Polyplasdones® employed.

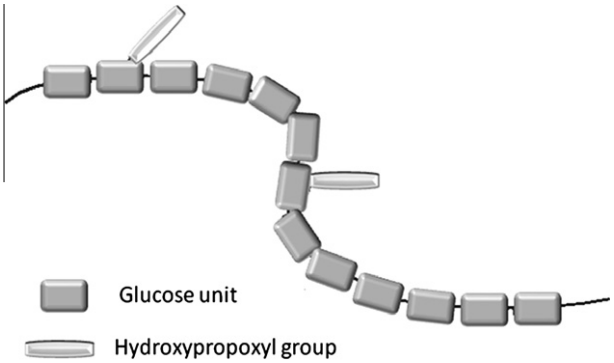

Fig. 3. Schematic representation of L-HPC®LH-21 structure.

Table 1
Tablet compositions and disintegration times.

Tablet	MgAl-HTlc-FURO[a] (mg)	Polyplasdone®INF-10 (mg)	Polyplasdone®XL (mg)	L-HPC®LH-21 (mg)	Lactose (mg)	Disintegration time (n = 3; ±SD)
1#	45	2	–	–	53	38.00″ ± 3.00″
2#	45	3	–	–	52	36.00″ ± 1.00″
3#	45	4	–	–	51	22.00″ ± 3.00"
4#	45	–	2	–	53	41.00″ ± 1.00″
5#	45	–	3	–	52	37.00″ ± 6.00″
6#	45	–	4	–	51	35.00″ ± 3.00"
7#	45	–	–	2	53	>5′
8#	45	–	–	3	52	>5′
9#	45	–	–	4	51	67.00″ ± 5.00″

[a] Corresponding to 25 mg of crystalline FURO.

encountered after the compression as the ejection step resulted difficult due to tablet sticking to the die. For this reason, talc was discarded from the study. Lactose is a naturally occurring simple carbohydrate, disaccharide constituted by D-galactose and D-glucose. It is widely used as filler or filler-binder in the manufacture of pharmaceutical tablets and capsules [32]. The main lactose properties that contribute to its popularity are the cost effectiveness, availability, bland taste, low hygroscopicity, compatibility to active ingredients and other excipients, excellent physical and chemical stability, and water solubility. Lactose resulted the most suitable filler as it confers high homogeneity and easy compressibility to powder blends.

The most suitable super disintegrants for final tablet preparation were firstly selected on the basis of powder blend (constituted by MgAl-HTlc-FURO, lactose and a super disintegrant) manageability and machinability. Explotab® and Polyplasdone®XL-10 were discarded from the study in the preliminary trials as some problems were encountered during the manufacturing procedure. In particular, in both cases, tablet ejection was very difficult due to its sticking to the die resulting in a defective final product. Each of the remaining super disintegrant was employed in three different percentages (2, 3, and 4% wt./wt.) for the preparation of tablets **1#**–**9#** which compositions are reported in Table 1.

Firstly, all the formulations were submitted to in vitro disintegration time tests with the aim to evaluate the minimum amount of super disintegrant able to produce tablet disintegration in short time. This was an essential requirement as the tablet must generate rapidly a MgAl-HTlc-FURO dispersion in acidic fluid keeping, at the same time, the stability to salivary fluid during the passage through the buccal cavity. The last consideration is an essential point because super disintegrants are generally used for orally disintegrating tablets (ODT) preparation [33], for which FDA guidance prescribes disintegration times of approximately 30 s or less [34]. From disintegration time, tests resulted that both tablet groups **1#**–**3#** (in which Polyplasdone®INF 10 is present as super disintegrant) and **4#–6#** (prepared by using Polyplasdone®XL as super disintegrant) disintegrate within 1 min. The disintegration times decrease as super disintegrants percentage increases (Table 1). Regarding the group of tablets prepared with Polyplasdone®INF 10, tablet **3#** showed a disintegration time under the limit value of 30 s [34] and for this reason was discarded from the study. As tablets **1#**–**2#** have similar disintegration times 38.00″ and 36.00″ respectively, the formulation with the lowest amount (2 wt.%) of Polyplasdone®INF 10 (tablet **1#**) was chosen for the next studies. Also in the case of tablets **4#**–**6#** (Polyplasdone®XL as super disintegrant), the increase in super disintegrant percentage reduces the disintegration times (Table 1) which resulted similar for the three tablets: 41.00″ (tablet **4#**), 37.00″ (tablet **5#**), and 35.00″ (tablet **6#**). Among them, tablet **4#**, containing the lowest amount of Polyplasdone®XL (2 wt.%), was chosen for next studies. Regarding the tablets containing L-HPC®LH-21, it was observed that using low percentages, namely 2 wt.% and 3 wt.% (tablet **7#** and **8#** respectively), delayed disintegration times have been registered (>5′) while increasing the amount to 4 wt.% (tablet **9#**), a most rapid tablet breaking can be obtained (67.00″) (Table 1); thus, the latter L-HPC®LH-21 percentage was used for final tablets preparation.

DSC analyses have been performed in order to evaluate the possible interactions among tablet ingredients (MgAl-HTlc-FURO/FURO, filler, superdisintegrant) and possible changes in tablet characteristics. Analyses were carried out on the simple physical blends of tablet ingredients and then after their compression. From obtained data (not reported) resulted that no interactions occurred among tablet ingredients. Moreover, comparing physical mixture thermal profiles, before and after compression, no differences were observed meaning that the compression procedure does not change the characteristics of the native ingredients.

Two important features must be evaluated during tablet design and preparation, (1) the aspects related to patient compliance and (2) final formulation mechanical properties.

Regarding the aspect of patient compliance, tablet thickness was considered and measured on tablets **1#**, **4#**, and **9#**, resulting in the regular size with a thickness of 2 mm (Table 2). Concerning the formulation mechanical properties feature, tablets must possess adequate strength to withstand mechanical shocks of handling in manufacturing, packaging, and shipping as well as the resistance to surface abrasion. These properties were determined by submitting tablets **1#**, **4#**, and **9#** to friability and crushing strength tests F.U. (XII Ed.). All the compositions showed the capability to confer similar crushing strength and low friability to final tablet (Table 2). The latter is expressed as weight loss percentage, calculated by Eq. (3) where W_1 and W_2 represent tablets weight before and after the test; in all cases, a weight loss less than 1% (limit value) was measured.

$$\text{weight loss}\% = \frac{(W_1 - W_2)}{W1} * 100 \quad (3)$$

Friability values below 1% indicate a good tablet mechanical resistance confirmed also by the crushing strength force values. This is an important tablet characteristic that can guarantee tablet integrity during their preparation, storage, transport, and also at the moment of tablet removal from blister.

3.2. In vitro release studies

The dissolution test was performed by using the flow through diffusion cell method (F.U. XII Ed.), pumping acidic medium at pH 3.0 for 2 h, and then changed with phosphate buffer at pH 6.8 pumped for further 2 h, in order to simulate the formulation transit from the stomach to the intestine. In order to evaluate the real contribution of the composite to drug release from tablets **1#**, **4#,** and **9#**, for each of them two tablets (as controls) were prepared by using (i) crystalline FURO physically mixed to MgAl-HTlc or (ii) crystalline FURO alone instead of MgAl-HTlc-FURO.

Table 2
Tablet thickness and mechanical properties.

Tablet	Super disintegrant (%)	Thickness (mm ± SD, n = 10)	Crushing strength force (N, n = 10)			Friability (weight loss %, n = 65)
			Max	Min	Mean	
1#	2	2.00 ± 0.10	99.00	62.00	75.40	0.61
4#	2	2.00 ± 0.10	92.00	44.00	69.00	0.43
9#	4	2.00 ± 0.10	82.00	47.00	67.10	0.40

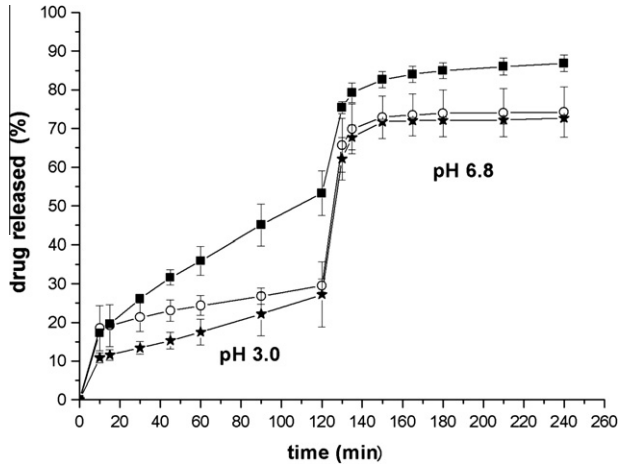

Fig. 4. In vitro release profiles of tablet **#1** (■), in which the drug is present as MgAl-HTlc-FURO, compared with that obtained from the tablets having the same compositions but containing, FURO alone (★) or physically mixed to MgAl-HTlc (°) instead of the intercalation product. The experiment was conducted pumping at 15 ± 1 ml/min a fluid at pH 3.0 ± 0.1 for 2 h, changed with a fluid at pH 6.8 ± 0.1 (USP XX) pumped further for 2 h in order to simulate the passage of the formulation from stomach to the first part of intestine (n = 5, mean ± SD).

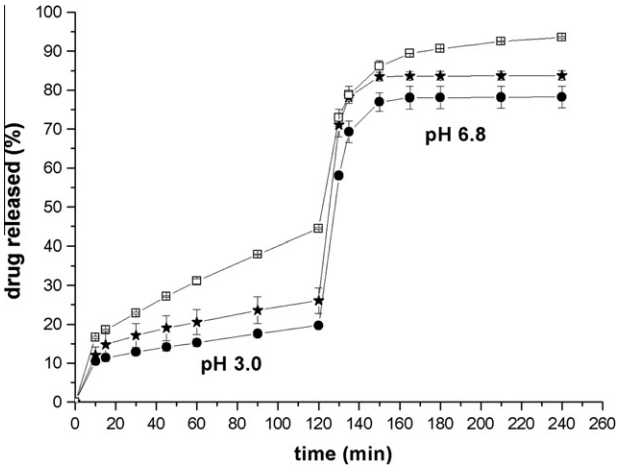

Fig. 5. In vitro release profiles of tablet **#4** (□), in which the drug is present as MgAl-HTlc-FURO, compared to that obtained from the tablets having the same compositions but containing FURO alone (★) or physically mixed to MgAl-HTlc (●) instead of the intercalation product. The experiment was conducted at 37 °C ± 0.5 pumping at 15 ± 1 ml/min a fluid at pH 3.0 ± 0.1 for 2 h, changed with a fluid at pH 6.8 ± 0.1 (USP XX) pumped for further 2 h in order to simulate the passage of the formulation from stomach to the first part of intestine (n = 5, mean ± SD).

Fig. 4 shows the release profiles of tablets containing Polyplasdone®INF 10 as super disintegrant. The main differences among the three kinds of formulation could be mainly highlighted at pH 3.0. After the first 15 min, lapse of time in which the amount of FURO dissolved is rather similar for the three formulations (tablet **1#** and the two controls), the amount of drug released from tablet **1#** (containing MgAl-HTlc-FURO) increases reaching 53% vs. 27% (FURO alone) and 29% (FURO/MgAl-HTlc physical mixture) after 2 h. Thus, the prevalence of drug released from tablet **1#**, compared to the controls, could be clearly recognized. After 2 h, the dissolution medium is changed with phosphate buffer (pH 6.8). Also in these conditions, tablet **1#** produces the better release profiles in comparison with the controls reaching 86% vs. 73% (FURO alone) and 74% (FURO/MgAl-HTlc-NO_3 physical mixture); however, the differences are less marked than the acidic conditions as FURO displays a better solubility at pH 6.8 [35].

Comparing the dissolution profiles of the formulations prepared with Polyplasdone®XL as super disintegrant, the improved FURO release from tablet **4#**, containing the composite MgAl-HTlc-FURO, can be observed (Fig. 5). The drug release at pH 3.0 from tablet **4#** is slow during the first 15 min, resulting similar to that coming from the two controls, and then it increases reaching after 2 h a maximum of 44% vs. 26% (FURO alone) and 20% (FURO/MgAl-HTlc physical mixture). Also at pH 6.8, the amount of FURO released from tablet **4#** prevails obtaining an amount of drug dissolved of 94% vs. 83% (FURO alone) and 80% (FURO/MgAl-HTlc physical mixture) at the end of the first part of the experiment (120 min).

Regarding the tablets containing L-HPC®LH-21, results that the amount of drug released from tablet **9#** exceeds that coming from the two controls reaching after 2 h 63% vs. 28% (FURO alone) and 38% (FURO/MgAl-HTlc physical mixture) (Fig. 6). Also at pH 6.8,

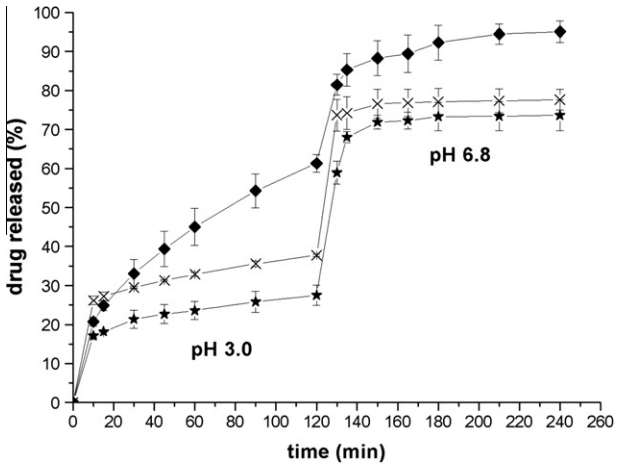

Fig. 6. In vitro release profiles of tablet **#9** (♦), in which the drug is present as MgAl-HTlc-FURO, compared to that obtained from the tablets having the same compositions but containing FURO alone (★) or physically mixed to MgAl-HTlc (×) instead of the intercalation product. The experiment was conducted at 37 °C ± 0.5 pumping at 15 ± 1 ml/min a fluid at pH 3.0 ± 0.1 for 2 h, changed with a fluid at pH 6.8 ± 0.1 (USP XX) pumped for further 2 h in order to simulate the passage of the formulation from stomach to the first part of intestine (n = 5, mean ± SD).

tablet **9#** shows the best performances giving a final release of 95% vs. 73% (FURO alone) and 77% (FURO/MgAl-HTlc physical mixture) at the end of the first 2 h.

In all cases, the formulations containing the intercalation product MgAl-HTlc-FURO (tablet **1#**, **4#** and **9#**) promote a better and higher drug release than the corresponding controls. In regard to

this consideration, it should be underlined the importance in drug intercalation into HTlc galleries instead of the simple physical blending. In fact, as observed from the release profiles of the FURO/MgAl-HTlc physical mixtures (Figs. 4–6), FURO dissolution from the simple drug/HTlc blend is similar to that deriving from drug alone, and this is explained considering that FURO is present in the controls in crystalline form. Thus, drug dissolution is strongly influenced from crystal lattice breaking, process that takes place slowly because of poor FURO solubility. On the other hand, the drug dissolution improvement from tablets containing the composite MgAl-HTlc-FURO is explained considering that FURO intercalation into HTlc generates a new product in which the drug is not present as crystals (physical state in which it is present in the two controls) [16], whose disruption is the rate determining step of drug dissolution, but is homogeneously dispersed in molecular form into the nanospaces of HTlc interlayer region generating a "shell-liquid state" [19]. Thus, the crystal lattice breaking is bypassed and drug dissolution takes place rapidly. The drug is released from MgAl-HTlc-FURO following two mechanisms: (1) by HTlc dissolution as it is noteworthy that at low pH values (<4) HTlc undergoes to gradual destruction [36,37] favoring the release of the intercalated drug molecules; (2) (from the not dissolved composite) by ion exchange with anions present in the dissolution medium, generally Cl^- (at pH 3.0) and PO_4^{2-} (at pH 6.8).

Through the careful analysis of tablets **1#**, **4#**, and **9#** release profiles and of their corresponding controls (Figs. 4–6), three important steps could be distinguished: (1) from zero to 30 min; (2) from 30 to 120 min; and (3) from 120 to 240 min. In the first step (1), in all cases, a low amount of released drug is registered from the three different tablets (MgAl-HTlc-FURO, FURO alone or physically mixed to MgAl-HTlc). In the case of tablets containing FURO alone and physically mixed to MgAl-HTlc, these results are explained considering that the drug, present in crystalline form, shows a low solubility in acidic conditions; thus, crystal lattice disruption takes place slowly. In the case of MgAl-HTlc-FURO tablets in the first minutes, drug is released by ion exchange mechanism as HTlc solubilization occurs gradually. This conclusion can be confirmed from the analysis of the second step (2), showing that the amount of FURO released from tablets **1#**, **4#**, and **9#** increases in comparison with the corresponding controls obtaining in all cases an higher amount of dissolved drug at pH 3.0. The best performances of tablets containing MgAl-HTlc-FURO could be evidenced in the third step (3) too, however, as FURO results more soluble at pH 6.8, the differences with controls are less marked.

It is important to note that, comparing FURO release profiles from tablets **1#**, **4#,** and **9#**, some differences could be recognized. Since the formulation composition plays an important role in the composite liberation and drug release, it was interesting to investigate the effect of the three super disintegrants on these two processes.

The amount of MgAl-HTlc-FURO available to the gastric fluids is strictly related to the dosage form disintegration rate and to the size of the particles deriving from this process. After administration, the dosage form gets in contact with the gastric fluid and disintegrates, generating large particles which must deaggregate to yield fine particles. In fact, fine particles offer a large surface area exposed to gastric fluid giving rise to a high amount of drug dissolved and available for absorption. Comparing FURO release profiles of tablets **1#** and **4#**, it might be hypothesized that the differences are related to the different size of primary particles generated after disintegration. These differences could affect drug dissolution since breaking tablets into smaller particles may promote drug dissolution by providing larger total surface areas for drug dissolution to take place. As shown in the pictures reported in Fig. 7, tablet **1#** (containing Polyplasdone®INF 10) breaking (Fig. 7) generates smaller particles if compared with tablet **4#** (containing Polyplasdone®XL) (Fig. 7). In the first case, the intercalation product is better dispersed in the dissolution medium in fact, and the small particle size guarantees a high surface area that increases interfacial activity enhancing drug dissolution. Regarding tablet **9#** (containing L-HPC®LH-21), presenting the better drug release in comparison with formulations **1#** and **4#**, the particles generated after the contact with the gastric medium are fine (Fig. 7), offering a high surface area exposed to dissolution. The differences in the disintegration behavior could also be connected to the different tablet wettability. In particular, it was demonstrated that L-HPC®LH-21 is able to absorb a large amount of water and in a lower time than Polyplasdones®, allowing a rapid water penetration and contact with the intercalation product [38].

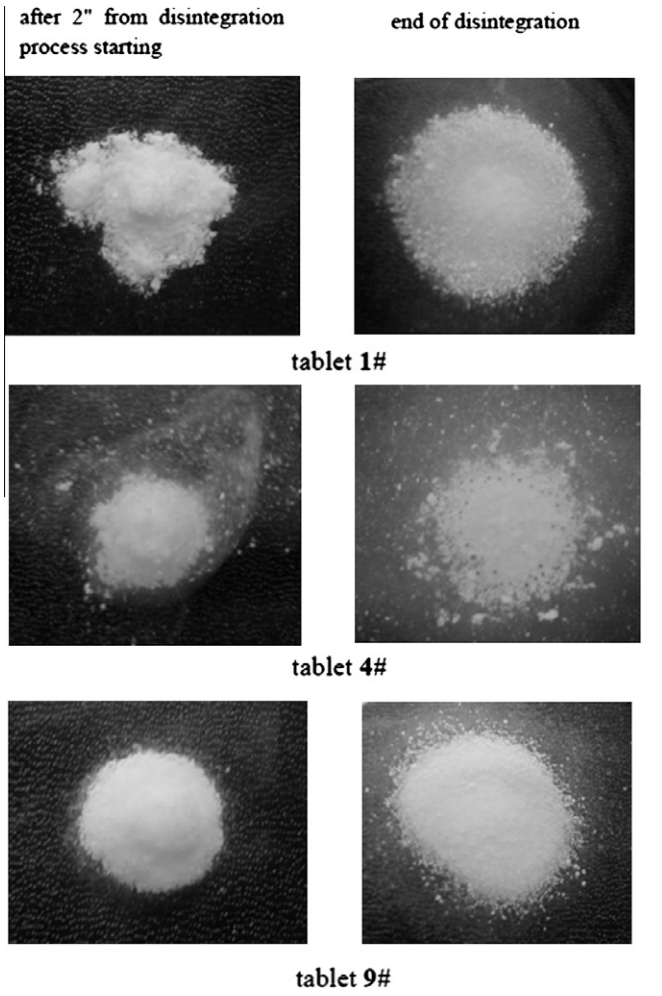

Fig. 7. Disintegration process of tablets #1, #4, and #9 after 2 s from disintegration starting (left) and at the end of the process (right).

Because of its weak acidity (p*K*a = 3.8), FURO shows the lowest solubility at low pH values of the stomach that in turn represents its preferential absorption site [6]; moreover, it was observed that at pH 3.0, FURO shows the most rapid absorption [39]. For this reason, a formulation able to release the drug preferentially in acidic conditions should be necessary. Comparing the amount of drug released from tablets **1#**, **4#**, and **9#**, the best results, in terms of higher amount of FURO released at pH 3.0, were observed for tablet **9#** that was chosen to be compared with the commercial product Lasix®. As is clearly evidenced in Fig. 8, FURO is released gradually from tablet **9#**, according to the mechanism of drug release from HTlc; after 2 h in acidic conditions, the total amount of drug dissolved resulted of 63% vs. the 49% of the commercial product. This

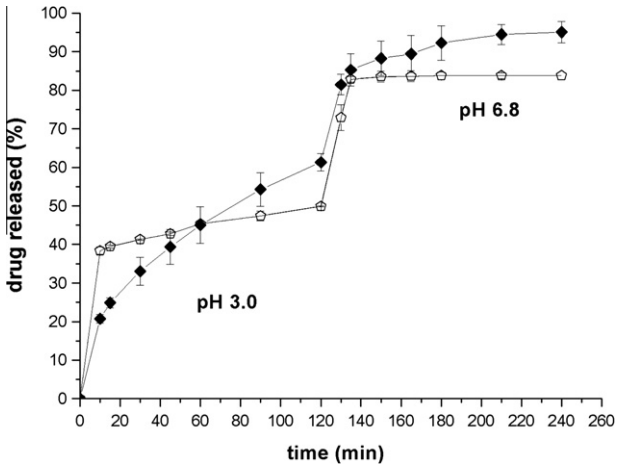

Fig. 8. In vitro release profile obtained from tablet **#9** prepared with MgAl-HTlc-FURO (♦) and from the market tablets Lasix® (⬠). The experiment was conducted at 37 °C ± 0.5 pumping at 15 ± 1 ml/min a fluid at pH 3.0 ± 0.1 for 2 h, changed with a fluid at pH 6.8 ± 0.1 (USP XX) pumped for further 2 h in order to simulate the passage of the formulation from stomach to the first part of intestine (n = 5, mean ± SD).

aspect is important and must be underlined as, and in comparison with the commercial product, tablet **9#** is able to produce a better and higher FURO release in acidic fluids resulting suitable to promote its release in the preferential absorptive region, the stomach.

The difference was maintained at pH 6.8 too where, after further 2 h, the amount of FURO released was 95% in comparison with the commercial product, with a total amount released of 83%.

4. Conclusions

The problems of low and variable bioavailability of FURO (class IV BCS) could be solved improving its biopharmaceutical properties. With this aim, a new formulation was proposed combining two technological approaches: the intercalation of FURO into the inorganic matrix hydrotalcite and its formulation as immediate release tablet in order to improve the release in the stomach, the preferential absorption region of this drug. With this aim, new super disintegrants, at different percentages, have been chosen and studied.

Results coming from dissolution assays highlighted the advantages in the use of the composite MgAl-HTlc-FURO instead of the crystalline drug; additionally, the tablets prepared by using L-HPC®LH-21 gave the higher release at pH 3.0 in comparison with the tablets prepared with other super disintegrants as well as the commercial tablets Lasix®.

The solid oral dosage form proposed represents an interesting tool to improve FURO biopharmaceutical properties because of the in vitro high and almost complete drug release observed. This means that the combination of the composite MgAl-HTlc-FURO with a suitable super disintegrant produces a synergic effect responsible for FURO release improvement.

Moreover, the drug confinement into an inorganic material offers advantages in terms of molecule isolation from the environment and improvement of long-term stability and storage, especially since many molecules are instable in certain conditions [40].

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Appendix A. Supplementary material

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.ejpb.2011.12.011.

References

[1] K. Marshall, E.M. Rudnic, in: G.S. Banker, C.T. Rhodes (Eds.), Drugs and the Pharmaceutical Sciences: Modern Pharmaceutics, vol. 121, Marcel Dekker, New York, 1990, pp. 23–66.
[2] N.Y. Golovenko, I.Y. Borisyuk, The biopharmaceutical classification system-experimental model of prediction of drug bioavailability, Biochem. Suppl. Ser. B: Biomed. Chem. 2 (2008) 235–244.
[3] R. Panchagnula, N.S. Thomas, Biopharmaceutics and pharmacokinetics in drug research, Int. J. Pharm. 201 (2000) 131–150.
[4] D.J. Hauss, Oral lipid-based formulations, Adv. Drug Deliv. Rev. 59 (2007) 667–676.
[5] M. Lindenberg, S. Kopp, J.B. Dressman, Classification of orally administered drugs on the World Health Organization model list of the essential medicines according to the biopharmaceutics classification system, Eur. J. Pharm. Biopharm. 58 (2004) 265–278.
[6] S.S. Davis, Formulation strategies for absorption windows, Drug Discov. Today 10 (2005) 249–257.
[7] I. Gomez-Orellana, Strategies to improve oral drug bioavailability, Expert Opin. Drug Deliv. 2 (2005) 419–433.
[8] T. Heikkilä, J. Salonen, J. Tuura, N. Kumar, T. Salmi, D.Y. Murzin, M.S. Hamdy, G. Mul, L. Laitinen, A.M. Kaukonen, J. Hirvonen, V.-P. Lehto, Evaluation of mesoporous TCPSi, MCM-41, SBA-15, and TUD-1 materials as API carriers for oral drug delivery, Drug Deliv. 14 (2007) 337–347.
[9] V. Ambrogi, L. Perioli, C. Pagano, L. Latterini, F. Marmottini, M. Ricci, C. Rossi, MCM-41 for furosemide dissolution improvement, Micropor. Mesopor. Mater. 147 (2012) 343–349.
[10] N.A. Ochekpe, P.O. Olorunfemi, N.C. Ngwuluka, Nanotechnology and drug delivery Part 1: background and applications, Trop. J. Pharm. Res. 8 (2009) 265–274.
[11] C. Del Hoyo, Layered double hydroxides and human health: an overview, Appl. Clay Sci. 36 (2007) 103–121.
[12] J.H. Choy, S.J. Choi, J.-M. Oh, T. Park, Clay minerals and layered double hydroxides for novel biological applications, Appl. Clay Sci. 36 (2007) 122–132.
[13] L. Tammaro, U. Costantino, A. Bolognese, G. Sammartino, G. Marenzi, A. Calignano, S. Tetè, F. Mastrangelo, L. Califano, V. Vittoria, Nanohybrids for controlled antibiotic release in topical applications, Int. J. Antimicrob. Agents 29 (2007) 417–423.
[14] V. Ambrogi, G. Fardella, G. Grandolini, M. Nocchetti, L. Perioli, Effect of hydrotalcite like-compounds on the aqueous solubility of some poorly water soluble drugs, J. Pharm. Sci. 92 (2003) 1407–1418.
[15] L. Perioli, V. Ambrogi, L. di Nauta, M. Nocchetti, C. Rossi, Effects of hydrotalcite-like nanostructured compounds on biopharmaceutical properties and release of BCS class II drugs: the case of flurbiprofen, Appl. Clay Sci. 51 (2011) 407–413.
[16] L. Perioli, V. Ambrogi, M. Nocchetti, M. Sisani, C. Pagano, Preformulation studies on host-guest composites for the oral administration of BCS class IV drugs: HTlc and furosemide, Appl. Clay Sci. 53 (2011) 696–703.
[17] R.F. Stockel, Bisphosphonates Inorganic Carriers, US Patent 0013893A1, 2006.
[18] G.K. Bolhus, Z.T. Chowhan, in: G. Alderborn, C. Nyström (Eds.), Pharmaceutical Powder Compaction Technology, Marcel Dekker, New York, 1996, p. 420.
[19] U. Costantino, V. Ambrogi, M. Nocchetti, L. Perioli, Hydrotalcite-like compounds: versatile layered hosts of molecular anions with biological activity, Micropor. Mesopor. Mater. 107 (2008) 149–160.
[20] L. Perioli, V. Ambrogi, S. Giovagnoli, M. Ricci, P. Blasi, C. Rossi, Mucoadhesive bilayered tablets for buccal sustained release of flurbiprofen, AAPS PharmSciTech 8 (3) (2007) E1–E8. <http://www.aapspharmscitech.org> (Article 54).
[21] U. Costantino, M. Nocchetti, in: V. Rives (Ed.), Layered Double Hydroxides and their Intercalation Compounds in Photochemistry and Medicinal Chemistry, Layered Double Hydroxides: Present and Future, Nova Science Publishers, New York, 2001, pp. 435–468.
[22] E.M. Rudnic, J.B. Schwartz, in: D.B. Troy (Ed.), Remington. The Science and Practice of Pharmacy, 21st ed., Lippincott Williams & Wilkins, Baltimore, 2005, p. 893.
[23] M. Andries, S. Mingna, M.M. De Villiers, Effect of compression force, humidity and disintegrant concentration on the disintegration and dissolution of directly compressed furosemide tablets using croscarmellose sodium as disintegrant, Trop. J. Pharm. Res. 2 (2003) 501–504.
[24] S.K. Nachaegari, A.K. Bansal, Coprocessed excipients for solid dosage forms, Pharm. Technol. 1 (2004) 52–64.
[25] A.H. Kibbe (Ed.), Handbook of Pharmaceutical Excipients, third ed., American Pharmaceutical Association, Washington, USA, 2000, pp. 501–504.
[26] A.H. Kibbe (Ed.), Handbook of Pharmaceutical Excipients, third ed., American Pharmaceutical Association, Washington, USA, 2000, pp. 163–164.
[27] G. Shlieout, K. Arnold, G. Müller, Powder and mechanical properties of microcrystalline cellulose with different degrees of polymerization, AAPS PharmSciTech 3 (2) (2002) 45–54. <http://www.aapspharmscitech.org> (Article 11).
[28] P.M. Fechner, S. Wartewig, M. Füting, A. Heilmann, R.H.H. Neubert, P. Kleinebudde, Properties of microcrystalline cellulose and powder cellulose after extrusion/spheronization as studied by Fourier transform Raman

spectroscopy and environmental scanning electron microscopy, AAPS PharmSci 5 (4) (2003) 1–13. <http://www.aapspharmsci.org> (Article 31).
[29] M.J. Tobyn, G.P. McCarthy, J.N. Staniforth, S. Edge, Physicochemical comparison between microcrystalline cellulose and silicified microcrystalline cellulose, Int. J. Pharm. 169 (1998) 183–194.
[30] A. Nokhodchi, An overview of the effect of moisture on compaction and compression, Pharm. Technol. 29 (2005) 46–66.
[31] A.H. Kibbe (Ed.), Handbook of Pharmaceutical Excipients, third ed., American Pharmaceutical Association, Washington, USA, 2000, pp. 555–556.
[32] M. Jivraj, L.G. Martini, C.M. Thomson, An overview of the different excipients useful for the direct compression of tablets, PSTT 3 (2000) 58–63.
[33] R. McLaughlin, S. Banbury, K. Crowley, Orally disintegrating tablets. The effect of recent FDA guidance on ODT technologies applications, Pharm. Technol. (Suppl.) (2009).
[34] US Department of Health and Human Services Food and Drug Administration Center for Drug Evaluation and Research (CDER), Guidance for Industry Orally Disintegrating Tablets, 2008.
[35] G.E. Granero, M.R. Longhi, M.J. Mora, H.E. Junginger, K.K. Midha, V.P. Shah, S. Stavchansky, J.B. Dressman, D.M. Barends, Biowaiver monographs for immediate release solid oral dosage forms: furosemide, J. Pharm. Sci. 99 (2010) 2544–2556.
[36] A.I. Khan, D. O'Hare, Intercalation chemistry of layered double hydroxides: recent developments and applications, J. Mater. Chem. 12 (2002) 3191–3198.
[37] M. Jobbágy, A.E. Regazzoni, Dissolution of nano-size Mg–Al–Cl hydrotalcite in aqueous media, Appl. Clay Sci. 51 (2011) 366–369.
[38] J. Fukami, E. Yonemochi, Y. Yoshihashi, K. Terada, Evaluation of rapidly disintegrating tablets containing glycine and carboxymethylcellulose, Int. J. Pharm. 310 (2006) 101–109.
[39] V.S. Chungi, L.W. Dittert, R.B. Smith, Gastrointestinal sites of furosemide absorption in rats, Int. J. Pharm. 4 (1979) 27–38.
[40] L. Perioli, V. Ambrogi, C. Pagano, M. Nocchetti, G. D'Alba, C. Rossi, Farmaci di classe IV del BCS: un approccio tecnologico-formulativo per migliorare le proprietà biofarmaceutiche, 50° Simposio AFI 23, 2010 pp. 277–278.