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SOME APPLICATIONS OF CURRENT ORGANIC MOLECULES FOR ORAL DRUG DELIVERY

D. Dupeyrón[1,2], J. Rieumont[3,4] and J.C.T. Carvalho[1]

[1]Laboratório de Pesquisa em Fármacos, Universidade Federal do Amapá, Rod. JK, km 2, 68902-380, Macapá-AP, Brasil
[2]Pós-Graduação em Biodiversidade Tropical, Universidade Federal do Amapá, Rod. JK, km 2, 68902-380, Macapá-AP, Brasil
[3]Facultad de Química, Universidad de la Habana, Esq. Zapata y G. 10400, Ciudad Habana, Cuba
[4]Instituto de Ciencia y Tecnología de Materiales, Universidad de la Habana, Esq. Zapata y G. 10400, Ciudad Habana, Cuba

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Abstract. Over the past 15 years, development of polymeric entities and their applications as drug delivery systems have been a promising alternative to overcome protein and anti-inflammatory oral administration drawbacks. Designing oral protein delivery systems has been a persistent challenge to scientists due to their instability in gastrointestinal fluids and poor absorption. Proteins would be denatured by enzymatic action or even by the acidic pH in the stomach, which results in the loss of biological activity. On the other hand, anti-inflammatories do not present stability problems but serious side effects, like stomach ulcers, caused in patients. In this regard, some pharmaceutical approaches such us molecules that act as carriers for proteins, bioadhesive systems and pH sensitive systems have been used with different degrees of success. Thus, the present mini-review discuss these approaches and details the latest state of pH-sensitive polymers development as drug delivery systems. It also shows the increasing interest in block copolymers and enteric nanoparticles structures and their potential applications in oral delivery of proteins and anti-inflammatories.

1. INTRODUCTION

Drugs can be delivered to patients by more than one route, but oral formulation is still the most commonly used in pharmaceutical industry. Oral drug delivery, although attractive compared to injections, cannot be utilized for the protein administration due to the poor epithelial permeability and enzymatic degradation within the gastrointestinal tract [1]. Also, other drugs such as peptides [2] and antigens [3] have shown some difficulties in oral administration. In this sense, more emphasis has been given to improve drug delivery systems since development of new active pharmaceutical ingredients is a complex process. Despite of several advances on drug delivery, oral administration of proteins still remains not only a great challenge but also an attractive research field.

Among proteins, insulin has been thoroughly studied since it is the most effective drug for the treatment of advanced-stage diabetes (Type 1) [4]. Several approaches have been proposed to increase insulin oral bioavailability, including permeation enhancers [5], enzyme inhibitors [6], encapsulation technologies such as nanospheres [7], hydrogels [8], microemulsions [9], and liposomes [10]. Permeation enhancers, such as bile salts and fatty acids, increase epithelial cell permeability of the gastrointestinal tract, increasing oral bioavailability [11,12]. Interestingly, some poly(acrylates) such as polycarbophil and chitosan derivatives have shown

Corresponding author: D. Dupeyrón, e-mail: danaydm@gmail.com

to act as both permeation enhancers and enzyme inhibitors [13]. Poly(acrylates) are able to enhance paracellular epithelial transport by loosening the tight junctions of the mucosal epithelium [14] and inhibit calcium-dependent enzymes such as trypsin by competitively binding to local calcium ions [15]. Other delivery strategies have been primarily focused on utilization of encapsulation technologies [7]. Micro and nanospheres can protect proteins from enzymatic degradation in the intestine, while nanospheres can further facilitate protein transport across the epithelium by way of the Peyer's patches that line the intestine [11].

Another problem in oral drug administration has been associated to the use of Non-Steroidal Anti Inflammatory Drugs (NSAIDs), known as the most prescribed anti-inflammatories. Conventional NSAIDs, such as aspirin, diclofenac, ibuprofen, and others, are reported to inhibit both the COX-1 and COX-2 forms of the enzyme cyclo-oxygenase (COX). The dual COX inhibitory effect of conventional NSAIDs results in gastrointestinal side effects due to the COX-1 is associated to the protection of the gastric mucosa against gastric juice and the coagulation. In the 90th, appears a new generation of anti-inflammatories, called COX-2 inhibitors, such as celecoxib, valdecoxib and rofecoxib. These drugs promise action against pain and fever without the side effects of the previous generation [16]. However, some recent studies have focused on the cardiac toxicity of these COX-2 inhibitors, related to an increased risk of myocardial infarction [17]. Hence, NSAIDs called COX 1 inhibitors are still the most used. Nevertheless, in order to reduce gastric side effects improved formulations of these anti-inflammatories are needed.

The aforementioned highlights the biggest challenges to obtain successful oral drugs formulations. The first problem mentioned is associated to the instability and poor absorption of proteins and the second one to the adverse effects of anti-inflammatories (when use for a long period). In this regard, some pharmaceutical approaches such us molecules that act as carriers for proteins, bioadhesive particles, bioadhesives patches, and pH sensitive polymers have been previously considered with different degrees of success. Thus, the present mini-review discuss these approaches and details the latest state of the development of pH-sensitive polymers as oral drug delivery systems. It also shows the increasing interest in block copolymers and enteric nanoparticles structures and their potential applications in oral delivery of proteins and anti-inflammatories.

2. PHARMACEUTICAL APPROACHES

2.1. Carrier molecules

Progress on oral drug delivery relies not only on the invention of novel processes but also on the development of new molecules, like cyclodextrins, that can serve as drug carriers. The most common pharmaceutical application of these new materials is to enhance the solubility, stability, safety and bioavailability of drugs [18,19].

Cyclodextrins are a family of cyclic oligosaccharides with a hydrophilic outer surface and a lipophilic central cavity. In the pharmaceutical industry they are used as complexing agents to increase the aqueous solubility of poorly soluble drugs and to increase their bioavailability and stability [19]. In addition, cyclodextrins can be used to reduce gastrointestinal irritation, convert liquid into powder (microcrystalline or amorphous), and prevent drug-drug or drug-excipient interactions [19].

Moreover, studies have described the synthesis and pharmacokinetics of a number of low molecular weight compounds that enable the oral delivery of some drugs [20]. When a solution of carrier and either recombinant human α-interferon, insulin or recombinant human growth hormone is orally administered in rats or primates significant serum concentrations of the proteins are detectable. Proteins can be reversibly destabilized, favoring a partially unfolded conformation, due to the interaction between them and carrier molecules [20]. These intermediate conformations are able to be absorbed through the intestinal tissue. The binding force of the carriers to the partially unfolded proteins is relatively weak and the activity of the proteins seems to be unaffected. As an example, development of molecules that can carry large molecules across the membrane of the gastrointestinal tract such as hydroxyl-benzyl aminophenyl butyric acid has been reported [20,21]. Some of these carriers are shown in Fig. 1.

2.2. Bioadhesive systems

Bioadhesion or mucoadhesion is the process whereby synthetic and natural polymers adhere to mucosal surfaces in living organism [22]. Different polymers such as the chitosans, carbopols and carbomers have been used as bioadhesive materials [23]. For most polymers, the molecular weight has a decisive effect on the physical penetration and subsequent entanglement of the polymer with the substrate. Interpenetration and entanglement of an

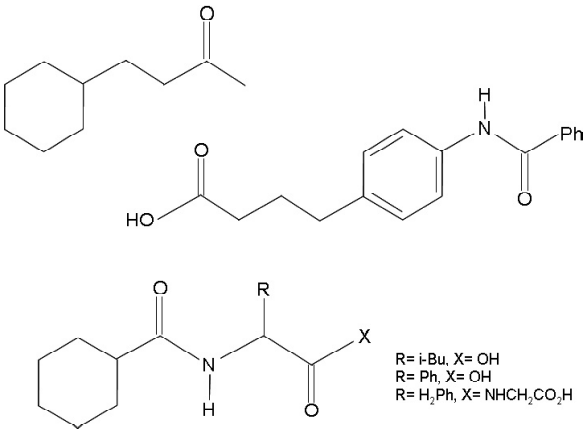

Fig. 1. Some carrier molecules for proteins (adapted from [20]).

adhesive polymer with a mucin substrate is partly responsible for its bioadhesive strength [24].

Mucoadhesive systems can prolong the residence time of the drug at the absorption site and facilitates the contact between the drug and the absorption surface, contributing to a better therapeutic performance of the drug [22]. Recent advances in polymer science and drug delivery have shown the development of novel drug carriers such as bioadhesive particles or bioadhesive patches that have enhanced the use of "bioadhesion" in drug delivery [23-25].

2.2.1. Bioadhesive particles

Several bioadhesive systems such us micro and nano polymeric particles have been developed to enhance the oral bioavailability of drugs [25-27]. Bioadhesive microparticles that adhere to the intestinal membrane have shown strong interactions with gastrointestinal mucus and cellular linings. These particles can traverse both the mucosal absorptive epithelium and the follicle-associated epithelium covering the lymphoid tissue of Peyer's patches. For instance, the absorption of three model substances of widely different molecular size (dicumarol, insulin and plasmid DNA) is increased by these systems [6].

Chitosan, considered a nontoxic and biodegradable polymer, has been widely used not only to obtain nanoparticles [26] but also to obtain modified liposomes with better mucoadhesive properties [27]. Modified liposomes have been developed by anchoring the polymer chitosan-thioglycolic acid (chitosan-TGA) to the liposomal surface to target intestinal mucosal membranes. In these systems, Gradauer et al have been established the formation of a covalent thioether bond between chitosan-TGA and the liposome, in contrast to the common coating procedure, in which polymers are attached to liposomes by ionic interactions [27]. *In vitro* mucoadhesion studies of thiomer-coated and uncoated liposomes showed that the residence time in the porcine small intestine could be almost doubled by the addition of the polymeric layer around the liposome (coated liposomes) [27]. Others studies have used different molecular weight chitosans, with different degrees of deacetylation, producing nanoparticles of varying sizes and charge ratios, affecting the levels of protein release [26]. In spite of many advances, this field has to progress beyond animal models and prove efficacy in humans.

2.2.2. Bioadhesive patches

Bioadhesive intestinal patches are a novel drug delivery system used to release drugs into systemic circulation [28-30]. Intestinal patches can localize drugs, such us insulin, near the mucosa inducing greater levels of absorption and stability at the intestinal epithelium. *In vivo* experiments in rats, performed via jejunal administration, showed that intestinal insulin patches with doses in the range of 1-10 U/kg induced dose-dependent hypoglycemia in normal rats, with a maximum drop in blood glucose levels of 75% (at 10 U/kg) [30]. Intestinal patches could not only offer a new methodology for the oral delivery of insulin, but for various other molecules, including heparin, growth hormones and vaccines [30]. Even though there is still a limited number of studies in this area, bioadhesive patches seem to be promising devices to improve proteins and peptides absorption [29].

2.3. pH-sensitive copolymeric systems

Over the past 15 years, the fast expansion of pH-sensitive polymeric systems has facilitated the development of several structures such as micelless, cross-linked micelles, hydrogels, copolymer nanoparticles [31]. Specifically, pH-sensitive copolymeric systems can be used not only for protein protection but also to minimize side effects of conventional NSAID formulations.

Copolymer is a polymer derived from two (or more) monomer units (for instance A and B units). Copolymers can be classified as alternating, random, block, graft and others; based on how those units are arranged along the chain [32]. Among copolymer structures, two main types can offer pH

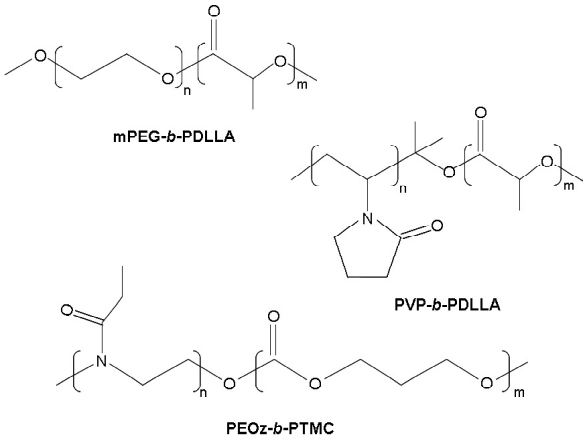

Fig. 2. Examples of amphipilic block copolymer (adapted from [33]). Abbreviations: mPEG-*b*-PDLLA, Poly(ethyleneglycol)-*block*-Poly(D,L-lactide); PVP-*b*-PDLLA, Poly(vinylpyrrolidone)-*block*-Poly(D,L-lactide); PEOz-*b*-PTMC, Poly(2-ethyl-2-oxazoline)-*block*-Poly(1,3-trimethylene carbonate).

sensitive characteristics: block amphiphilic copolymers and random copolymers. A block copolymer is a linear copolymer with one or more long uninterrupted sequences of each monomer (eg, -A-A-A-A-B-B-B-B-), while copolymers formed by two or more monomers units distributed randomly are referred to as random copolymers (eg, A-B-B-A-B-A-B-A) [32]. Copolymers are attractive materials for development of pH-sensitive systems since they possess ionizable and hydrophobic monomer units in their structure, as shown in Fig. 2.

2.3.1. Block copolymeric systems

As can be seen in Fig. 3, block amphiphilic copolymers are able to self-assemble in different structures like spherical micelles, reversed micelles, cylindrical micelles, bicontinuous structures, lamellar phases and vesicles, at or above the critical micelle concentration [34,35]. The most widely used application of these structures is as drug delivery vehicles, since they are able to transport not only hydrophilic compounds but also hydrophobic ones. In addition, several studies have shown their potential applications as pH-sensitive systems [31,36,37]. For instance, micelles, able to release pharmaceuticals at acidic pH, can be used in tumor tissue or endosomic or lysosomic cellular compartments. Moreover, their sizes (20-100 nm) are small enough to avoid uptake by the reticuloendothelial system and also prevent rapid renal exclusion [38]. Although micelles offer great versatility, their applications are limited due to their inherent characteristics such as low encapsulation efficiency, poor storage stability and fast leakage of water-soluble drug in the blood circulation. In this regard, the use of alternatives

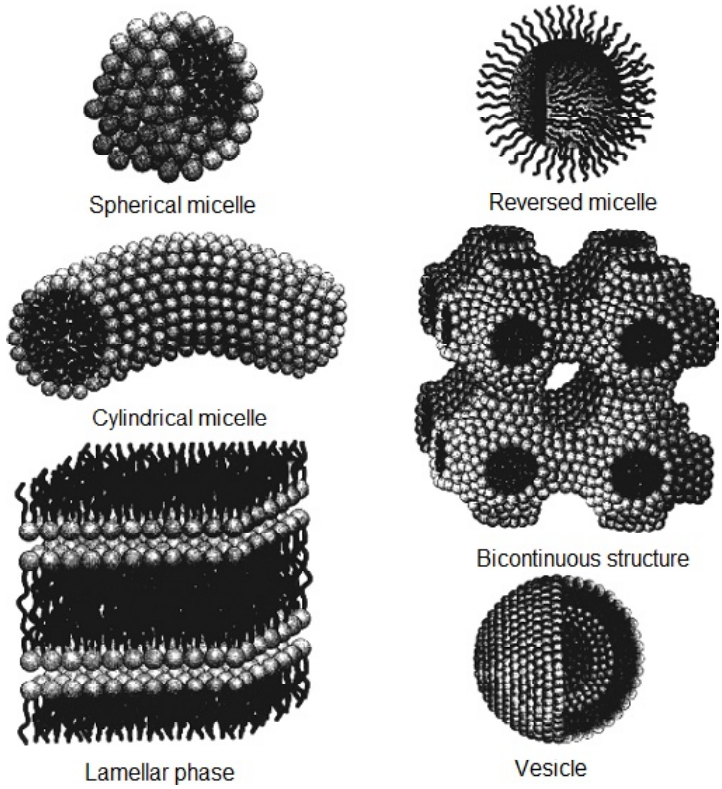

Fig. 3. Structures of block amphiphilic copolymers self-assembly (adapted from [35]).

Table 1. The pH values and the transit time at different segments of the Human GI tract [1].

Anatomical Site	Fasting Condition		Fed Condition	
	pH	Transit Time (h)	pH	TransitTime (h)
Stomach	1-3.5	0.25	4.3-5.4	1
Duodenum	5-7	0.26	5.4	0.26
Jejunum	6-7	1.7	5.4-6	1.7
Ileum	6.6-7.4	1.3	6.6-7.4	1.3
Cecum	6.4	4.5	6.4	4.5
Colon	6.8-7	13.5	6.8-7	13.5

systems like hydrogels has gained increasing attention.

As an example, pH-sensitive hydrogel nanoparticles that respond to the local acidic pH have been prepared from pullulan acetate/oligo-sulfadimethoxine conjugates. These nanoparticles showed great potential since acidic environments can be found in various diseased states such as tumors, ischemia and inflammation. Doxorubicin, released from this system, showed pH-dependent profiles around physiological pH and was significantly promoted at pH<6.8 [39].

One of the most used block copolymer for medical purposes are Poloxamers, also known by the trade name Pluronic®. These polymers are nonionic triblock copolymers composed of a central hydrophobic chain of poly(propylene oxide) flanked by two hydrophilic chains of poly(ethylene oxide) and arranged in the basic A-B-A structure [40]. Recently, Wang et al have used Pluronic®, poly(å-caprolactone) (PCL), and methylacrylic acid (MAA) polymers to obtain a biodegradable and pH-sensitive P(CL-Pluronic-CL-co-MAA-MEG) hydrogel to deliver 5-aminosalicylic acid. *In vitro* release tests of this hydrophobic drug show a good response of the hydrogel to pH change. This indicates great potential applications in smart drug delivery systems, especially in colon targeting therapy [40].

Otherwise, in order to minimize side effects of conventional NSAID formulations, a sequential interpenetrating network of poly(vinyl alcohol) and poly(acrylic acid) has been prepared. This system was crosslinked with glutaraldehyde to form pH-sensitive microspheres by using water-in-oil (w/o) emulsion method [41]. These microspheres were used to deliver diclofenac sodium to the intestine, as a model anti-inflammatory drug. The advantages of such controlled release preparations containing NSAIDs over their conventional dosage forms have been reported earlier by Kulkarni *et al.* [42].

2.3.2. Random copolymeric systems

Many pH-sensitive systems, based on random copolymers, have been synthesized and widely used in development of new formulation. These systems can release the drug at different sites in the gastrointestinal tract, depending on pH values, avoiding not only the peak and valley phenomenon of conventional forms but also the possible side effects.

pH-sensitive polymers are a potent and versatile tool, as targeted drug delivery systems, since the pH of the human gastrointestinal tract is progressively increased from the stomach to the colon [1,43]. The major physiological differences from stomach to colon under fasting and fed states are summarized in Table 1. As can be seen, after oral administration, a pharmaceutical form can take up to 8 h to arrive at the colon [1,44]. Hence, to develop a success pH-sensitive system it must be considered not only pH differences in the gastrointestinal tract but also the transit time of dosage form.

2.3.2.1. Enteric nanoparticles

The word enteric means related to the intestines. Thus, in pharmaceutical development, enteric coating is commonly understood as a formulation designed to pass through the stomach unaltered and dissolve in the intestine. Other terms such us gastro resistant, enterosoluble, delayed release, and pH-sensitive can be used to refer to enteric dosage forms [1].

Nanoparticles, for pharmaceutical purposes, are dened by the Encyclopedia of Pharmaceutical Technology as solid colloidal particles ranging in size from 1 to 1000 nm [45]. They consist of macromolecular materials, in which the active principle (drug or biologically active material) is dissolved, entrapped, or encapsulated, or to which the active principle is adsorbed or attached [46].

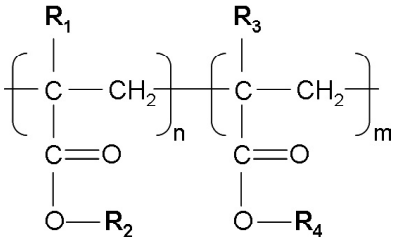

Fig. 4. Schematic view of some enteric copolymer structures.
L100; L12,5; S100 and S12,5: where $\mathbf{R_1} = \mathbf{R_3} = \mathbf{R_4} = CH_3$, $\mathbf{R_2} = H$;
L100-55 and L30D- 55: where $\mathbf{R_1} = CH_3$, $\mathbf{R_2} = \mathbf{R_3} = H$, $\mathbf{R_4} = CH_2CH_3$.

Table 2. pH-sensitive polymer with their threshold pH (adapted from [43]).
Abbreviations: HPMC, Hydroxy propyl methyl cellulose; HPMCP: Hydroxy propyl methyl cellulose phthalate.

Polymer	Threshold pH
HPMCP	4.5
Cellulose acetate trimelliate	4.8
Cellulose acetate phthalate	5.0
Polyvinyl acetate phthalate	5.0
HPMCP 50	5.2
HPMC 55	5.4
Eudragit® L100-55	5.5
Eudragit® L-30D-55	5.5
Eudragit® L100	6.0
Eudragit® L12,5	6.0
Eudragit® FS- 30D	6.8
Eudragit® S100	7.0
Eudragit® S12,5	7.0

Among the major methods used to prepare polymeric nanoparticles are included ionic gelation, coacervation, solvent evaporation, spontaneous emulsification/ solvent diffusion, supercritical fluid technology and polymerization [43,47].

Progress on nanoparticulate systems relies not only on the design of novel methods but also on the development of new polymers. Recently, some commercially random copolymers were introduced to the market by Evonik® & Röhm Pharma. It is claimed that these copolymers are ideal for oral administration because they not only contain methacrylic acid but also acrylic or methacrylic ester units in their structures (see Fig. 4) [48,49]. Since these copolymers are essentially insoluble in the gastric juice, they can be used to impart enteric solubility to the encapsulated drug serving as a drug target device. As can be seen in Table 2, many polymers have been synthesized and exploited in designing gastro resistant drug delivery systems. Table 3 shows some characteristics of two polymers commonly used to obtain enteric nanoparticles.

In aqueous solution, synthetic pH-sensitive polymers are typically either weakly acidic or weakly basic polyelectrolytes. In other words, in their ionic form these polymers are hydrated and swollen, but dehydrate become deswollen and compact (neutral form) [31]. This swelling and deswelling process induces a size and volume change (see Fig. 5), ideal for controlled drug release applications. As an example, polymers like hydroxypropyl methyl cellulose phthalate (HPMCP) are being studied [50,51]. Also enteric submicron particle formulations of papain were prepared by w/o/w emulsion solvent evaporation method using HPMCP, Eudragit® L100, and Eudragit® S100, to avoid gastric inactivation of papain [52].

Site-specific targeting can be achieved using enteric systems able to release the drug at a specific pH. Thus, enteric nanoparticles are such attractive since the nanoencapsulated drug can persist throughout the whole gastrointestinal tract, resulting in delayed or retarded therapeutic action, as a

Table 3. Characteristic of some enteric Eudragits®.

Polymer	Eudragit® L100	Eudragit® L100 55
Description	White powder	White powder
Composition	Methacrylic Acid - Methyl Methacrylate Copolymer (1:1)	Methacrylic Acid - Ethyl Acrylate copolymer (1:1)
Average Molecular Weight	~ 135 000	~ 250 000
Solubility in instestinal fluid	above pH 6	above pH 5.5
Apparent viscosity	50 - 200 mPa . s.	100 - 200 mPa . s.
Refractive index	n_D^{20}: 1.390 - 1.395	n_D^{20}: 1.387 - 1.392
Relative density	d^{20}_{20} : 0.831 - 0.852	d^{20}_{20} : 0.821 - 0.841

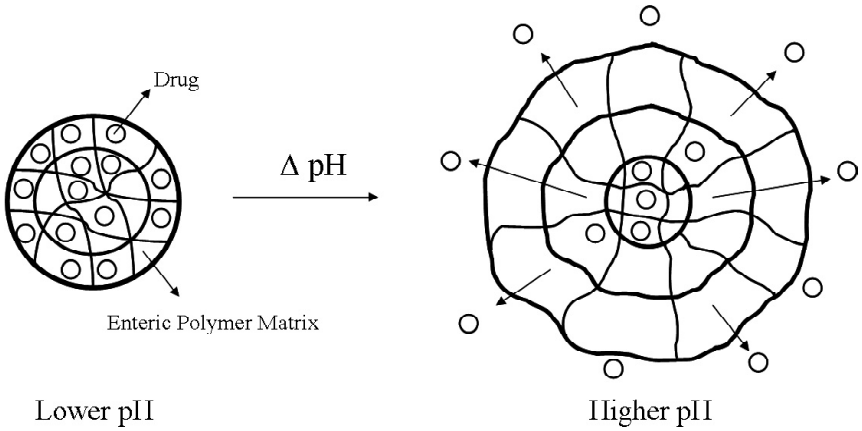

Fig. 5. Schematic representation of swelling and deswelling process of the enteric systems, as a function of pH value.

Table 4. Examples of drug formulations using enteric Eudragits®.

Polymer	Drug	Method	References
Eudragit® L100	Insulin solvent	evaporation (w/o/w)	Jain et al, 2006 [57]
	Prednisolone	coevaporation procedure	Carelli et al., 2000 [58]
	Acetylsalicylic Acid	solvent evaporation (o/w)	González et al., 2008 [59,60]
	Aceclofenac	solvent evaporation	Radhika et al., 2008 [61]
		fluidization	Thiruganesh, 2011 [62]
	Naproxen	solvent evaporation (o/w)	Maghsoodi, 2009 [63]
	Indomethacin	coprecipitation	Karnachi, 1996 [64]
Eudragit® L100 55	Insulin	dispersion-solvent evaporation	Jelvehgari et al., 2010 [65]
	Furosemide	co-administration	Terao et al., 2001 [66]
	Mesalazine	spraying-drying	Khan et al., 1999 [67]
	Diclofenac	electrospinningsolvent	Shen et al., 2011 [68]
	Sodium	evaporation (o/w)	Cetin et al., 2010 [69]
	Ibuprofen	coacervation	Weiß et al., 1993 [70]
			Dong and Bodmeier, 2006 [71]
	Indomethacin	coacervation	Dong and Bodmeier, 2006 [71]

function of pH. Consequently, nanoencapsulation of proteins has been revealed as a useful tool to decrease enzymatic degradation [53-55].

As an alternative approach, pH-sensitive polymers/poly(ethylene oxide) blends have been used to facilitate dissolution of drugs with low solubility in the intestine. Consequently, it is important to understand blends interactions, since they may cause both precipitation and complex formation. It has been reported that drug release, for miscible blends, can be adjusted by varying their composition [1]. As an example, blends have been used to deliver non-proteic drugs to the ileo-colonic region of the intestine [56]. In general, Table 4 summarizes some examples using enteric polymers in drug delivery.

As seen, enteric nanoparticles used as drug delivery systems can combine the inherent benefits of nanoparticles with the special interest of pH-sensitive systems, offering several advantages like [43]:

· Enhance drug absorption
· Decrease dose to be administered
· Lower cost to patient
· Decrease side effects
· Protect mucosa from irritating drugs (e.g., antinflammatories)
· Avoid extensive first pass metabolism
· Drug targeting to specific site (e.g., colon)

In spite of all these advantages, nanoparticles have shown some limitations due to their small size and large surface area. They can rapidly lead to

particle agglomeration and also limit drug loading and burst release [72]. Thus, these problems have to be overcome to made nanoparticles clinically and commercially available.

3. GENERAL REMARKS

In this paper it has been possible to cover some of the most relevant approaches in the expanding field of drug delivery. Oral drug delivery is still by far the most preferable route of drug delivery due to its easy administration, patient compliance and formulations flexibility. As observed, carriers molecules, bioadhesive particles, bioadhesive patches and pH-sensitive systems like enteric nanoparticles offer several possibilities as oral drug delivery systems.

In general, bioadhesive systems seem to be promising devices to improve proteins absorption since they can prolong the residence time of the drug at the absorption site and facilitates the contact between the drug and the absorption surface.

On the other hand, the use of pH-sensitive systems as oral drug delivery devices can provide benefits such us reduced gastrointestinal toxicity, increased stability, enhanced absorption and targeted drug delivery. Development of these systems has multidisciplinary approaches combining medicine, chemistry, engineering and others scientific fields. Their action can be explained in terms of cause and effect, since drug delivery is stimulated by pH change within the human gastrointestinal tract. Considering this behavior, further advances are needed in order to achieve an efficient formulation for oral administration of proteins and AINEs (COX 1 inhibitor).

As a conclusion, pH-sensitive forms, when used considering gastrointestinal physiology, therapeutic needs and formulations strategies, can be a potent tool for designing a product to target drugs. Accordingly, we expect to see such pH-sensitive systems on the market in the foreseeable future.

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