Enteric-coated HPMC capsules: Comparison of enteric coatings and investigation of relationship between in-vitro disintegration and dissolution times

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1. Introduction

Polymeric film coating of oral solid dosage forms may generate products that exhibit controlled release of active ingredients, physical and chemical protection of the ingredients from the external environment as well as taste masking properties [1]. Among controlled release products, sustained and delayed release profiles are prominent. For the former, the drug is released gradually over time whereas for the latter, drug release occurs rapidly following a predefined lag-time. Thus, drugs can be delivered to the small intestine as exemplified by pH-sensitive coatings of solid dosage forms [2].

HPMC (hydroxypropyl methylcellulose) capsules have been available commercially, mainly to the dietary supplement industry as a vegetarian alternative to gelatin, for more than 10 years [3]. Enteric-coated filled HPMC hard capsules are also frequently applied as clinical trial formulations in early stages of clinical investigations [4] since some new chemical entities (NCE's) are prone to instability in gastric fluids or irritate the gastric mucosa. Furthermore, the limited amount of drug substance available at early development stages often precludes the development of other solid dosage forms, such as coated pellets or tablets. Advantages result from the ability to coat a capsule, since the coating process is independent of the capsule contents thus avoiding extensive formulation development.

During capsule coating challenges are generally due to the characteristics of the capsule wall. In particular, for gelatin-based capsules, the shell may soften and become sticky upon spraying of aqueous enteric polymer dispersions or it may become brittle due to water evaporation during drying with consecutive loss of mechanical stability. Furthermore, insufficient adhesion of the film with splintering and peeling of the coat (orange peel effect), especially with organic spray formulations has been reported [5]. In this respect, HPMC capsules offer the advantage of less sensitivity to aqueous coatings if appropriate sealing, i.e. closure of the gap between the capsule body and the cap is obtained to avoid leaking of the capsule content into the stomach or vice versa. Moreover, site-specific delivery of HPMC capsules was claimed by using coating materials with different pH-dependent solubilities, i.e. dissolving at pH $\geq$5.5 and pH $\geq$7 for upper GI and colonic delivery, respectively. A comparison between gelatin versus HPMC capsules coated with acrylic polymers Eudragit® L and S 12.5 demonstrated that the gelatin capsules resulted in brittle film coats with insufficient adhesion on the smooth capsule surface [6]. Since the surface of HPMC is more ragged compared with gelatin capsules, HPMC materials may lend themselves to better polymer coat adhesion [7].

1.1. Enteric polymers

Several aqueous coating suspensions are commercially available based on different polymers: Cellulose acetate phthalate, C-A-P (Aquateric®); Hypromellose acetate succinate, HPMCAS (AQOAT®); Hypromellose phthalate, HPMCP (HPMCP®); Polyvinyl acetate phthalate, PVAP (Opadry®); Polymethacrylate-polymethylmethacrylate copolymer, PMA-PMMA (Eudragit®, Kollicoat® MAE). Current enteric polymers contain carboxylic functional groups, enabling water solubility at weakly acidic to neutral pH, i.e. pH 5 to 6.5 and above, through salt formation. In addition to pH, dissolution / solubility profiles are dependent on structural elements of the polymer such as the number of carboxylic groups, the type of acidic functional groups and the polymer molecular weight. Thus, a range of different products may be created differing in onset of drug release in the intestine.

In the present study, AQOAT® (HPMCAS), HPMCP (HPMCP®), Eudragit® L100-55 and Eastman® C-A-P as coating materials for HPMC hard capsules are compared. Their structures are displayed in Figure 1:

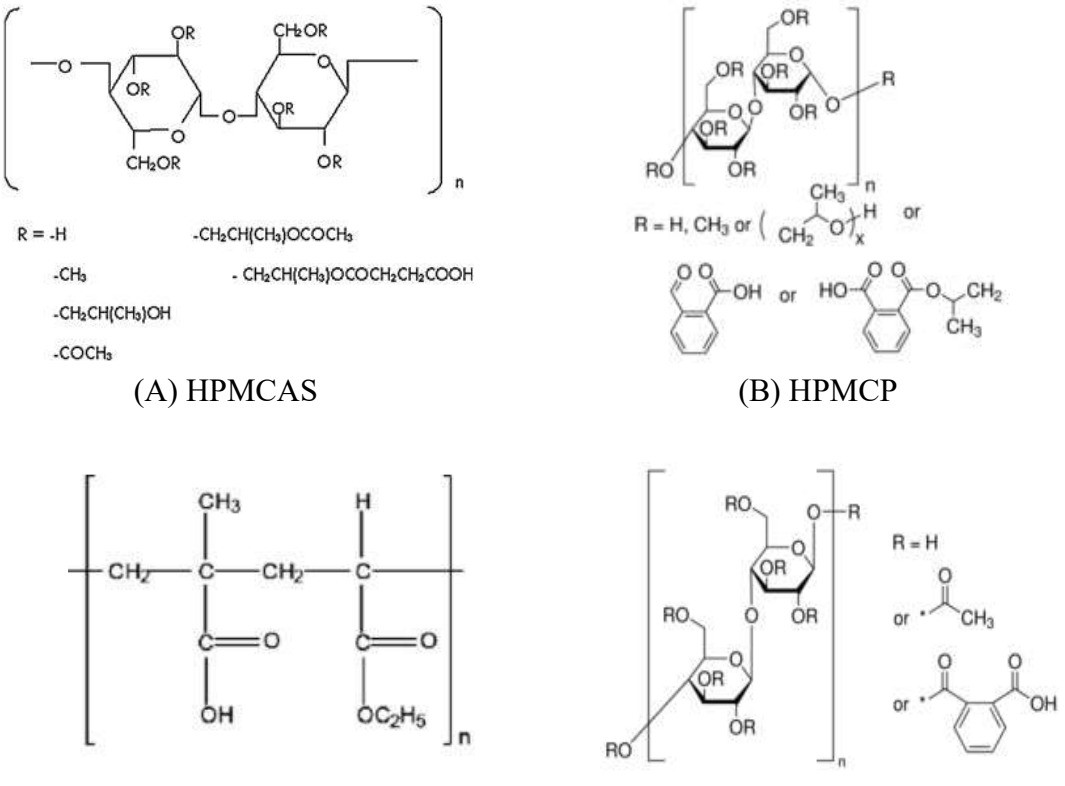


Figure 1: Chemical structures of (a) HPMCAS; (b) HPMCP; (c) PMA-PMMA; (d) C-A-P.

1.1.1. Structure-functionality relationship for enteric polymers

1.1.1.1. HPMCAS

Hypromellose acetate succinate (HPMCAS)—a mixture of acetic acid and monosuccinic acid esters of hydroxypropyl methylcellulose (HPMC)— is available in several grades that vary in their extent of substitutions of acetyl and succinoyl groups and particle size [8]. HPMCAS is a cellulose based polymer with four types of substituents, semi-randomly substituted on the hydroxyls: (1) methoxy (mass content of 20-26 wt %); (2) hydroxypropyl (mass content of 5-10 wt %); (3) acetate (mass content of 5-14 wt %); (4) succinate (mass content of 4-18 wt %) [8]. The succinate groups of HPMCAS have a pKa ≈5, therefore, the polymer is less than 10% ionized at pH values ≤4 and is ≥50% ionized at pH values ≥5. Due to the presence of relatively hydrophobic methoxy and acetate substituents, HPMCAS is water-insoluble when un-ionized (pH <4) and remains predominantly colloidal at intestinal pH of 6.0-7.5. Acetyl and succinoyl substitutions on the HPMC backbone (Figure 1) dictate the pH-solubility behavior of the polymer. There are three commercial grades of HPMCAS. The approximate pH values above which each grade becomes dispersible or soluble in aqueous media are -H grade, pH 6.5; -M grade, pH 6.0; and -L grade, 5.5 [8].

1.1.1.2. HPMCP

HPMCP is produced by transesterification of HPMC with phthalic anhydride [9] and was first introduced in 1971 as a cellulose derivative for enteric coating. HPMCP has been admitted in the European and Japanese pharmacopeias and is included in the USP/NF under the name hypromellose phthalate. Depending on the degree of phthalyl substitution, HPMCP is soluble in aqueous media at pH 5 to 5.5 and above. Three primary grades of HPMCP are available: HP-50, HP-55 and HP-55S. HP-50 has a nominal phthalyl content of 24% and dissolves at pH >5.0, while HP-55 and HP-55S have 31% nominal phthalyl content and dissolve at pH >5.5.

The HP-55S grade has a higher molecular weight than the HP-55 grade which results in a higher solution viscosity, increased film tensile strength and resistance to simulated gastric fluid [8]. The HP-55S grade therefore requires less coating thickness for enteric functionality and exhibits stronger crack resistance than the HP-55 grade [10]. Specifications of the different HPMCPs are summarized in Table 1.

1.1.1.3. C-A-P

C-A-P is produced by reacting the partial acetate ester of cellulose with phthalic anhydride in the presence of a tertiary organic base or a strong acid [11]. The USP specifies that C-A-P must contain 21.5-26.0% acetyl ($COCH_3$) groups and 30-36% phthalyl (o-carboxybenzoyl, COC_6H_4COOH) groups on the cellulose backbone as calculated on an anhydrous basis. The degree of substitution is equal to about half of the acylation of the available hydroxyl groups, and about one quarter is esterified with one of the two carboxyl groups of phthalic anhydride. Since only one carboxyl group on the phthalic acid moiety participates in the substitution, the other remains free to form a salt, thus providing enteric dissolution to the polymer [12]. At this degree of substitution, C-A-P showed water solubility in the vicinity of pH 6 and above[13]. The

degree of substitution is the key for dissolution in the intestinal fluid. It has been determined that C-A-P has a threshold of about 20% phthaloyl substitution to ensure rapid dissolution at intestinal pH [14].

1.1.1.4. Methacrylic acid copolymers

Methacrylic acid copolymer (IUPAC name Poly (methacylic acid-co-ethylacrylate) 1:1, is an anionic copolymer of methacrylic acid and ethyl acrylate with methacrylic acid as a functional group, allowing effective and stable enteric coatings dissolving in the upper bowel at pH >5.5. Among the anionic copolymers, it is the only polymer that is available as an aqueous, ready-to-use 30% dispersion (Eudragit L30D-55). Other types available include solutions in acetone or alcohols.

Besides the choice of the polymer and other excipients in the coating fluid, several process variables can influence the coating efficiency. These include the type of coating equipment, air pressure used for atomization, spraying nozzle position, inlet air temperature and humidity, flow rates of the air and coating suspension and subsequent curing process [15]. The enteric coating polymers used in this study and their properties are presented in Table 1 below.

Table 1: Enteric coating polymers used in the study.

Polymer	Name	Grade	Function related characteristic	Opening pH value
Hypromellose acetate succinate, HPMCAS	Shin-Etsu AQOAT®	AS-LG	Acetyl: 8.2 % Succinoyl: 14.9 %	>5.5
Hypromellose acetate succinate, HPMCAS	Shin-Etsu AQOAT®	AS-MG	Acetyl: 9.3 % Succinoyl: 11.3 %	>6.0
Hypromellose acetate succinate, HPMCAS	Shin-Etsu AQOAT®	AS-HG	Acetyl: 11.7 % Succinoyl: 7.5 %	>6.5
Hypromellose Phthalate	HPMCP	HP-50	Phthalyl: 23.1 % Viscosity: 55 mPas	>5.0
Hypromellose Phthalate	HPMCP	HP-55	Phthalyl: 32.9 % Viscosity: 43 mPas	>5.5
Hypromellose Phthalate	HPMCP	HP-55S	Phthalyl: 33.2 % Viscosity: 167 mPas	>5.5
Methacrylic acid and ethyl acrylate copolymer	Eudragit®	L100-55	Ratio of methacylic acid to ethyl acrylate ~ 1:1	>5.5
Cellulose acetate phthalate	Eastman™ C-A-P	C-A-P Cellulose Ester NF	Acetyl: 21.5-26 % Phthalyl: 30-36 %	n.a.

1.2. Performance testing of enteric coated dosage forms

Disintegration tests have been used to assess dosage form performance starting from the early 20th century with the current basket rack assembly apparatus being compendial since the early 1950's[16]. Despite the predicament of the released quantity of the drug (API) no longer being measured, these tests are still broadly utilized in pharmaceutical quality control because of their simplicity and speediness in comparison to dissolution tests. This contributed to the International Conference on Harmonization (ICH) guideline allowing disintegration tests to be used as dissolution test surrogates if (among other things) a correlation between disintegration and dissolution is proven [17].

In this regard, it seems from the literature information that the probability of establishing a disintegration–dissolution correlation varies substantially from formulation to formulation, with Gupta et al. [18] testing twelve different verapamil hydrochloride formulations only one of which gave a good correlation. Radwan et al. [19] investigated different trospium chloride tablets in media of different viscosities and found that a correlation is feasible in those cases when the disintegration is not too rapid. Nickerson et al. [20] on the other hand found good disintegration-dissolution correlations for several immediate release formulations of an unnamed API, indicating that disintegration times are suitable to demonstrate biopharmaceutic drug product quality. However, the focus has commonly been restricted to immediate release dosage forms, most probably due to the ICH guidance restricting the possibility of employing the disintegration test as a dissolution test surrogate to non-modified release dosage forms.

One type of modified release dosage form, particularly enteric-coated (EC) formulations, offers a theoretical possibility for obtaining proper disintegration - dissolution relationships. For, in the presence of a swiftly disintegrating and dissolving core, having a scenario wherein the disintegration of the enteric coat strongly influences the overall release performance is likely. Therefore, this work is also going to investigate the correlation between disintegration and dissolution of enteric-coated hard-shell capsules so one can discover the feasibility of using the disintegration test as a dissolution test surrogate for EC dosage forms.

1.3. Behavior of EC dosage forms in the fasted state

1.3.1. Stomach.

Enteric-coated dosage forms are not supposed to release appreciable quantities of the API into the stomach. Therefore, they are designed to remain intact in the stomach until they are emptied into the duodenum, and only after that can drug release begin. The gastric emptying time of the enteric dosage form will impact the pharmacokinetic profile of the API. The exact way gastric emptying can affect the API's pharmacokinetic profile depends on whether the dosage form is a single unit or multiparticulate one. The emptying of a single unit dosage form is an all or none single impulse event so it will generally be reflected in the absorption onset lag time (i.e., the degree of the shifting of the pharmacokinetic profile) since significant drug absorption is supposed to start only after the dosage form is emptied from the stomach.

The emptying of a multiparticulate dosage form is a continuous function rather than a single impulse, and so the overall absorption rate can be viewed as the result of convoluting this gastric emptying function with the API release in the small intestine as a weight function followed by convoluting the resulting output with the absorption of dissolved API from the small intestine as a weight function. This means that the gastric emptying kinetics of such a dosage form will be reflected not only in how much the pharmacokinetic profile is shifted but in its shape as well since they will be a component of the kinetics of the post lag time API absorption.

For single unit EC dosage forms, the gastric emptying time is strongly affected by the migrating motor complex (MMC) phase during which the dosage form has arrived at the stomach [21].It most commonly occurs during the phase III (the "housekeeper waves") of the MMC [21]. Dosage form size and density could affect the gastric emptying time, with some data indicating that small tablets with high densities could have longer and more variable gastric emptying times because they could become entrapped within the folds of gastric mucosa and thus can escape emptying during their first exposure to housekeeper waves [22],[23]. The emptying of larger single unit dosage forms could pose a problem in patients with pyloric stenosis. A case report has been published about a 70 year old patient with subclinical pyloric stenosis who accumulated 65 g of enteric-coated aspirin tablets over 14 weeks (the accepted fatal dose of aspirin for an adult lies between 24 and 30 g) [24].

As for multiparticulate systems like pellets and beads, these tend to empty gradually, but the process is still affected by the MMC cycle phase, with individual emptying profiles often showing lag times, interruptions, and different emptying speeds at different time intervals [25]. For modeling the mean gastric emptying profiles of pellets, Locatelli et al. suggested using the Weibull distribution function as follows:

$$\%\text{remaining to be emptied} = 100 * \exp\left(-(t/61.9)^{0.895}\right)$$

It has been observed that size has little effect on the gastric emptying rate of beads and pellets in the fasted state [26],[27]. As for density, there seems to be a threshold value of about 2.4 g cm^{-3} , with values above this threshold leading to prolonged gastric residence [28].

Another important aspect of the performance of an enteric dosage form in the stomach is the degree of protection it affords to an acid-labile active ingredient against degradation by gastric juice. The permeability of an enteric film-former to acid is an important factor governing this aspect with cellulose acetate phthalate (CAP) and hypromellose phthalate (HPMCP) being known for their relatively high permeabilities to acids [29].

Concerning *in vitro* prediction of the product's performance in the fasted stomach, dissolution and disintegration tests are typically performed for one or 2 h in 0.1 M HCl, simulated gastric fluid (SGF) of the USP, or 1st dissolution and disintegration fluids of the Japanese Pharmacopoeia (JP) (practically identical to the USP SGF except for their lack of pepsin). In a disintegration test, the dosage form must remain intact at the

end of the acid stage, while in a dissolution test not more than 10% of the dose should be released.

The limitations to these tests include the fact that the fasted gastric pH tends to be a bit higher than the pH 1−1.2 typically employed by compendial media [30]. In addition, dosage forms could spend highly variable times in the stomach. In particular for single unit enteric-coated dosage forms, instances of very long gastric residence times have been reported [29]. This could affect not only the ability of the dissolution system to evaluate the probability of premature drug release in the stomach, but also that of the next stage to predict the speed of postgastric emptying release. This is because longer gastric residence time results in more accumulation of protons in the enteric coat, and this leads to more delayed onset of release postgastric emptying [31]. Another important aspect that is dependent on the length of the dosage form's gastric residence time is the degree of inactivation of acid-labile drugs. Concerning this aspect, Stefanic et al. developed a flow-through cell-based method where lansoprazole enteric-coated pellets were subjected to acid for different periods of time and then using the Weibull distribution model proposed by Locatelli et al. calculated the estimated amount of drug remaining intact [32]. This work could serve as a good starting point for developing *in vitro* methodologies to predict an enteric coat formulation's overall ability to protect an acid-labile drug.

An additional issue is that the media used in *in vitro* dissolution studies have been designed based on conditions found in healthy people. However, disease states and certain drugs can alter gastric pH. A significant issue for enteric-coated dosage forms regarding this aspect could be drug-induced achlorhydria. The most commonly used class of drugs to elevate gastric pH is the proton pump inhibitors (PPI's). These drugs are typically enteric-coated owing to their rapid degradation in acid. Therefore, following multiple dosing, the enteric coat will face a pH higher than 4 in the stomach [33]. The USP does not account for this, but the British Pharmacopoeia (BP) does and specifies that the acid stage of the dissolution test be done in a pH 4.5 phosphate buffer for these drugs. However, by omitting the test in HCl, the BP approach overlooks that the first dose will face a normally acidic gastric juice. Therefore, the most appropriate approach would be to perform the dissolution test twice: once with a normal HCl medium, and once more with a pH 4.5 phosphate buffer. It should be taken into account; however, that achlorhydric gastric fluid contains no phosphate but rather has an elevated pH owing to lack of HCl secretion, and, therefore, using highly diluted HCl might be more appropriate (similar to the approach of Matsui et al in simulating the effect of acid-reducing agents [34]).

As for how elevated gastric pH affects the performance of enteric-coated products, the studies of which the authors are aware have so far not found any effect: one study found that the pharmacokinetics of EC prednisolone were not affected by cimetidine co-administration suggesting that the elevated gastric pH would not affect the performance of this EC product [35]. However, the gastric pH was not measured and treatment administration was after light breakfast rather than during the fasted state. Therefore, this study cannot be considered to conclusively eliminate the possibility of elevated gastric pH affecting the performance of enteric-coated dosage forms. A

second study found that the pharmacokinetics of EC-mycophenolate sodium were not affected by the discontinuation of pantoprazole in heart or lung transplant recipients on EC-mycophenolate sodium therapy [36]. However, the rather long half-life of the API (9-17 hours)[37] makes the pharmacokinetic parameters less sensitive to alterations in absorption rate, and this rather low sensitivity to changes absorption rate is even further lowered by the fact that the API was administered in multiple twice daily doses. Moreover, the gastric pH was not measured in this study as well. Last but not least, those two studies investigated only two products with maximally two out of the many available enteric polymers.

Yet another limitation of the usual disintegration and dissolution test is related to the involvement of mechanical destructive forces brought about by the contraction of the stomach wall muscles. In the stomach, these mechanical destructive forces (1.5 N) were found to be significantly higher than those in compendial disintegration and dissolution testers [38]. This could affect a method's ability to predict mechanical stress-induced dose dumping in the stomach. However, for disintegration testing of tablets, the pharmacopoeial provision that no tablet should show signs of softening during the acid stage helps to reduce this risk since unsoftened tablets generally can resist forces higher than the 1.5 N reported for the stomach. This makes the issue of mechanical forces in the stomach less likely of being problematic for EC dosage forms than for immediate release ones which are generally softened by gastric fluid and disintegrate in the stomach.

1.3.2. Intestine.

1.3.2.1. Enteric Coatings Targeted to the Small Intestine.

Following gastric emptying, the enteric coat begins to dissolve and erode allowing fast ingress of water that triggers the swelling of the disintegrant inside the core leading to dosage form disintegration and start of drug release. Factors influencing the speed at which this happens include [29]:

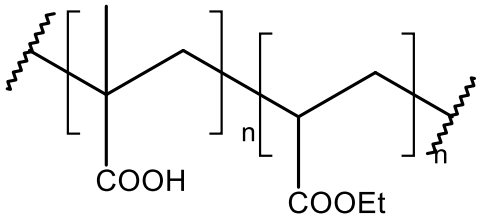
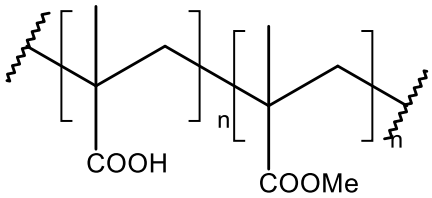
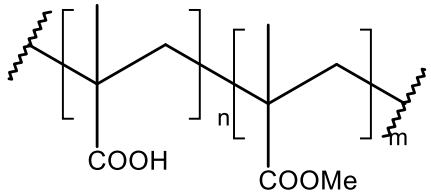
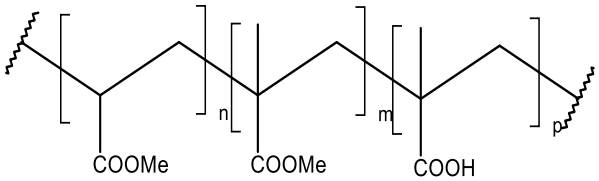
1) The nature of the employed film former.
2) The nature and quantities of the additives used together with the film former in the dosage form.
3) The thickness of the applied film.
4) The physic-chemical properties of the drug, with acidic drugs in the core exerting an acidifying effect on the coat and thus prolonging coat dissolution (and the opposite happens with basic drugs).
5) Osmotic properties of the core.
6) Swelling properties of the core.

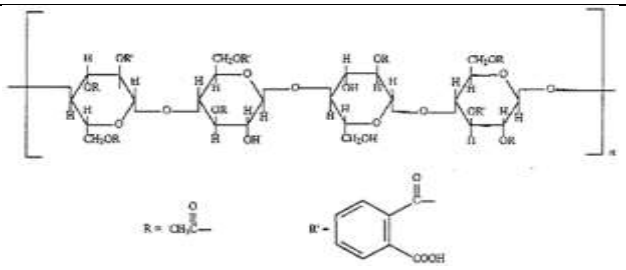
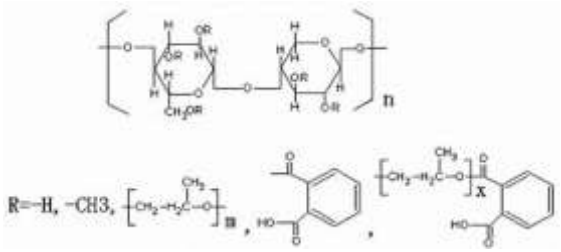
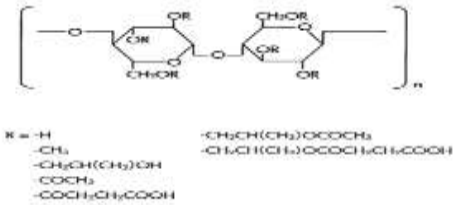
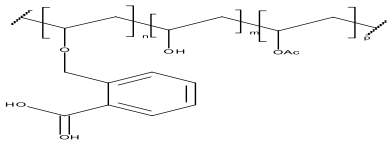
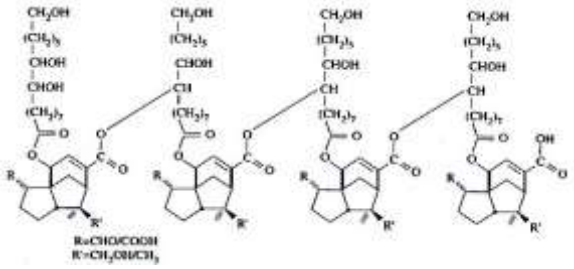
Different enteric polymers have been determined to have different dissolution pH thresholds (as outlined in Table 2). The polymers with dissolution pH thresholds of 5.5 and lower are considered to target the onset of release to the duodenum while those with a dissolution pH threshold of 6.0 are considered to target the jejunum. As for those with dissolution pH threshold values of 7.0-7.2, they are considered to delay the onset of release until the dosage form reaches the last parts of the ileum and are employed for colon-targeting. However, the reality of what happens to these dosage

forms in the human intestine *in vivo* does not seem to be that simple and straightforward.

Enteric-coated products were once described as the most difficult to predict among all dosage forms [39]. This has been typically attributed to erratic gastric emptying patterns with the onset of release assumed to take place quickly after the dosage form passed through the pylorus. However, closer examination of data available in literature shows that, even though the gastric emptying of enteric-coated products can often be erratic, it is not the sole culprit. Indeed, the onset of release often does not take place quickly following exit through the pylorus, and it can be an unpredictably variable as well.

Table 2: Examples of enteric film-formers and their range of dissolution pH threshold values

Polymer name	Structure	Dissolution pH threshold
Methacrylic acid-ethyl acrylate 1:1 copolymer (Eudragit L55; Kollicoat MAE)		5.5
Methacrylic acid-methyl methacylate 1:1 copolymer (Eudragit L)		6.0
Methacrylic acid-methyl methacylate 1:2 copolymer (Eudragit S)		7.0
Methyl acrylate-methyl methacrylate-methacrylic acid 7:3:1 copolymer (Eudragit FS)		7.0

Polymer name	Structure	Dissolution pH threshold
Cellulose acetate phthalate (CAP)		6.0
Hydroxypropylmethylcellulose phthalate (HPMCP)		5.0 (HPMCP-50); 5.5 (HPMCP-55)
Hydroxypropylmethylcellulose acetate succinate (HPMAS)		5.5(HPMCAS-L); 6.0(HPMCAS-M); 6.8(HPMCAS-H)
Polyvinylacetate phthalate (PVAP)		5.0
Shellac		7.2

The unpredictability of the post-gastric emptying onset of release is clearly illustrated by enteric tablets observed exiting the body intact in the stool following a bowel movement [40]. Another notable example is a case report of a man hospitalized for intestinal obstruction caused by the accumulation of undisintegrated enteric-coated prednisolone tablets in his bowel [41]. A less dramatic example is the poor bioavailability of iron from enteric-coated dosage forms [42]. This has been attributed

to the release of iron occurring after the dosage form has already passed this nutrient's preferential absorption site in the duodenum and proximal jejunum [42], illustrating that EC dosage forms targeted to the duodenum often release their load too distally *in vivo*, thus missing their target site.

These discrepancies are attributed to the overestimation of the disintegration and dissolution rates of EC dosage forms by the compendial disintegration and dissolution tests as shown by a few studies [4],[43],[44],[45]. While most of the aforementioned studies and case reports date back to long time ago when the pH of the buffers used was above 7, some of them were done with the currently employed pH 6.8 buffers.

For instance, Cole et al evaluated enteric-coated HPMC hard-shell acetaminophen capsules both *in vitro* using the current compendial dissolution procedure and *in vivo*, in humans, using gamma ray scintigraphy [4]. The *in vitro* testing predicted the onset of release to be approximately 15 minutes; however, the times (post-gastric emptying) of initial tablet disintegration recorded scintigraphically *in vivo* gave an estimate of 63 minutes as shown by Table 3.

The capsules used were coated with a formulation based on the film-former Eudragit L 30 D-55 [4]. According to the manufacturer, this film-former targets the dosage form release to the duodenum [46]; however, the *in vivo* scintigraphic data show that, in half the subjects, the release onset does not occur even in any part of the proximal small bowel but in the middle and distal small bowel; something that, based on the *in vitro* dissolution results, would not have been expected. When formulating an API with e.g. a preferential absorption site in the duodenum and proximal jejunum, this could have major impact on drug bioavailability. Therefore, this study showcases that the current compendial test methods still have the potential to mislead the formulator into developing an unsuccessful enteric-coated product.

Table 3: In vivo disintegration onset times of enteric-coated hard-shell capsules recorded scintigraphically by Cole et al.

Volunteer	Gastric emptying time (h)	Disintegration onset time (h)	Gastric emptying-corrected disintegration onset time (h)	Location of disintegration onset
1	2.0	2.8	0.8	Proximal small bowel
2	0.2	1.4	1.2	Middle small bowel
3	0.4	1.5	1.1	Middle small bowel
4	0.9	1.3	0.4	Proximal small bowel
5	0.8	1.9	1.1	Distal small bowel
6	0.1	2.5	2.4	Proximal small bowel
7	0.4	1.3	0.9	Proximal smal bowel
8	1.5	2	0.5	Distal small bowel
Mean ±SD	0.8 ±0.7	1.8 ±0.571	1.1 ±0.6	
In vitro onset of release	~0.25 hours			

In a more recent study [45], it was shown that the difference between the *in vivo* post-gastric emptying drug release lag times of two commercially available enteric-coated aspirin formulations is significantly greater than that predicted by the USP dissolution method (45 vs 10 minutes; p-value<0.050). This supports the conclusions derived from the Cole et al study.

When investigating the discrepancies those reports have exhibited, the first factor that comes in mind would be the pH of the employed buffers. While this is clearly valid concerning the formerly used buffers with pH values exceeding 7, the currently used ones have a pH of 6.8, which, though still somewhat higher than most estimates of proximal intestinal pH [30], is not the sole culprit. Another highly significant cause is the employed phosphate buffers' having too high buffer capacity (3-6 fold compared to the human intestinal fluid(HIF)) and, in some cases, even too high ionic strength (3-fold compared to HIF in the case of the Ph Eur disintegration testing method) [45],[47].

Bicarbonate buffer systems that, to an appreciable extent, simulate the composition of the intestinal buffer system have been proposed for use to improve the accuracy of *in vitro* predictions of the post gastric emptying behavior of enteric-coated dosage forms [48],[49]. The problem with such buffers is that, technically, bicarbonate buffer is hard to operate for *in vitro* studies. This is due to the thermodynamic instability of carbonic acid and the resulting evaporation of carbon dioxide requiring continuous sparging with carbon dioxide to maintain the target pH. Automated computer-controlled systems have been proposed that can automatically adjust the pH of test buffer conditions as the experiment progresses to simulate the pH increase faced by the dosage forms as it traverses the small intestine [48],[49]. However, such technologically advanced systems are not readily accessible for routine use to all laboratories, and, moreover, due to the previously outlined technical challenges, bicarbonate buffer systems are extremely difficult to establish for disintegration testers, reciprocating cylinder-type dissolution testers and flow through cells. However, when fully optimized, such systems could serve as effective optimization tools for the development of simpler more convenient to handle phosphate or other buffer systems. This would be similar to what has been done by other researchers who used the bicarbonate buffers to develop simpler surrogate phosphate-based systems for the intrinsic dissolution of drugs [50].

Indeed, the concept of developing a simple phosphate-based surrogate method has recently been applied to enteric coatings [45]. As a result, a relatively simple phosphate-based method has been proposed. This method also involves increasing the pH and the buffer molarity by adding concentrated disodium monohydrogenphosphate solutions when testing late-disintegrating products. This procedure allows the simulation of the increasing pH and bicarbonate molarity faced by such products as they move along the small bowel. The method showed significantly improved biopredictability, compared to the currently most established methods: the USP and the Fasted State Simulated Intestinal Fluid (FaSSIF)-based method, even though room for further improvement still exists. During the development of this method, the

investigators also showed the importance of simulating the increasing concentration of the buffering system's conjugate base in the distal direction along the small intestine.

One important factor that has so far been largely ignored when working to develop improved *in vitro* tests for enteric-coated products (and other dosage forms as well) is the mechanical stresses and hydrodynamic conditions experienced by the dosage form in the human small intestine. Because EC dosage forms enter the small intestine still undisintegrated, the mechanical destructive forces exerted by the small intestinal contractions play more important role in their disintegration than in that of immediate release dosage forms which usually disintegrate in the stomach. Stefanic et al. introduced a continuous flow dissolution method incorporating a stirring element exerting additional mechanical stress on EC lansoprazole pellets to improve the predictions of *in vivo* performance [51]. Even though this way of introducing additional mechanical stress improved the prediction of the *in vivo* dissolution for EC lansoprazole pellets, the magnitude of the stress force in the dissolution apparatus remains to be validated. If such validation is done, it could, together with the work of Garbacz et al., described in the next subsection, provide a starting point for systematic work on mechanical stresses in *in vivo*-predictive dissolution apparatus development. Such work could help to further narrow the remaining gaps left by works that have focused solely on the buffer systems.

There exists a further issue, which, though not unique to EC dosage forms, could be, in many cases, of greater significance to them than it is to immediate-release (IR) drug products. This issue is that, in the small intestine, water is present in discrete small-volume pockets rather in a large continuous pool [52],[53]. At peak total small bowel water volume, achieved at 12 min following ingestion of 240 mL of water, 15 ± 2 pockets with volumes in the range of 6 ± 2 mL were observed [53]. This situation is markedly different from that faced during a typical dissolution test where 900−1000 mL continuous pools of fluid are typically employed. This could contribute to the actual in vivo dissolution being slower than predicted in vitro except in the case of extremely highly permeable drugs, where assuming the presence of sink onditions will not lead to too large deviation from the real situation despite the large discrepancy in fluid volumes. The effect is expected to be often larger in EC (particularly single-unit) dosage forms compared to IR ones because IR dosage forms disintegrate in the stomach where the fluid pool is much larger (ca. 250 mL immediately after ingestion of 240 mL of water) [53], and so the dissolving API particles are dispersed through a larger volume not to mention that, in the case of many API's, a significant fraction will dissolve in the stomach.

Another important issue that needs to be taken into account is that *in vitro* methods tend to be developed based on average values of *in vivo* parameters. However, many individuals exhibit deviations from the average values of GI physiological parameters leading to unpredictably erratic performance. Therefore, during formulation development, running additional *in vitro* tests under conditions simulating deviation of certain critical parameters (e.g. pH) from the average could be helpful to increase the chance of consistent product performance.

In regard to reducing the effect of such deviations, EC polymers that show a performance less affected by variation of *in vivo* conditions would be helpful in reducing the impact of such issues.

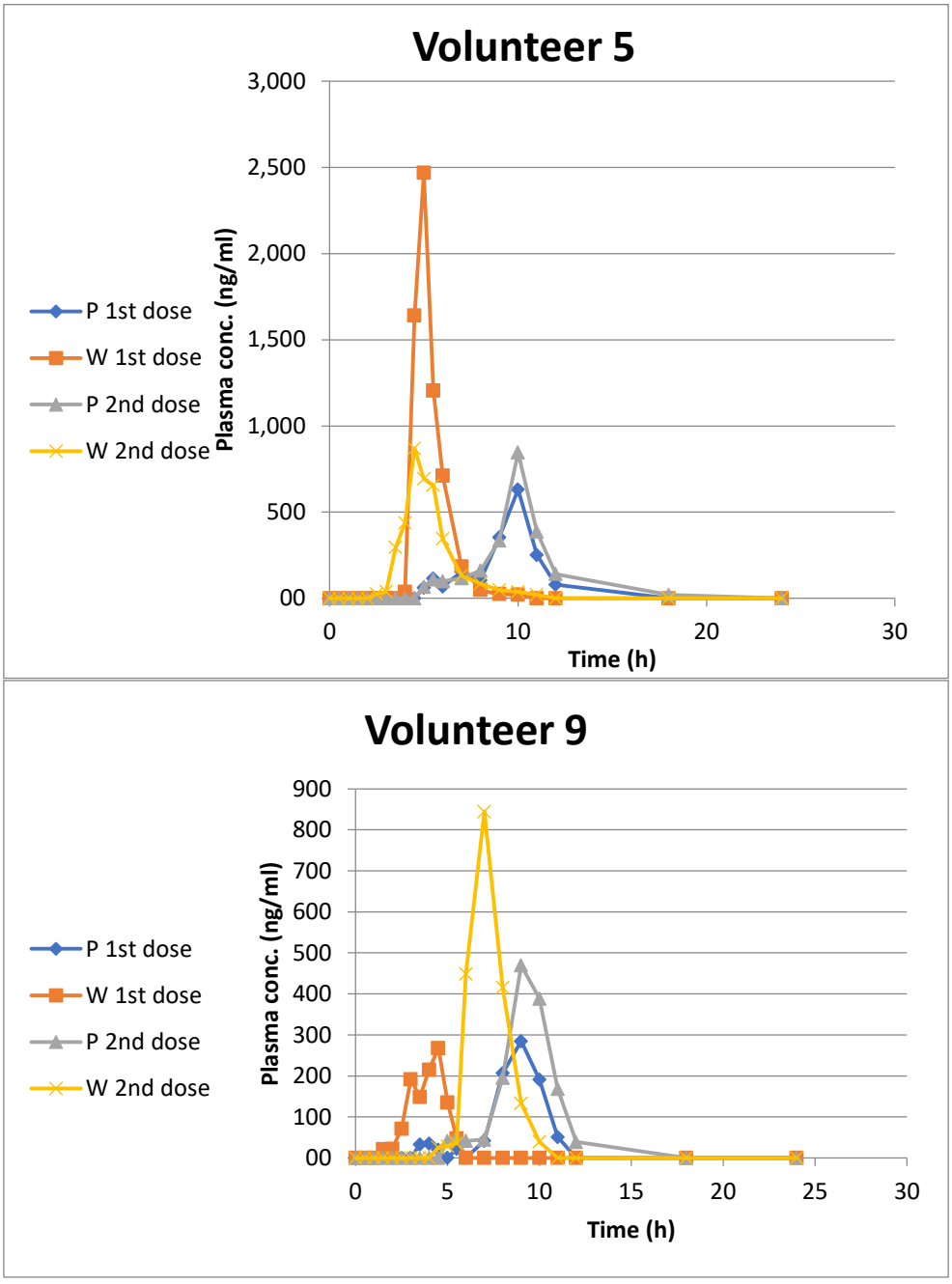


Figure 2 Pharmacokinetic profiles following the administration of EC Aspirin in two subjects (Volunteers 5 and 9) participating in the replicate design study of Al-Gousous et al. P represents Aspirin Protect and W represents Walgreens Aspirin. The variations in areas under the curve (AUC's) are, most probably, mainly due to the highly variable first pass metabolism of Aspirin (Al-Gousous et al., 2016). The variability in the lag times is due to variability in gastric emptying (Al-Gousous et al., 2016).

For example a parameter that would be critical for the performance of an EC dosage form is the intestinal pH. Using a film-former with lower pKa values could help reduce the impact of intestinal pH variation on dosage form performance as shown in Figure 2.

In those two volunteers from the Al-Gousous et al study on EC aspirin [45], the plasma concentration profiles consistently show a relatively long period of slow drug release for Aspirin Protect (immediately after the lag time). In Walgreens aspirin, this period is considerably shorter and in the case of one profile (volunteer 5: first dose of Walgreens aspirin) even almost absent. One explanation could be that those periods reflect a slow initial release in the stomach due to prolonged gastric emptying. However, considering that the astric emptying of single unit dosage forms is a random event with high intrasubject variability [21], the rather consistent presence of this observation in those two volunteers makes this explanation unlikely. Therefore, a more likely explanation would be that those subjects have lower intestinal pH values leading it to take longer for the dosage form to release its load of the drug following gastric emptying.

Walgreens aspirin is coated with a coat based on PVAP as a film-former in contrast to Aspirin Protect which is coated with a coat based on methacrylic acid-ethyl acrylate 1:1 copolymer as a film-former. By virtue of the lower pKa of its monomers [45], PVAP has a lower dissolution pH threshold(pH 5.0) compared to methacrylic acid-ethyl acrylate 1:1 copolymer(pH 5.5). The lower pKa also enables its performance to be less affected by variations in intestinal pH since the farther from the medium pH the pKa value lies, the less the variation of the ionized fraction with pH is (Figure 3). When a change in the pH value results in a smaller change in the fraction ionized, the variability of formulation performance brought about by variability in intestinal pH can be expected to be lower. Therefore, polymers with lower pKa values could be useful in this regard.

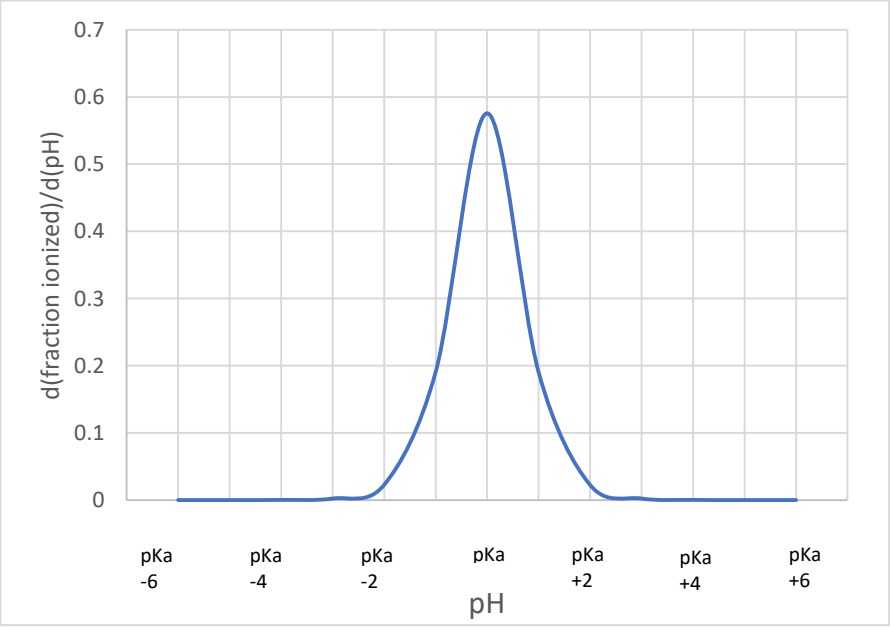


Figure 3: The rate of the change of the ionized fraction of a substance with respect to pH calculated by differentiating the expression fraction ionized=1/(1+10pka-pH) derived from re-arranging the Henderson-Hasselbalch equation for a weak acid. It should be noted that for a polymeric material, owing to the effect of the ionization of a carboxylic group on the ionizability of the neighbouring ones, this curve would be broader and the pKa will be replaced with an average pKa.

Developing such polymers with lower pK a could also help to achieve more efficient targeting of drug release to the proximal small intestine through enhancing polymer solubility at lower pH values (though care should be taken not to induce premature drug release under conditions of elevated gastric pH). For this purpose, formulation approaches could also be employed. For instance, a formulation approach involving a double coating technology with the dosage form being surrounded by a bilayer composed of an outer conventional enteric coat and an inner coat composed of partially neutralized enteric polymer, and an organic-acid based buffer system has been proposed [54],[55],[56]. This system achieved considerable improvement in reducing the time to onset of dosage form disintegration postgastric emptying in vivo [55],[56]. The mechanism behind it was an increase in the buffer capacity and ionic strength to which the outer conventional enteric coat is exposed through the action of the partially neutralized polymer's carboxylates and the organic salts in the inner coat [54],[55],[56].

Yet another important aspect that needs to be taken into account is that the *in vitro* tests are designed based on the physiological conditions encountered in the intestines of healthy subjects. However, some populations, like patients with exocrine pancreatic insufficiency, might have lower proximal small intestinal pH [57], and those considerations for disease state patients have to be taken into account to conduct *in vitro* studies for its meaningful *in vivo* prediction.

1.3.3. Colon-targeted dosage forms

In some cases, the enteric coat is not intended to dissolve in the proximal small intestine but rather to delay drug release until the distal small intestine or the colon is reached. A classic example is formulations of mesalamine, a drug used to treat ulcerative colitis and Crohn's disease. Targeting drug release to the distal parts of the GI tract will ensure maximal drug concentration at the inflammation site in the lower small intestine and colon. Another potential application for colon targeting is the per-oral delivery of therapeutic proteins and peptides since it will result in them being released to a region of relatively lower proteolytic activity and thus reduce their pre-systemic degradation.(though the lower permeability in the large intestine poses another challenge) [58]. Film-formers with a dissolution pH threshold of more than 7.0 are typically used for this purpose. Examples include Eudragit S, Eudragit FS and shellac.

A multi-stage dissolution test is needed for such products to show that they manage to withhold their drug load in both the stomach and the proximal and middle small bowel. This approach is employed by the FDA-recommended dissolution testing methods for mesalamine colon-targeted pH-dependent products as listed in Table 4 [59]:

Table 4: Dissolution Testing Methods for Mesalamine Colon-Targeted pH-Dependent Products

Drug Name	**Dosage Form**	**USP Apparatus**	**Speed (RPMs)**	**Medium**	**Volume (mL)**	**Recomme-nded Sampling Times (minutes)**	**Date Updat-ed**
Mesalamine	Capsule (Delayed Release)	II (Paddle)	Phase 1 & 2: 100 rpm; Phase 3: 50 rpm	Phase 1: 0.1N HCl (degas); Phase 2: pH 6.0 Buffer (degas); Phase 3: pH 7.2 Buffer	Phase 1: 500; Phase 2: 900; Phase 3: 900	Phase 1: 120; Phase 2: 60; Phase 3: 20, 30, 45, 60, 75, 90 and 120	06/30/ 2016
Mesalamine (1.2 gram)	Tablet (Delayed Release)	II (Paddle)	100	Acid stage (A): 100 mM HCl Buffer stage (B): Phosphate Buffer, pH 6.4 Buffer stage (C): Phosphate Buffer, pH 7.2	Acid stage (A): 750 mL; Buffer stage (B): 950 mL; Buffer stage (C): 960 mL	Acid stage (A): 2 hours; Buffer stage (B): 1 hour; Buffer stage (C): 1, 2, 4, 6 and 8 hours	06/10/ 2009
Mesalamine (400 mg and 800 mg)	Tablet (Delayed Release)			Refer to USP			11/05/ 2010

For the 400 and 800 mg delayed release tablet products, the FDA recommends the USP-proposed method. This method is made up of a first two-hour stage in 0.1 M HCl to simulate gastric conditions, a second one-hour stage in a pH 6.0 buffer to simulate

the proximal small intestinal conditions followed by a third stage in a pH 7.2 buffer to simulate the distal bowel where the major part of drug release should occur.

The USP method outlined above suffers from two serious shortcomings:

1. It specifies one hour to simulate exposure to proximal small intestinal conditions followed immediately by the distal small intestinal conditions where drug release should take place. Thus it ignores the exposure of the dosage form to middle small intestinal conditions where the dosage form will face a higher pH than in the proximal bowel [30].
2. The pH 7.2 buffer used to simulate distal GI conditions has a buffer capacity of approximately 70 mmol L^{-1} pH^{-1} calculated using the van Slyke equation.[60] This is more than ten times the experimental value reported in literature (6.4 mmol L^{-1} pH^{-1}) for ileal fluid [30]. Therefore, drug release *in vitro* could be expected to be generally much faster than *in vivo* (especially for API with acidic functionalities like mesalamine).

The same media are recommended for the capsule products. The media for the high strength (1200 mg) tablet products are different with regard to the first buffer stage (pH 6.4 instead of pH 6.0 making it less unsuitable to ensure that the dosage form will not release the API in the middle small bowel).

In addition to media-related shortcomings, the absence of mechanical stresses matching those found in the human intestine (a problem not restricted to dissolution testing of colon-targeted dosage forms) also adversely affects the ability of compendial methods to predict *in vivo* performance [61].

These issues might contribute to the extremes of product performance variability that have been observed with mesalamine delayed release products. For instance, in a study by Varum et al, despite exhibiting consistently quick release when tested according to the USP dissolution method for mesalamine delayed release tablets, ileo-colonically targeted Eudragit S prednisolone tablets exhibited highly variable disintegration performance *in vivo* with 2 out of 10 tablets being voided intact [62]. Moreover, the USP test failed to distinguish between the performance of two different formulations [62]. This variability is further illustrated by the results of a bioequivalence study[63] as shown by selected individual pharmacokinetic profiles in Figure 4. The figure clearly shows the presence of strong variability at both intra- and inter-individual levels with occasions on which no or little drug can be quantified in the plasma. These findings are supported by reports of tablets being egested intact that have even been mentioned in the prescribing information of Asacol® mesalamine delayed release tablets [64]. Employing more biopredictive *in vitro* testing methodologies could reduce the frequency of such occurrences through being more able to identify inadequately formulated products.

Dynamic physiological bicarbonate-based buffers the pH of which is adjusted by automated computer-controlled systems have been proposed for dissolution of mesalamine delayed release dosage forms.[65] Indeed, the ability of a static pH 7.4 bicarbonate-based Krebs buffer to discriminate between two ileo-colonically targeted

formulations with different *in vivo* performance, in contrast to the USP method [62], shows that such systems are worth further research and evaluation. However, as discussed in the previous subsection, bicarbonate buffer is difficult to use for the routine screening of oral products. A possible solution could be using an approach similar to the one employed by Al-Gousous et al. for non-colon targeted enteric-coated dosage forms to develop a simple phosphate-based system where the formulation performance matches that in such a bicarbonate system. So, when fully optimized, dynamic bicarbonate systems can be useful standards for developing such phosphate buffers.

As for mechanical stresses, Garbacz et al. have proposed a modified dissolution tester that incorporates presumably biorelevant mechanical stresses (and even combined it with the use of abicarbonate-based buffer).This is achieved through placing the dosage form in a chamber made of stainless steel netting that is immersed in the dissolution vessel. Mechanical stresses are introduced through pre-programmed inflation and deflation of a balloon inside the chamber and pulses of intense rotation of the chamber [61]. They found that this device could, potentially, predict instances of premature drug release that cannot be predicted by a conventional paddle apparatus showcasing the significance of the mechanical stresses in drug release from such dosage forms.

However, it should be noted that so far the design of *in vitro* testing methodologies to evaluate EC dosage forms has been done based on the conditions in healthy individuals, and the major application of colon-targeted dosage forms is to treat patients with inflammatory bowel disease (ulcerative colitis and Crohn's disease) where the physiological conditions (like pH) in the intestinal lumen could be different from those in a healthy human. Regarding the ileo-colonic pH in inflammatory disease, the studies published so far gave conflicting results with some studies showing decreased pH, others showing increased pH and others showing no difference from controls [66]. A summary of these results is provided in a review by Nugent et al. [66]. The authors of that review concluded that the small numbers of subjects studied so far make it difficult to definitively establish the influence of these diseases on GI pH [66]. Therefore, further studies involving large numbers of subjects are necessary.

Active inflammatory bowel disease introduces another deviation from conditions in healthy subjects. The presence of diarrhea leads to reduced contact time between the drug product and the patient's intestine. This will reduce the time window during which the dosage form can release its load of drug. Therefore, more effort needs to be dedicated to develop *in vitro* methodologies that are clinically relevant.

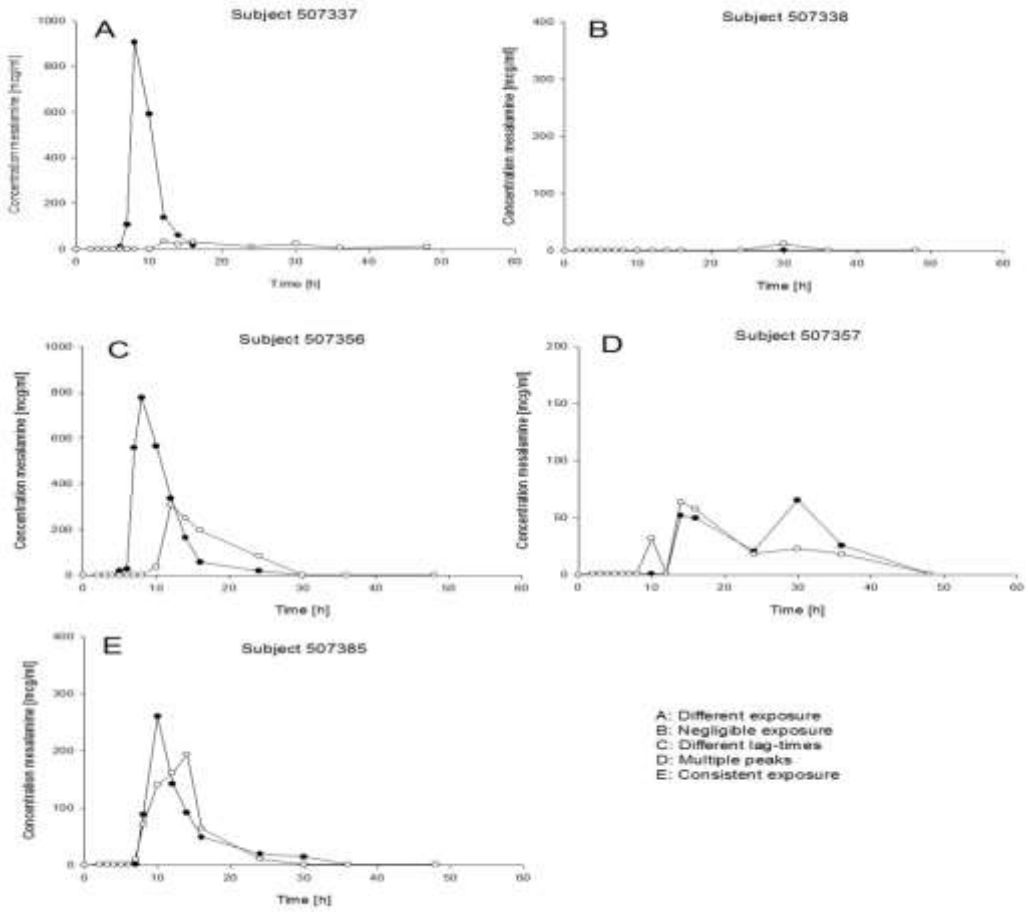


Figure 4: Examples on inter- and intra- individual variation in mesalamine absorption from a delayed release dosage form (Asacol 400 mg tablets). For explanations of the effects see insert.

1.4. Effect of food on EC-coated dosage forms

First of all, ingestion of food disrupts the MMC cycle thus delaying the emptying of larger single unit dosage forms [67]. The duration of this disruption has been shown to primarily depend on the caloric value of the meal [68]. As for multiparticulate dosage forms, it is believed that pellets/beads with a diameter not exceeding 2 mm can pass together with food through the contracted pylorus, and therefore, their emptying is less affected by food [67]. However, a study points out that this is the case only in some subjects, while in others pellet emptying is delayed until the stomach becomes nearly empty and the MMC cycle is restored [69]. The investigators concluded that restricting data evaluation to average profiles misled scientists into believing that pellets smaller than 2 mm empty together with food [69]. An additional issue is that under fed conditions, a dosage form is consistently subjected to high pressure events inside the stomach in contrast to the fasted state where it is subjected to a smaller number of highly variable pressure events [70]. When also taking into account the pH changes described later, this could contribute to an increased risk of premature drug release in the stomach.

As for small intestinal transit time, data are contradicting. Some data indicate no significant effect of food on small bowel transit time [71], while other data indicate accelerated small intestinal transit time with food in some subjects [72]. This contradiction could be explained by the findings of Fadda et al. where it was found that when food, given 45 min postdose, arrived at the stomach after a nondisintegrating tablet was emptied, the small bowel transit was accelerated, while food that arrived at the stomach before the tablet being emptied (as well as food administered before dosing) did not significantly impact the small bowel transit time. 54 The effects of possible acceleration of small intestinal transit can have different effects depending on the properties of the active ingredient. For example, in a study on enteric-coated erythromycin tablets, some subjects exhibited very fast small bowel transit times that led to the tablet reaching the cecum still undisintegrated [72]. There, in the large intestine, the tablets underwent very slow disintegration and dissolution resulting in poor bioavailability.

In addition, food leads to the presence of elevated pH in the stomach [30] making it more difficult for the dosage form to prevent release into the stomach (especially when taking into account that the larger number of high pressure events in the fed stomach increases the chance of dosage form softening resulting in coat rupture and premature drug release). Food also lowers the pH of the proximal small intestine [30] making it more difficult for the dosage form to release its load there quickly, but, however, it increases the local buffer capacity [30], which could have an opposing effect (depending on the exact value of the pH).

As for selecting pH values for simulating the fed state during in vitro dissolution studies (assuming healthy GI tract), Jantratid and Dressman have proposed a medium with pH 5.0 and buffer capacity of 25 mM/pH to represent fed state gastric fluid and pH 5.8 with a buffer capacity of 25 mM/pH to represent fed state proximal small intestine [73]. These values generally agree with what has been reported concerning the in vivo situation except for possibly overestimating the jejunal buffer capacity in the fed state (reported to be 13.2−14.6 mM/pH) [30].

For these reasons the patient instructions for many enteric-coated products instruct the patients to take the medicine on empty stomach. However, a particular enteric-coated product, pancreatin, must be taken together with meals since its function is to provide replacement for the pancreatic digestive enzymes for patients with exocrine pancreatic insufficiency.

With pancreatin there is the additional challenge that, in subjects with pancreatic insufficiency, the duodenal pH might be lower than normal due to impaired pancreatic bicarbonate secretion [57]. Indeed, it has been proposed that the incomplete resolution of digestive symptoms of exocrine pancreatic insufficiency with pancreatin therapy is due to its strongly delayed release from the available formulations [74] owing to the lowered proximal intestinal pH in these patients. Therefore, designing enteric coatings, the release performance of which is more independent of food and of lowered proximal intestinal pH, could help improve the clinical outcomes of pancreatin therapy.

Another issue is that of interactions between enteric coatings and specific foodstuffs and nutrients. For instance, it has been known that enteric film formers are liable to dissolve prematurely in the presence of ethanol. That is why the FDA requires dissolution testing to be also performed in 0.1 M HCl containing 5, 20, and 40% v/v alcohol USP [75]. However, according to the findings of a recent study by Rubbens et al., the peak ethanol gastric fluid concentration was between 2.6 and 7.3% after drinking 200 mL of wine and between 6.3 and 21.1% after drinking 80 mL of ethanol [76]. This indicates that the recommendation to perform dissolution testing with 40% ethanol might be too strict. However, the small number of subjects involved (five subjects) does not allow a definite judgment to be made.

Recently, in vitro data have been published regarding possible interaction between enteric coatings and calcium leading to retarded disintegration due to reduced coat solubility through ionic cross-linking by calcium ions [77]. This issue needs to be thoroughly investigated to ascertain the extent of the interaction in vivo since it could have clinical significance in the context of coingestion of calcium-rich foods as well as in the context of coadministration of calcium supplements.

There are additional food effects that are relevant, albeit not particular, to EC dosage forms. These include increased viscosity of the gastric and proximal small intestinal fluids and pellicle formation by food components around the disintegrating dosage form delaying disintegration [19].

2. Aims

Despite their widespread use, comparative studies of HPMC-based hard capsules with different enteric polymers are lacking, in particular when considering methods without the use of a sealing step. This step closes the gap between the cap and the body and is generally recommended to avoid leaking of the capsule content into the stomach and vice versa. Thus, the comparative evaluation of coated capsules in the present investigations should include their enteric properties, batch-to-batch consistency, disintegration in pharmacopoeial and biopredictive media and stability. In this work, particular emphasis was put on identifying a formulation with comparatively rapid drug release following gastric emptying. Since biopredictive disintegration test conditions are still in the process of evaluation, a new release testing medium [45] as well as the official compendial fluids were thus employed for disintegration testing. In this respect, a study was performed that showed good correlation between the disintegration and the dissolution properties of EC hard capsules, thus justifying the use of disintegration tests.

3. Evaluation of formulation factors on disintegration time of enteric-coated capsules

3.1. Materials and Methods

3.1.1. Materials

Size 0 hydroxypropyl methylcellulose (HPMC) capsules prefilled with maltodextrin (450 mg of maltodextrin per capsule) were received from Lallemand Inc. (Montreal, Canada). Hypromellose acetate succinate (HPMCAS, Shin-Etsu AQOAT®) and hypromellose phthalate (HPMCP) were received as gifts from Shin-Etsu (Shin-Etsu Chemical Co. Ltd., Japan). HPMCAS was used as -LG, -MG and -HG (different substitution grades of the polymer) [78]and HPMCP was used as HP-50, HP-55 and HP-55S[79]. Methacrylic acid and ethyl acrylate copolymer (EU 100-55, Eudragit®) was used as L100-55 and obtained as free sample from Evonik (Darmstadt, Germany). Cellulose acetate phthalate NF (C-A-P, Aquateric®) was received from Eastman via Krahn Chemie GmbH (Hamburg, Germany). Triethyl citrate (TEC) was purchased from Sigma-Aldrich (Bornem, Belgium) and talc (particle size 0.8-35 µm) was obtained from Luzenac SE (Imerys, France). All other chemicals were of analytical grade.

3.1.2. Methods

3.1.2.1. Coating formulations

The preparation of the coating solution is illustrated by the example of HPMCAS polymer (Shin-Etsu AQOAT®). All other polymer solutions were prepared following this method and are described in Table 5-Table 8. First, HPMCAS was dissolved in ethanol (96 % v/v) and stirred for 0.5 h at room temperature. Talc was homogeneously dispersed in approximately 150 mL purified water and afterwards combined with the polymer solution. Next, TEC was added to the dispersion. After 1.5 h of ambient stirring the dispersion was filtered through a 200 um sieve (VWR). During the coating experiment the dispersion was stirred gently to prevent sedimentation of talc. Based on dry polymer mass the HPMCAS- and HPMCP-formulations differed in plasticizer content while the talc content stayed constant. For the C-A-P-formulations the plasticizer content was constant, and the amount of talc varied. The Eudragit batches differ in their processing parameters. (spray rate and coating time as well as inlet and outlet processing temperatures).In formulation E02 a coloring agent (cherry red food dye, Roth GmbH, Bochum) was used. Due to its low concentration (Table 5) the influence on the mechanic properties and disintegration time is expected to be negligible. Viscosity of coating formulations was determined using a Brookield LV rotational viscometer at 22-23.5 ℃.

Table 5: Composition of HPMCAS coating dispersion. All concentrations are given in percent (w/w).

Series	HPMCAS				
Batch No.	E02	E06	E04	E09	E18
Polymer type	AS-MG	AS-MG	AS-MG	AS-LG	AS-HG
content	4.99	5.00	5.00	5.00	5.00
TEC	1.25	3.00	2.00	2.00	2.00
Talc	7.49	7.50	7.50	7.50	7.50
Water	17.23	16.90	16.50	16.50	16.50
Ethanol 96% (v/v)	68.95	67.60	69.00	69.00	69.00
Food color	0.09	0	0	0	0
Total	100.10	100.00	100.00	100.00	100.00

Table 6: Composition of HPMCP coating dispersion. All concentrations are given in percent (w/w).

	HPMCP							
Batch No.	E03	E08	E07	E21	E11	E12	E10	E17
Polymer type	HP-55S	HP-55S	HP-55S	HP-55S	HP-55	HP-55	HP-50	HP-50
content	6.00	6.00	6.00	6.00	6.00	6.00	6.00	6.00
TEC	0.00	0.60	1.20	1.50	0.00	2.40	0.00	3.00
Talc	7.50	7.50	7.50	7.50	7.50	7.50	7.50	7.50
Water	12.97	17.90	17.00	17.00	12.97	17.90	12.97	16.70
Ethanol 96% (v/v)	73.53	68.00	68.30	68.00	73.53	68.00	73.53	66.80
Total	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00

Table 7: Composition of Eudragit® coating dispersion. All concentrations are given in percent (w/w).

	Methacrylic acid and ethyl acrylate copolymer	
Batch No.	E15-02	E15-03
Polymer type	EU 100-55	EU 100-55
Content	6.00	6.00
TEC	1.20	1.20
Talc	3.00	3.00
Water	14.00	14.00
Ethanol 96% (v/v)	75.80	75.80
Total	100.00	100.00

Table 8: Composition of C-A-P coating dispersion. All concentrations are given in percent (w/w).

	Cellulose acetate phthalate	
Batch No.	E19	E20
Polymer type	C-A-P	C-A-P
content	6.00	6.00
TEC	2.00	2.00
Talc	0.00	3.00
Water	46.00	44.50
Ethanol 96% (v/v)	46.00	44.50
Total	100.00	100.00

The prefilled HPMC capsules were coated in a Solidlab 1 drum coater (Hüttlin GmbH, Schopfheim, Germany) with different polymer formulations. Each batch consisted of 700 ml (0.368 kg) capsules. The process parameters are described in Table 9 and Table 10. The weight gain corresponds to a coating thickness of the dry coating formulation of 10 mg x cm^{-2}.

Table 9: Operating parameters for coating of HPMC capsules with enteric polymers on a Solidlab 1 drum coater.

	Shin-Etsu AQOAT®/HPMCP	Eudragit® L100-55	Aquateric®
Before coating			
Preheating to ℃	30	25	30
Coating			
Nozzle diameter (mm)	0.5	0.5	0.5
Spray rate (g/min)	6.5-7	E15-02:2.7-2.8 E15-03:1.2-1.3	7
Atomizing pressure (bar)	2.0	0.5	2.0
Inlet air volume (m^3/h)	55	60	50
Inlet air temperature (℃)	58-60	40-75	55
Product temperature (℃)	35-38	25-40	37
Coating time (min)	65	181-310	80
Drum speed (r/min)	30	30	30

*Table 10: Processing parameters for each formulation. The order in the table is given by the type of polymer used, starting with HPMCAS series (E02-E18), followed by HPMCP series (E03-E17) and Eudragit (E15-02 – E15-03) and CAP (E19-E20). *Viscosity of enteric polymer coating solution at 22-23.5 ℃ at 30 rpm stirring rate. Weight gain is based on polymer mass.*

Batch name	Viscosity* (mPas)	Spray rate (g/min)	Inlet temp. (℃)	Outlet temp. (℃)	Weight gain (%)
E02	32.6±0.1	6.5-7.0	57-64	38-39	7.10
E06	32.6±0.1	6.9-8.0	59-62	39-41	7.20
E04	32.6±0.1	6.5-8.5	58-60	39-40	7.15
E09	29.6±0.1	6.3-7.4	58-60	39-40	7.26
E18	40.2±0.2	8.6-10	57-60	38-40	7.23
E03	99.2±0.2	6.5-7.3	58-60	39-40	6.95
E08	99.2±0.2	7.2-9.0	59-60	39-41	7.21
E07	99.2±0.2	6.6-8.2	59-64	39-42	7.06
E21	99.2±0.2	7.1-7.9	58-60	40-41	7.12
E10	48.1±0.2	6.0-7.1	59-61	39-42	7.17
E17	48.8±0.2	7.0-8.9	58-60	39-40	7.09
E11	46.0±0.2	5.3-6.7	59-64	39-40	7.28
E12	47.5±0.2	6.7-8.2	58-62	38-41	7.25
E15-02	45.5±0.2	2.7-2.8	73-80	45-48	7.40
E15-03	45.2±0.2	1.2-1.3	35-40	24-31	7.50
E19	41.5±0.2	4.4-4.9	55-62	39-40	7.05
E20	47.2±0.2	4.6-5.0	46-52	35-38	7.13

3.1.2.2. Disintegration test

Enteric-coated capsules were tested in a USP conform disintegration tester (DT2, SOTAX AG, Aesh, Switzerland) according to the USP chapter <701>. Disintegration test was performed first in an acidic media for one hour followed by a buffer media with near neutral pH at 37±0.5 ℃. The volume of the disintegration media was 700 mL 0.1 N hydrochloric acid was used as acidic media. For the buffer media *Simulated Intestinal Fluid* (SIFsp) pH 6.8 as well as a 15 mM phosphate buffer pH 6.5 (*Al-Gousous medium*), that previously demonstrated biopredictivity for enteric-coated tablets [45], were used. The disintegration times recorded are the times at which the capsules ruptured, which helps reduce the uncertainty associated with determining the disintegration times based on "complete disintegration"[80]. Disintegration tests were performed with disks unless otherwise specified.

3.1.2.3. Acid Uptake test

6 capsules of each batch were accurately weighed (W_0) and exposed to 0.01 N HCl or 0.1 N HCl for one or two hours at 37 ℃ in a disintegration apparatus (see above). Thereafter, the capsules were removed and blotted dry to remove excess liquid on the capsule surface and reweighed (W_t). The percent acid uptake was calculated from the capsule weight difference before and after the test according to equation (1).

$$\%\ \text{acid uptake} = \frac{W_t - W_0}{W_0} \times 100\ \% \qquad (1)$$

3.1.2.4. Scanning electron microscopy (SEM)

SEM analysis was performed on a JEOL JSM-IT100 scanning electron microscope equipped with a secondary electron detector in high vacuum mode. The analysis was performed on the body-cap-joint part of the capsules, as well as on the flat face of the cap part using an acceleration voltage of 1 kV and probe current of 30% without prior modification of the sample (e.g. sputtering).

3.1.2.5. Statistical analysis

Comparison of different formulation effects on the disintegration time was done a two-way ANOVA for independent samples using the VassarStat website[81]. First factor (rows) were the two different buffers teste and the second factor (columns) were the tested formulation factor (e.g. plasticizer content). p-values < 0.05 were consider as significant.

3.1.2.6. Mechanical properties of the capsules

The mechanical properties of coated capsules were tested on a TA.XTplus Texture Analyzer (Stable Micro Systems, Godalming, UK) equipped with a HDP/90 Heavy Duty Platform table (flat insert with target on one side) and 1 cm P/1 KS Kobe Stainless punch. Basic principles of the test including data analysis and interpretation

have been published previously [82]. Briefly, the force (N) and the deflection (mm) were recorded during the test procedure. The data were then converted to stress and strain by Exponent software (Stable Micro Systems, Godalming, UK). The tensile strength of the film at the fracture point was determined. The area under the stress-strain curve was calculated to obtain the tensile toughness of the film-coated capsule. Six capsules from each lot were tested. Non-coated capsules were tested as reference material. Fracture points were obtained for both, the film and the capsule.

3.2. Results and Discussion

3.2.1. Surface structure of the enteric-coated capsules

All formulations could be applied successfully on the capsules and formed a film coating (see Figure 5) although there are visible differences between the different formulations. Within the capsules coated with HPMCAS-MG (E02, E04 and E06) and HP-50 (E10 and E17) an increasing amount of TEC results in a smoother and brighter surface. The surface difference was not observed with the HP-55 (E11 and E12) and the HP-55S (E03, E07 and E08) coating. Among the HPMCP series, HP-55S shows the best performance among regarding the surface structure of the coating and good sealing of the junction between cap and body of the capsule. This is most probably related to its higher degree of polymerization (higher viscosity) resulting in higher entanglement and accordingly superior film formation. Actually, HP-55S resulted in the best sealing of the gap among all the studied polymers. Between the L100-55 formulations (E15-02 and E15-03) there were no differences observed. Within the C-A-P coatings, talc seems to have a beneficial effect on the surface structure. With a higher talc concentration, the gap between the body and cap of the capsule is closed. This could be related to the bulking effect of the added talc.

Figure 5: Surface structure of enteric-coated HPMC capsules. % TEC and talc are based on dry polymer weight.

More detailed SEM images were prepared to evaluate the gap closing performance of selected formulations with different polymers (Figure 6) and the body parts surface structures of some coated formulations (Figure 7). Similar to the results above, the HP-55S (E03) formulation shows the best sealing of the gap. The film in formulation E03 is smooth and the gap is completely close. In formulation E10 and E11 the gap is clearly visible and could cause problems in the acid resistance. Coatings of the formulation E15-03 and E04 are closing the gap properly, although it is still apparent. Considering the C-A-P (E19 and E20) formulations, talc seems to have a positive effect on the gap closing. In formulation E20 the gap is almost completely closed.

These differences in the gap closing can influence the acid resistance of the coated capsules. For example, the HP-55S and Eudragit formulations which show the best sealing of the gap also exhibit the lowest acid uptake values (Figure 8).

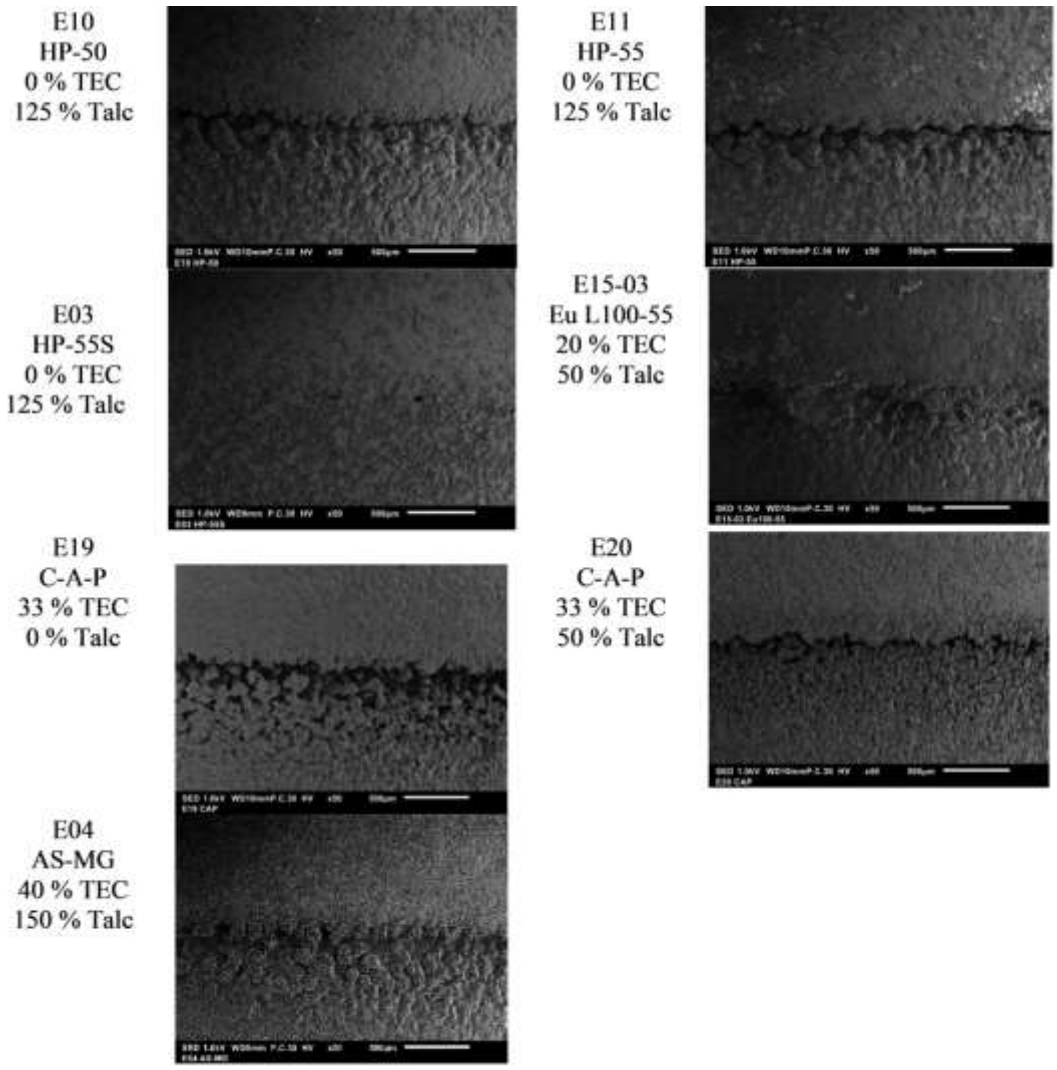


Figure 6: SEM images displaying the gap junction between capsule top and bottom after coating. % TEC and talc are based on dry polymer weight.

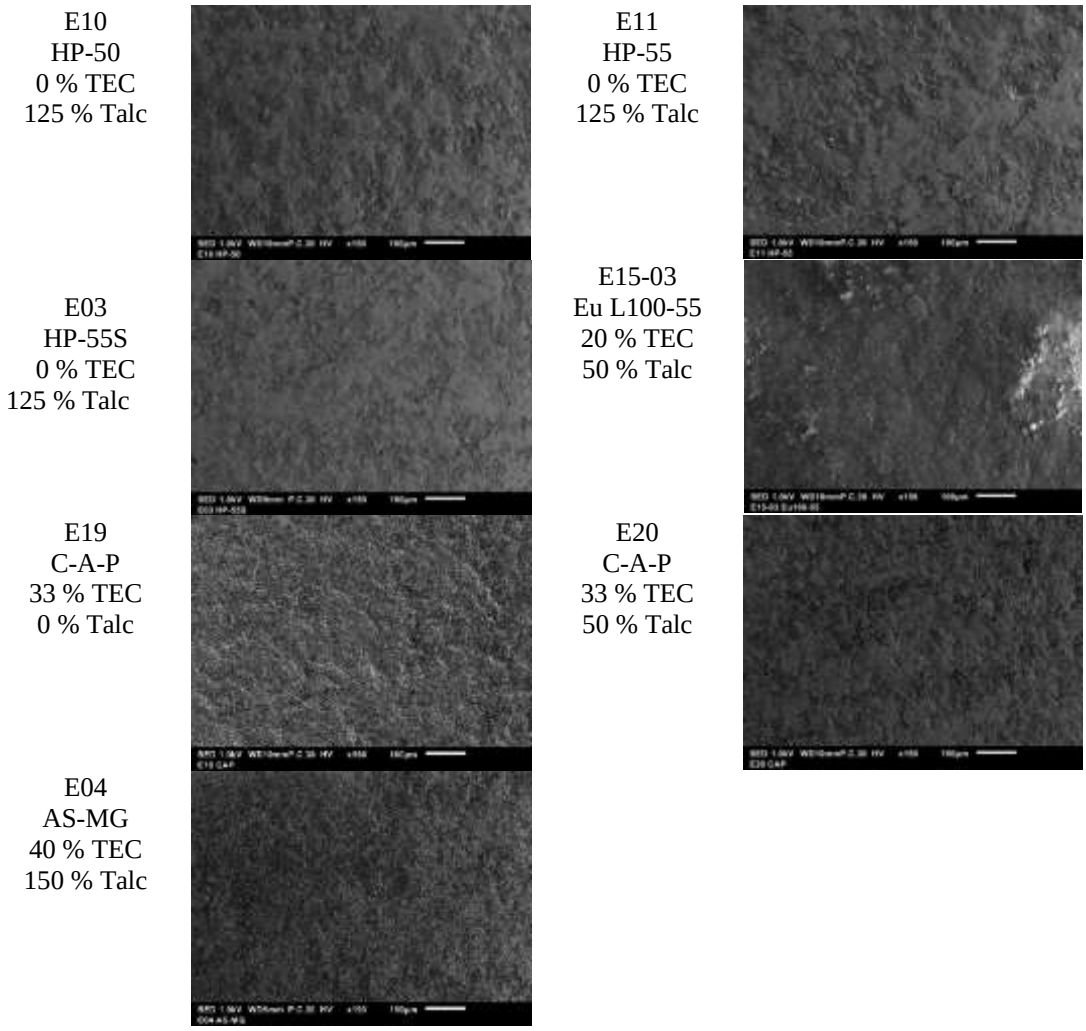


Figure 7: SEM images displaying the capsule body after coating. % TEC and talc are based on dry polymer weight.

3.2.2. Disintegration test and acid uptake

After one and two hour of disintegration testing in acidic media all capsules remained intact with no sign of coat rupture. An effective enteric coating results in low values of acid uptake and values up to 10 % acid uptake have been shown to be acceptable for protecting highly acid-labile drugs such as proton pump inhibitors (PPIs) [83]. As shown in Figure 8, all formulations showed a sufficient acid resistance at both pH 1 and 2 with an uptake of < 10 % over 2 h of testing. HPMCP HP-55S showed the lowest liquid uptake as would be expected from the images in Figure 6. Good results were also obtained with the Eu L100-55 and AS-HG (E18) formulations. These results are in good agreement with the observations from section 3.2.1 where these formulations sealed the gap between the capsules body and cap fairly well (Figure 6). As expected, the uptake after two hours increased compared to one hour of acid treatment. Comparing the results at pH 1 and pH 2 all capsules showed similar values.

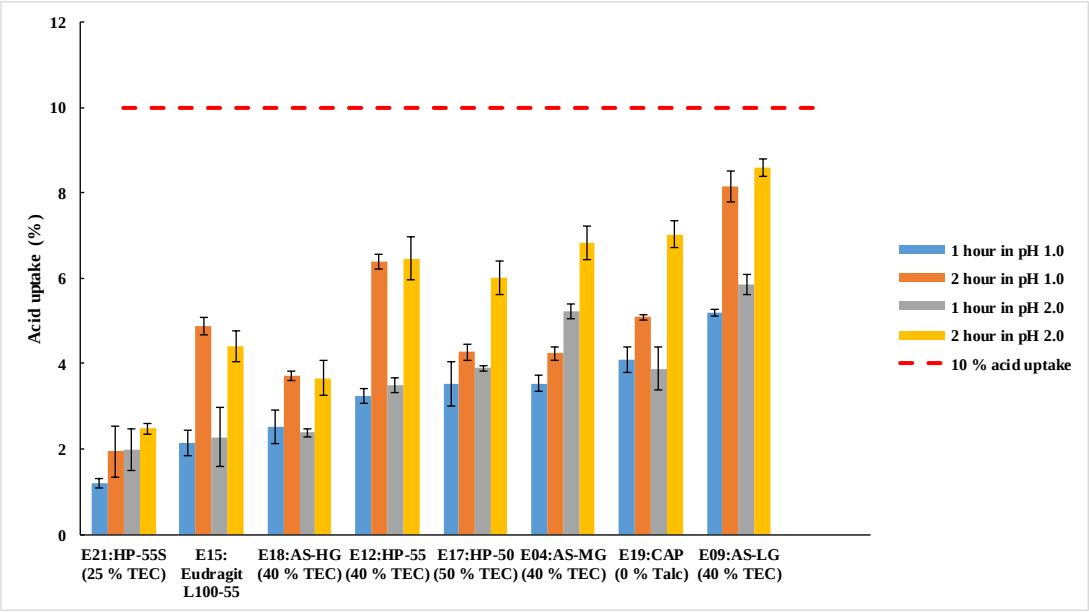


Figure 8: Acid uptake of enteric coated HPMC-capsules following incubation in 0.1N HCl and 0.01N HCl. Horizontal line represents 10 % acid uptake. Given are average acid uptake (%) ± SD; n=6. % TEC and talc are based on dry polymer weight.

The disintegration times in phosphate buffered media are depicted in Figure 9. All capsules showed a faster disintegration in the USP media due to the higher buffer capacity of the buffer [4],[45],[84]The capsules coated with HP-50 (E10 and E17) exhibit the fastest disintegration in both media, as expected. Among the tested polymers HP-50 has the lowest dissolution pH-threshold (>pH 5.0, Table 1) and a lower dissolution pH is generally associated with faster polymer dissolution, although other factors like molecular weight and viscosity of the gel layer during dissolution are influencing the dissolution rate as well. For capsules coated with Eu L100-55, HP-55S and AS-HG the longest disintegration times were the observed.

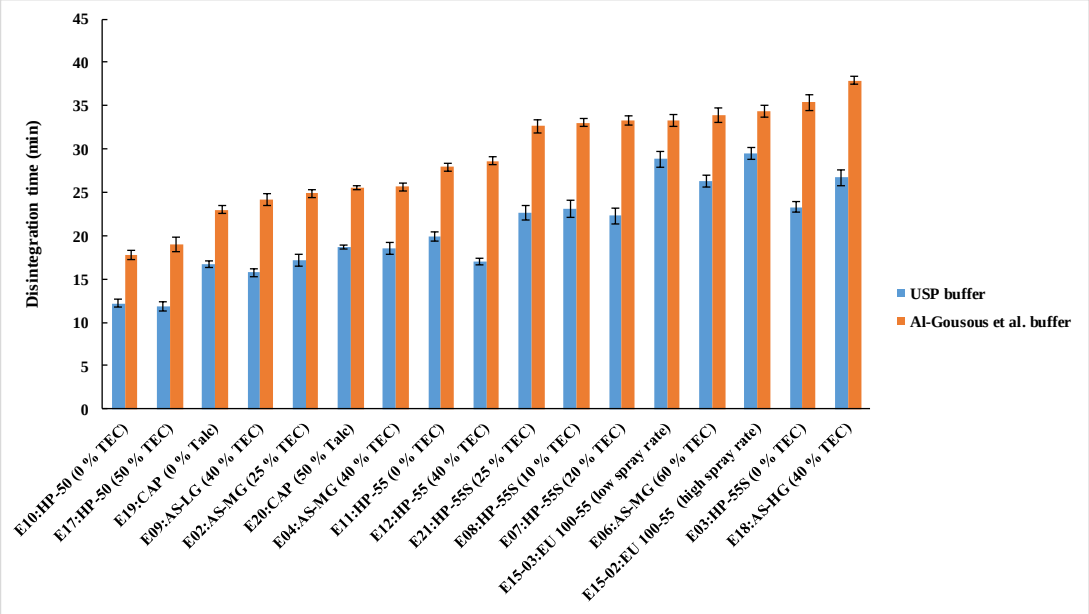


Figure 9: Disintegration times of enteric-coated capsules in 50 mM USP phosphate buffer pH 6.8 and 15 mM phosphate buffer pH 6.5 (Al-Gousous et al. buffer) without disc. Given are average disintegration times ± SD; n=6. % TEC and talc are based on dry polymer weight.

While uncoated capsules tend to dissolve at the end parts of the capsule first (not shown), this behavior may change when coated capsules are disintegrating. On the one hand, the coated capsule may begin to disintegrate at the gap part first. This is found with most tested capsules and exemplary shown in Figure 10 A and B. In this case, the initial disintegration of HPMCAS coated capsules is depicted. However, depending on the type of polymer used, the site of initial disintegration can move to the top or bottom parts as shown in Figure 10 C and D for capsules with intact gap coverage between top and bottom parts. Figure 6 showed that HPMCP can close the gap well. Therefore, the gap is not the weakest part of the capsule and the disintegration starts at the ends of the capsule.

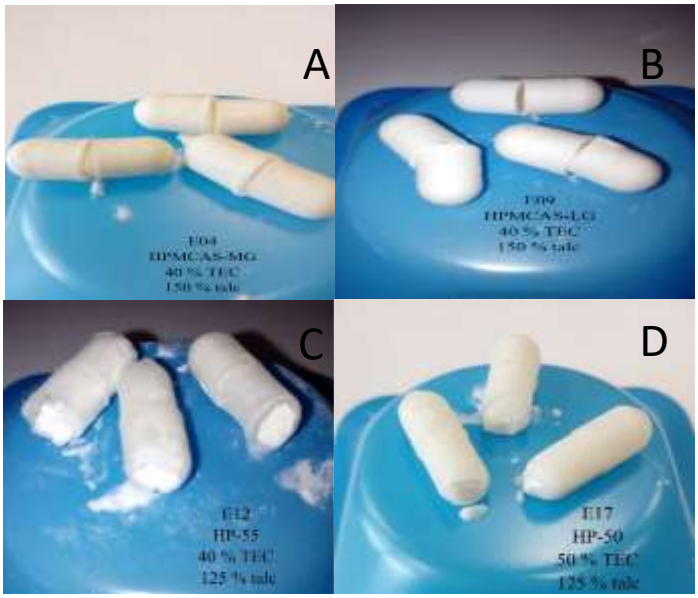


Figure 10: Appearance of capsules coated with different HPMCP and HPMCAS grades as they start disintegrating.

3.2.3. Comparison of formulation-specific factors on disintegration time

Formulation-specific factors at different levels were grouped and compared by statistical analysis, in order to distinguish between significant versus non-significant observations with respect to the effect of coating composition on enteric coated capsule disintegration times. Analysis was performed using two-way ANOVA.

3.2.3.1. Effect of different polymer grades on disintegration time

The effect of the substitution pattern of HPMCAS and HPMCP was investigated on three formulations for each polymer that just differ in their polymer grade (E09, E04 and E18 and E10, E11 and E03).

Figure 11 shows the disintegration time of the HPMCAS capsules. The observed disintegration times are significantly different (p-values < 0.05) between all grades of HPMCAS. In both buffers the rank order of the disintegration times is similar: AS-LG < AS-MG < AS-HG. It is noteworthy, that there is only little difference between the LG and MG disintegration times in both buffers. The polymers AS-LG, AS-MG and AS-HG have dissolution pH of 5.5, 6.0 and 6.5, respectively [8]. Therefore, the difference between all polymers could be expected to be similar. The varying amounts of acetyl (LF < MG < HG)- and succinoyl (HG < MG < LG) -groups are responsible for different dissolution pH thresholds. An increase in acetyl groups and reduction in succinoyl groups increases the hydrophobicity of the polymer. As consequence, the polymer requires ionization to a higher degree to become soluble, resulting in a higher opening pH value[85]. Figure 12 shows the acetyl- and succinoyl distribution between the polymers. The AS-L- and AS-M-grade have very similar contents whereas the AS-H-grade shows a larger difference to both other polymers which is in line with the experimental disintegration data.

That the solubility and disintegration behavior are not exactly mirror images of each other is not surprising, for the former is a thermodynamic parameter while the latter is a function of a kinetic phenomenon (polymer dissolution). And the dissolution of a polymer (which is a prerequisite for disintegration) is not solely dependent on the polymer solubility behavior and but also on other polymer properties like the diffusional and rheological behavior of polymer chains.

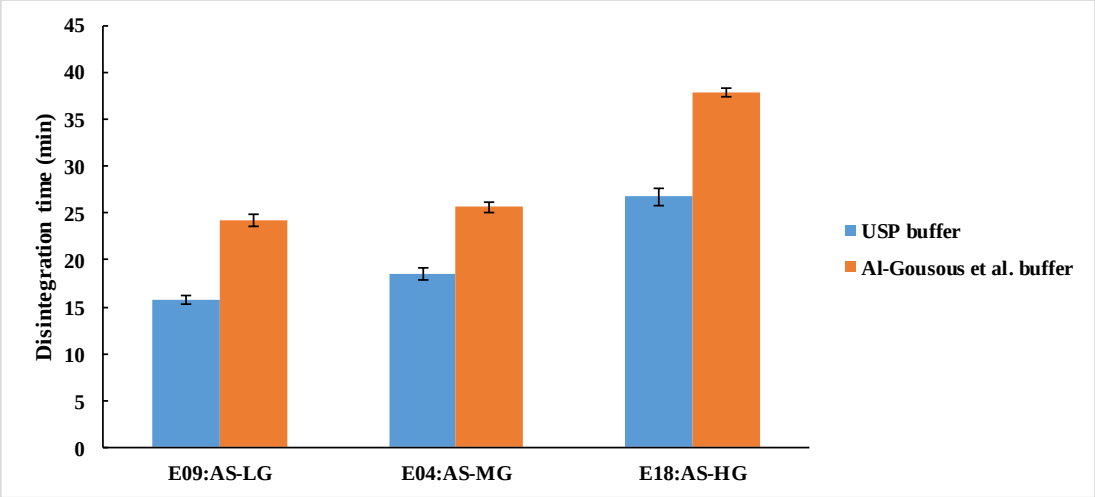


Figure 11: Disintegration time of capsules coated with HPMCAS of different grades (LG, MG and HG). Given are average disintegration times ±SD; n=6.

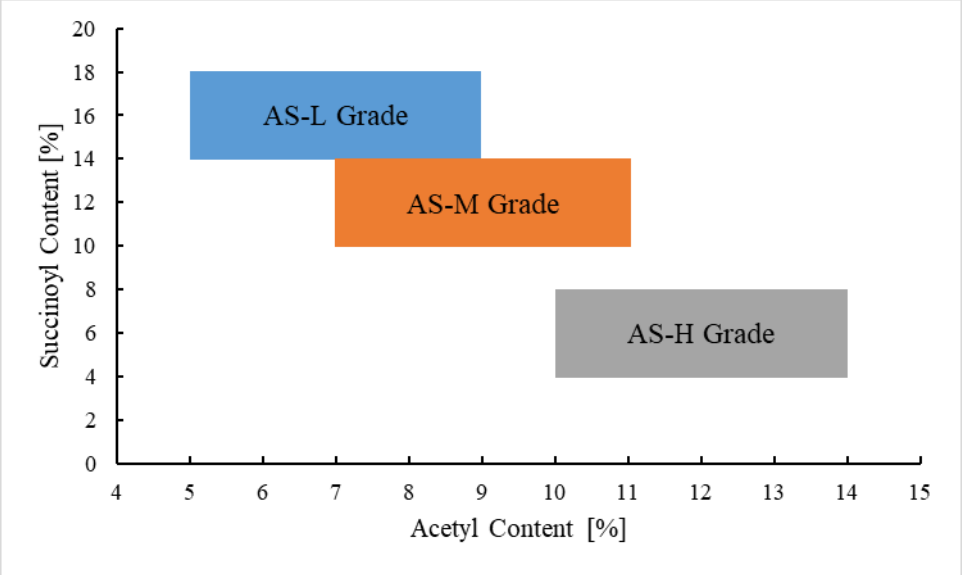


Figure 12: Acetyl- and succinoyl content of the different HPMCAS-grades [8][86]

As shown in Figure 13, HPMCP was tested in three different grades: HP-50, HP-55 and HP-55S. HP 50 has a phthalyl content of 24 % and a dissolution pH of 5.0 whereas HP-55 and HP-55S have a phthalyl content of 31 % and a dissolution pH of 5.5 [79]. The relationship between phthalyl content and dissolution pH threshold is inverse to the acetyl/succinoyl content in HPMCAS. With more ionizable groups the dissolution pH is increased. A possible explanation lies in the bulky hydrophobic nature of the introduced aromatic ring overcoming the solubilizing effect of additional ionizable groups. Referring to the dissolution pH, as expected the HP-50 disintegration time is lower compared to the HP-55/HP-55S disintegration times. It must be emphasized the difference in disintegration times of HP-55 and HP-55S was smaller when tested in the USP buffer while the difference is obvious in the more biorelevant Al-Gousous et al. buffer, although all differences between the formulations were significant (p-value < 0.05). The difference in disintegration time

between HP-55 and HP-55S is due to the molecular weight (resulting in different viscosities, Table 1) of these two polymers. Polymers of the same chemical structure but with higher molecular weight tend to dissolve slower because of greater entanglement and higher gel viscosity[87].

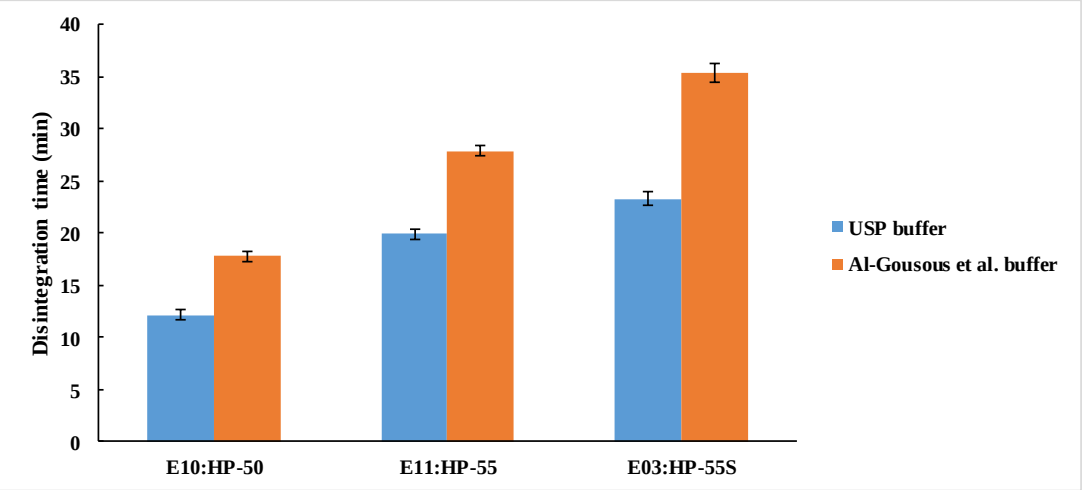


Figure 13: Disintegration time of capsules coated with HPMCP of different grades (HP-50, HP55 and HP-55S). Given are average disintegration times ±SD; n=6.

3.2.3.2. Effect of plasticizer content on disintegration time

TEC is often used as plasticizer in pharmaceutical coatings due to its GRAS-status [88]. TEC is a hydrophilic plasticizer which often increases the dissolution of coated formulations. This is in line with the disintegration times of the HP-55S formulations (Figure 14B) in the 15 mM phosphate buffer. Although there seems to be a limit for the acceleration of dissolution. The difference in disintegration time in the 15 mM phosphate buffer between 0 % TEC and 10 % TEC is significant while the difference between 10 % TEC and 20 % TEC is not. Unexpectedly, the disintegration time increased with increasing TEC content in the HPMCAS-MG formulations (Figure 14A). This was observed in both buffers tested. As for the HP-55S polymer, there is a limit at which the disintegration time is not affected by the plasticizer concentration anymore. The disintegration time of the HP-55 and HP-50 capsules (Figure 14C, D) does not change with the tested TEC concentrations. Due to the higher molarity the USP buffer is less discriminative and could not point out the TEC influence on disintegration for the HP-55S formulation which showcases the importance of using biopredictive buffers. The ANOVA testing with post hoc analysis showed significant differences in the disintegration times of the HP-55S formulations in the 15 mM phosphate buffer, where the higher TEC content led to a slightly faster disintegration most probably because of the pore forming effect of the water-soluble TEC, while this trend was not detected in the USP buffer.

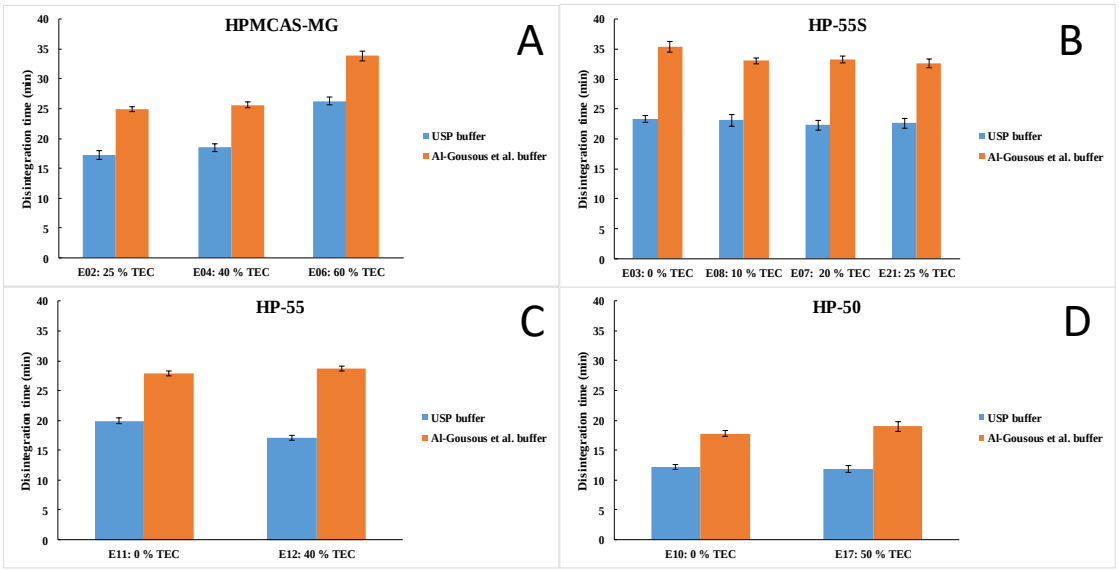


Figure 14: Effect of plasticizer content on the disintegration time in different formulations. A: HPMCAS-MG formulations. B: HP-55S formulations. C: HP-55 formulations. D: HP-50 formulations. Given are average disintegration times ±SD; n=6. TEC content is based on dry polymer weight.

3.2.3.3. Effect of talc-content in C-A-P formulations on disintegration time

In pharmaceutical coatings, talc is used as anti-tacking agent. Because of its lipophilicity high amounts of talc can delay the disintegration and dissolution of solid oral dosage forms. On the other hand, talc can have a positive effect on sealing the gap of coated hard capsules (see E19 and E20 in Figure 5). The tested formulations showed only little influence of the different talc concentrations, although these findings were significant (p-value < 0.05). The hydrophobicity of talc seems to exert a rather weak effect on disintegration time most probably because of the talc particles being coated by polymer chains which reduces their negative effect on wettability.

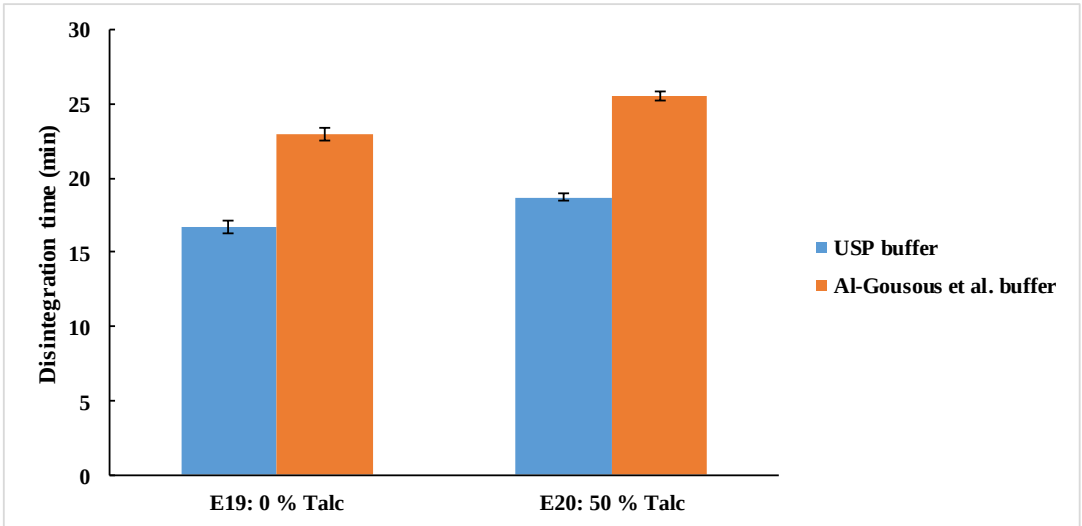


Figure 15: Disintegration time of C-A-P formulations with different talc content. Given are average disintegration times ±SD; n=6. Talc content is based on dry polymer weight.

3.2.3.4. Effect of different processing parameters on the disintegration time of Eudragit L100-55 formulations

The parameters of the coating process can have various effects on the film formation and therefore on the disintegration and dissolution of a coated dosage form [85]. The influence of two spray rates at different process temperatures on the disintegration time was evaluated. As can be seen in Figure 16 the disintegration times of the two batches are almost similar (difference approx. 1 min, p-value > 0.05). Since the inlet air temperature of both batches was above the minimum film forming temperature (MFFT) of 30-35 ℃ [89] there were homogeneous films formed. As long as the coating thickness is comparable and there is no spray drying observed the disintegration time should not be affected by the different spray rates when the process temperature is above the MFFT.

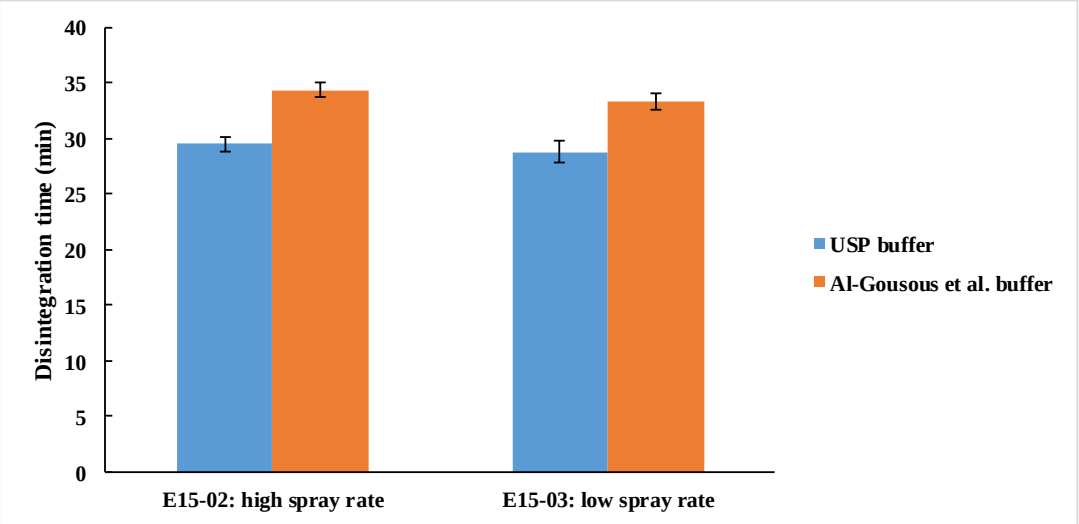


Figure 16: Effect of different coating parameters on the disintegration time. Given are average disintegration times ±SD; n=6. Effect of disc vs sinker on the disintegration time.

3.2.4. Difference between disintegration test performed with disc vs without disc but with sinker

In disintegration testing a disc is often added to the setup to prevent the dosage form from floating and may introduce added mechanical stress on the dosage forms. To investigate whether the mechanical stress on the capsule because of the disc has an influence on the disintegration time, further disintegration tests were performed without disc but with sinker. The resulting disintegration times are depicted in Figure 17. In the USP buffer only little differences between the disintegration time of the single formulations were observed. In the biorelevant Al-Gousous et al. buffer only the HPMCAS-MG capsules showed differences in disintegration time. With increasing concentrations of plasticizer, these differences increased.

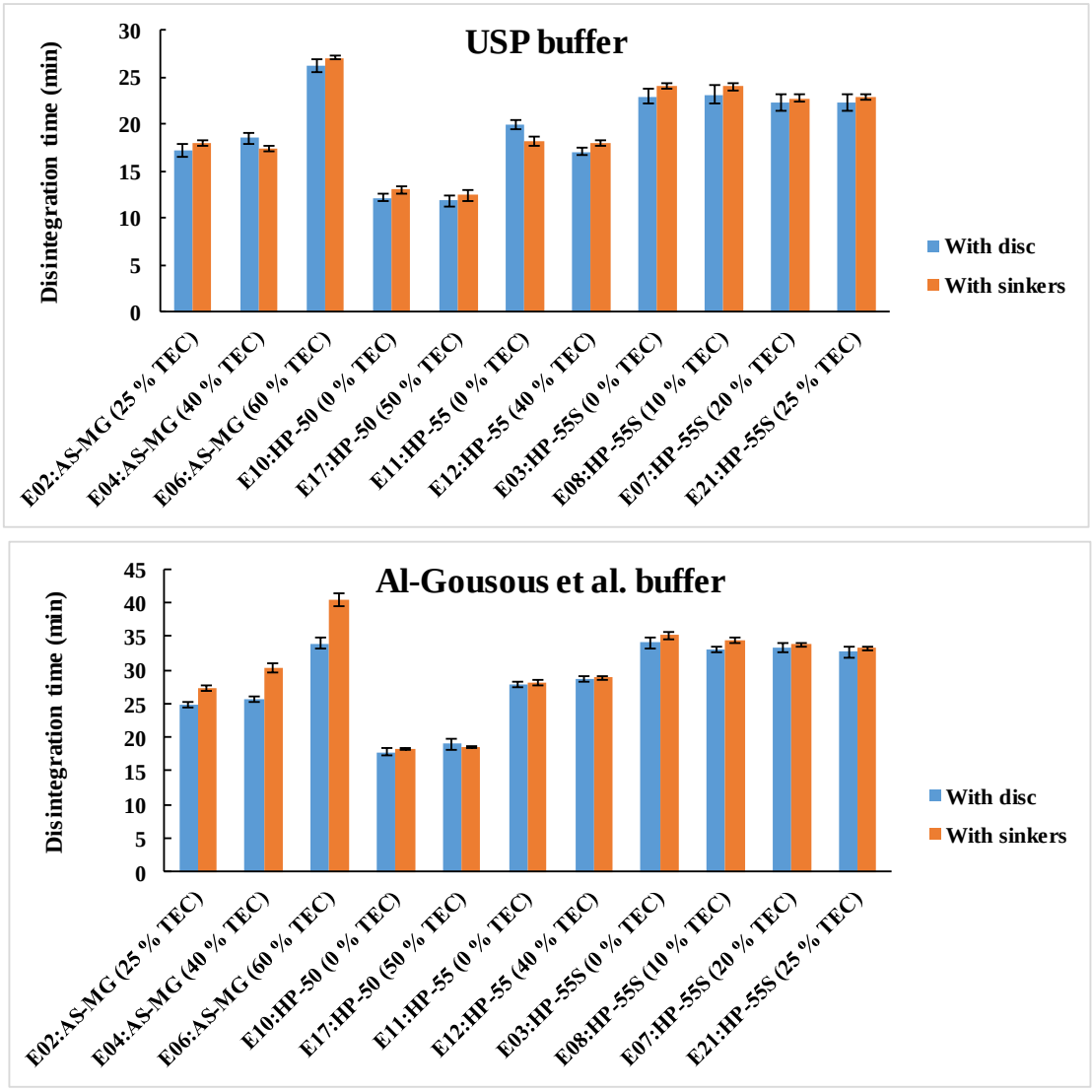


Figure 17: Comparison of disintegration test with disc vs. without disc but with sinker. Given are average disintegration times ±SD; n=6. TEC content is based on dry polymer weight.

3.2.5. Mechanic properties of coated capsules

Enteric-coated capsules were tested for their mechanic properties. In Figure 18A the % deflection (based on the outer diameter of the capsules (76.5 mm)) is shown. For most polymers a trend between increasing TEC concentration and increased deflection is observed. TEC increases the flexibility of the polymer. Therefore, the deflection is increased. Although for some formulations the effect was negligible. Figure 18B depicts the breaking force (N) that was needed to break the capsule. The HPMCAS-MG formulation shows the expected results that with increasing flexibility less force is required to break the capsule. The HPMCP HP-55 formulation shows an inverse trend. A higher TEC content results in a higher breaking force. The C-A-P formulation shows that an increased talc concentration requires a higher breaking force. This is due to the incorporation of solid talc particles in the coat which increases the mechanic stability. The work done (N*mm) until the breaking point is depicted in Figure 18C. Here, no trend between TEC or talc concentration and work done was observed.

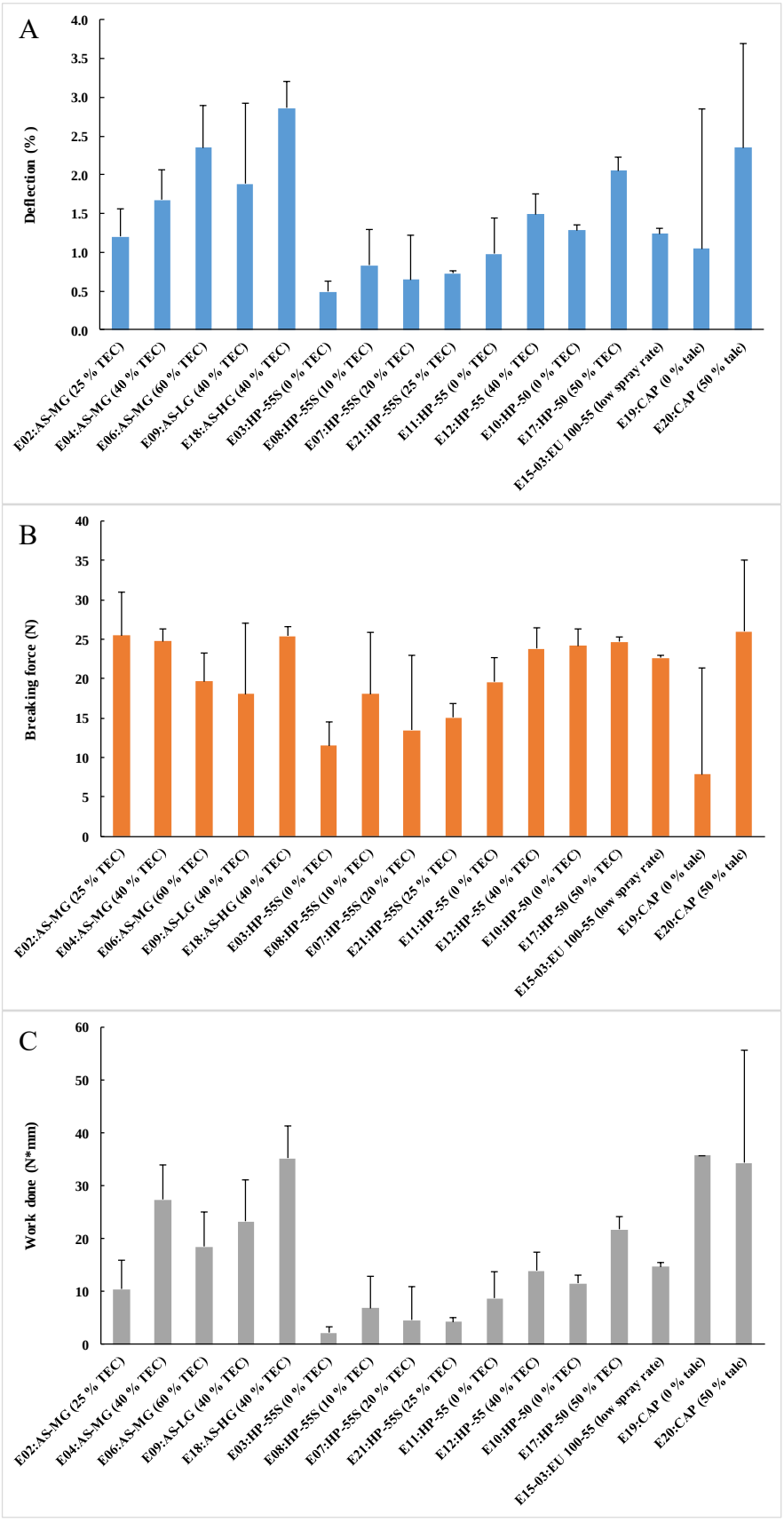


*Figure 18: Mechanic properties of the tested capsules. A: % deflection based on the capsule diameter (76.5 mm). B: breaking force (N). C: Work done until the capsule breaks (N*mm).*

Figure 19A shows the deflection (%) and breaking force (N) of the HPMCAS-MG capsules. As mentioned above, the deflection until the capsule breaks is increased but the force required is decreased. Comparing these values with the observed disintegration times a good correlation in all tested buffers was found (Figure 19C, D).

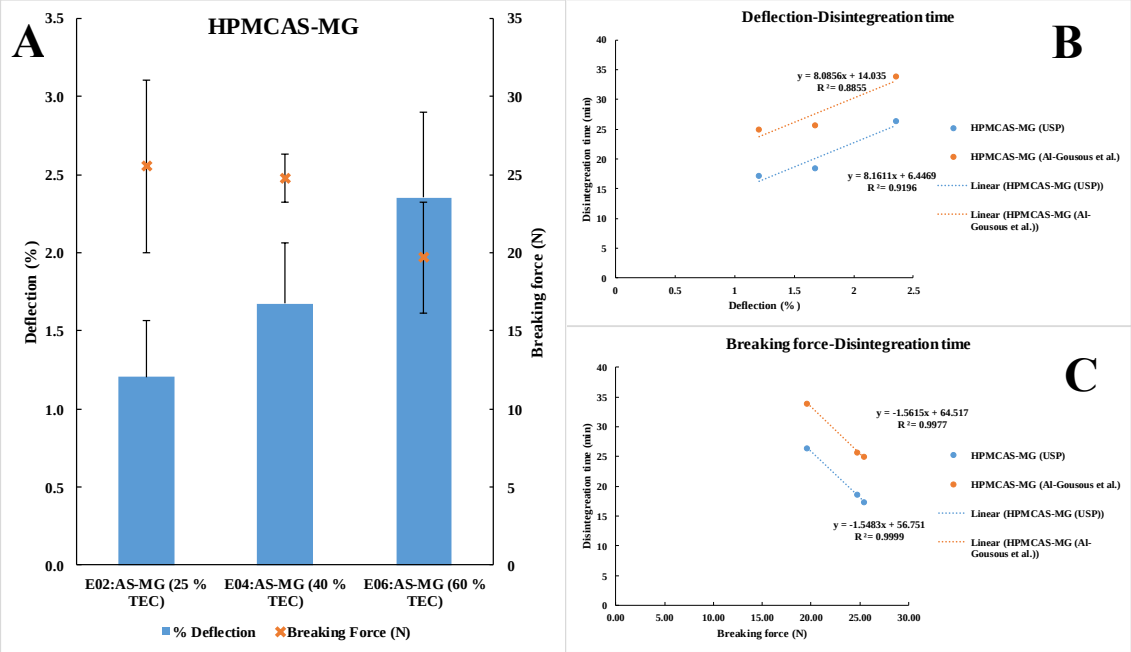


Figure 19: A: Mechanic properties of capsules coated with HPMCAS-MG. Given are the average deflection (% based on capsule outer diameter (76.5 mm)) and the Breaking force. ± SD; n=6. TEC content is based on dry polymer weight. B Correlation between disintegration time and deflection. C: Correlation between breaking force and deflection.

This provides a potential insight toward explaining the disintegration times' increasing with %TEC for the HPMCAS-MG-based formulations. The effect of the presence vs the absence of disks indicate that mechanical properties can present a significant factor affecting the said disintegration times. When correlating that to the mechanical properties measured by Texture Analyzer, good correlations between disintegration times and the deflection at break was obtained (Figure 19). Therefore, it might be presumed that the enhanced film flexibility imparted by increased TEC content had a retarding effect on the disintegration time that overweighed the accelerating effect of the pore formation by the leaching of the water-soluble TEC. No type of correlation could be obtained for the breaking force and energy values. This might have to do with the fact that texture analysis was performed with the capsules being in a dry state, whereas they are soaked with water during the disintegration testing. The resulting confounding effects brought about by the plasticizing effects of water and the leaching of the water-soluble TEC impair the degree to which the measured mechanical properties correlate with the disintegration behavior.

4. The relationship between disintegration and dissolution testing of caffeine enteric-coated HPMC capsules

In order to justify the use of disintegration tests in the previous sections, a study investigating the correlation between dissolution and disintegration was performed.

4.1. Materials and Methods

4.1.1. Materials

Hydroxypropyl methylcellulose size 0 capsules (ACG Nature Caps Plus) were received from ACG Associated Capsules Pvt Ltd (Mumbai, India). DRcaps® (nutraceutical capsules with inherent enteric properties of the capsule shell) were obtained from Neue Lebensqualität (Badendorf, Germany). Caffeine (median particle size 48 µm) and magnesium stearate were purchased from Caesar & Loretz GmbH (Hilden, Germany). Fumed silica was purchased from Fagron GmbH & Co KG (Glinde, Germany). Hypromellose phthalate (HP-50), hypromellose acetate succinate (HPMCAS-HG, AQOAT) and low substituted hydroxymethyl cellulose (L-HPC) were received as a gift from Shin-Etsu (Wiesbaden, Germany). Lactose (FlowLac® 90) was received from Molkerei Meggle (Wasserburg, Germany). Triethyl citrate (TEC) was purchased from Sigma-Aldrich (Overijse, Belgium). Talc was acquired from Imerys (Luzenac, France). All other materials were of analytical grade.

4.1.2. Capsule filling

The HPMC capsules and DRcaps® were filled manually (Aponorm® Kapselfüllgerät, WEPA Apothekenbedarf GmbH & CO.KG, Hillscheid, Germany) with a powder formulation. The powder formulation was prepared using a 1.6 L Turbula mixer (Willy A. Bachofen GmbH, Muttenz, Switzerland) at 49 rpm (batch size 600 g), and its composition is described in Table 11. The capsules were filled with 375 mg of the powder (i.e. 75 mg of caffeine). The filled capsules complied with the content uniformity requirements of the European Pharmacopoeia 9.0.

Table 11: Formulation of the capsule filling. All values (%) refer to the total weight (m/m).

Substance	**% (m/m)**	**weights of the components per capsule (mg)**
Caffeine	20	75
L-HPC	15	56.25
Lactose	63.75	239.06
Silica	0.25	0.94
Magnesium stearate	1	3.75
Total	100	375

4.1.3. Capsule coating

The ACG Nature Caps Plus capsules have been coated. Two different batches of coated capsules were prepared from them. The composition of coating formulations is given in Table 12. First, the ethanol–water solution was prepared. The polymer was dissolved in 80% of the solvent and the remaining 20% of the solvent were used to disperse the talc. Afterwards, the polymer solution was combined with the talc dispersion. In the last step, triethyl citrate was added to the formulation (in case of the HPMCAS-HG formulation). Before coating, the polymer solution is filtered using a sieve with a pore size of 0.2–0.4 mm. The coating levels of the capsules coated with HP-50 and HPMCAS-HG are 10 mg polymer/cm^2 and 9 mg polymer/cm^2 (30% and 27% weight gains), respectively. A Solidlab 1 drum coater (Robert Bosch Packaging Technology GmbH, Waiblingen, Germany) was used for coating with the following parameters: 230 g of capsules/batch preheated to 30 ℃; spray rate of 6.5–7 g/min and an atomizing pressure of 2.0 bar; nozzle diameter of 0.5 mm; the inlet air was heated to 58–60 ℃ and had a flow rate of 55 m^3/h; product temperature was maintained at 35–38 ℃.

Table 12: Coating formulation. All values (%) refer to the total solution

Substance	HP-50 formulation		HPMCAS-HG formulation	
	% (m/m)	weights of the components (g)	% (m/m)	weights of the components (g)
Polymer	6	60	5	40
Talc	7.5	75	7.5	60
TEC	-	-	2	16
Ethanol (96% (v/v))	69.2	747.7	68.4	591.25
Water	17.3	117.3	17.1	92.75

4.1.4. Disintegration test

The capsule disintegration was performed with disks using a DT2 Disintegration Tester (Sotax AG, Aesh, Switzerland) which corresponded to the European Pharmacopoeia specifications for a type A disintegration testing apparatus. All capsules were first exposed to 700 mL of 0.1 M HCl for one hour followed by one hour testing in 700 mL of buffer. The temperature was kept at 37.0 ± 0.5 ℃. There were three different buffers tested, namely the 50 mM USP phosphate buffer pH 6.8, blank FaSSIF buffer (28.4 mM) pH 6.5 and a 15 mM phosphate buffer pH 6.5 that showed to be bio-predictive in previous studies (henceforth referred to as the "Al-Gousous et al. medium"[45]). Disintegration times recorded are the times at which the capsules ruptured, which helps to reduce the uncertainty associated with determining the disintegration times based on "complete disintegration" [80]. Accordingly, these times are defined as the times at which first visible cracks in the capsule shell appear. In order to avoid observer distortion, disintegration tests were carried out before the dissolution tests.

4.1.5. Acid Uptake Test

Six capsules were individually weighed and then tested in a disintegration tester as outlined above (the Disintegration Test subsection) but without disks, and only in HCl (0.1 and 0.01 M) for one and two hours. Sinkers (Japanese Pharmacopoeia Standard, Pharma Test, Hainburg, Germany) were used to prevent the capsules from floating. At the end of the test the capsules were removed, blotted and the percent weight gain was calculated as described previously by equation 1.

$$\%\ weight\ gain\ in\ acid = \frac{mass\ after\ acid\ exposure - mass\ before\ acid\ exposure}{mass\ before\ acid\ exposure} x\ 100\ \%$$

4.1.6. Dissolution test

The drug release was tested with a DT6R dissolution tester (Erweka GmbH & CO.KG, Langen, Germany). The device was used as a USP type I dissolution tester at 100 rpm as well as a USP type II dissolution tester with sinkers (same as in previous subsection) at 50 rpm. The use of sinkers prevented the capsules from floating in the paddle apparatus. In accordance with the disintegration test, the capsules were studied for one hour in 0.1 M HCl followed by a media change to either one of the buffers described previously. The volume of the dissolution media was 700 mL. The temperature was maintained at 37.0 ± 0.2 °C. The 5 mL samples were filtered through a 0.8 μm cellulose acetate nitrate filter (Rotilab Spritzenfilter CME, Carl Roth, Karlsruhe, Germany). The first 1 mL of the filtrate was discarded to saturate the membrane. Blank buffer was used to replace the sample volume. Caffeine was quantified spectrophotometrically at λ= 275 nm.

4.1.7. Correlation between disintegration and dissolution results

Disintegration time was correlated with the time required to achieve 10, 50 and 80% release (t10%, t50%, and t80%, respectively), which represent the early, middle and late part of the dissolution profiles. The above times were calculated by linear interpolation. The correlation was performed using a simple linear regression performed by Microsoft Excel (Microsoft Office 2013). Hypothesis testing was done on the slope using a one-sided t-test (with the null hypothesis being slope = 0). One-sided p-values were calculated because a positive correlation is expected. The hypothesis tests were performed using the vassarstats website [81].

4.2. Results and Discussion

4.2.1. Disintegration and Acid Uptake

As shown in Figure 20, DRcaps® Enteric gave the fastest disintegration while HPMCAS-HG gave the slowest. Disintegration tended to be fastest in the USP dissolution testing medium and slowest in the Al-Gousous et al. medium as would be expected based on the buffer molarities of the media. The fast disintegration of DRcaps® Enteric, however, appears to be associated with poor resistance to acid as evidenced by the acid uptake values shown in Table 13 and by the deformation exhibited by those capsules (Figure 21). This shows that it is rather the weakened capsule shell structure that results in rapid disintegration in buffer. This is rather in line with the findings of Al-Tabakha et al. [80], in which rupture of such capsule shells in the simulated gastric fluid was observed.

Interestingly, the HPMCAS-HG capsules ruptured into acid within about 1.5 to 2 hours, despite the lowest acid absorption after one hour during the acid absorption tests. This could be related to mechanical instability. Figure 20 shows that the DRcaps® show a strong deformation, but the HP-50-coated ones show only a little wear at the gap between the body and the cap. HPMCAS-HG-coated capsules behave similarly to the HP-50 -coated capsules, but the wear at the gap appears to be somewhat greater, which may impart mechanical instability to the capsule. The causes behind this need to be further investigated.

As shown in Figure 20, after 1 hour in acid, the capsules coated with HPMCAS-HG still show the longest disintegration times. Only the high buffer capacity USP medium [45] showed considerable acceleration in disintegration compared to the situation where testing in acid was continued for one further hour (in the acid uptake tests with sinkers and without disks). This further supports the hypothesis of mechanical instability. Since the presence of disks in the disintegration test (compared to their absence in the acid uptake test) does not seem to have a dramatic effect. This could be due to the fact that the disk hits the capsule from above instead of tearing it apart. Other factors could be the force generated by the contact between the capsules and the sinks in the disc-free structure, the inclined orientation of the capsules in the disintegration tester tubes inside the sinkers and their possible effects on hydrodynamics. Additional investigation is needed regarding this issue, which is outside the scope of current research.

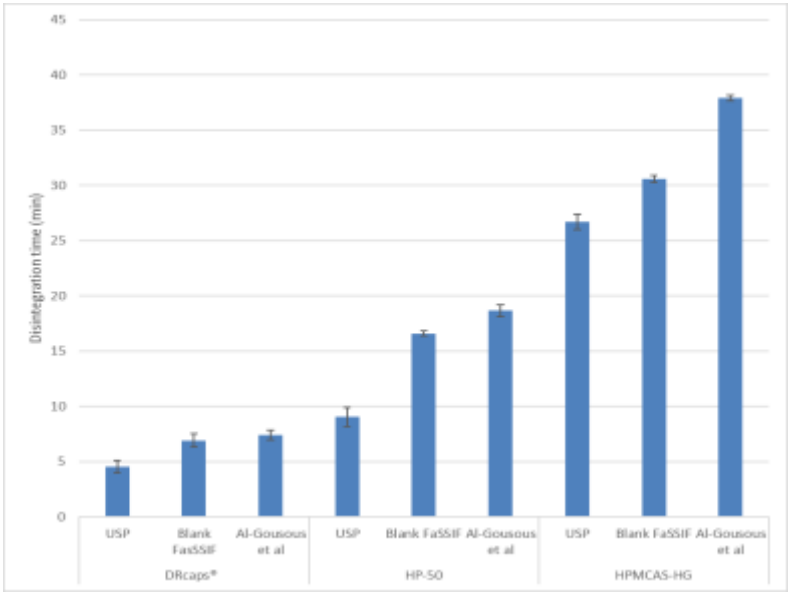


Figure 20: Disintegration time (mean ±SD) of the formulations tested (n = 6).

Table 13: Weight gain (mean ±SD) of the formulations tested (n = 6) after 1 h in acidic media.

Formulation	% Weight gain in 0.1 M HCl		% Weight gain in 0.01 M HCl	
	After 1 h	After 2 h	After 1 h	After 2 h
DRcaps®	6.5 ±0.7	7.4 ±0.3	11.2 ±0.2	13.5 ±0.6
HP-50	3.4 ±0.6	4.2 ±0.3	3.8 ±0.1	6.1 ±0.7
HPMCAS-HG	2.5 ±0.4	Ruptured	2.8 ±0.6	Ruptured

Figure 21: The appearance of DRcaps® and coated capsules after 1 hour in 0.1 M HCl.

4.2.2. Dissolution

As shown in Figure 22, the dissolution results followed the trends exhibited by the disintegration times. The disintegration times are lesser even than the t10% values, that is likely related to the capsule rupture being a pre-requisite for significant drug release and with the bigger mechanical stresses within the disintegration tester [38]. Dissolution seems slower in the basket equipment. This could be related to the lower fluid velocities within the central and higher regions of the basket (at 100 rpm) wherever the capsule tends to be located owing to its buoyancy compared to the bottom of the vessel in a paddle equipment (at 50 rpm) [90]. This suggests that hydrodynamic differences between the various apparatuses play a significant role within the obtained correlations.

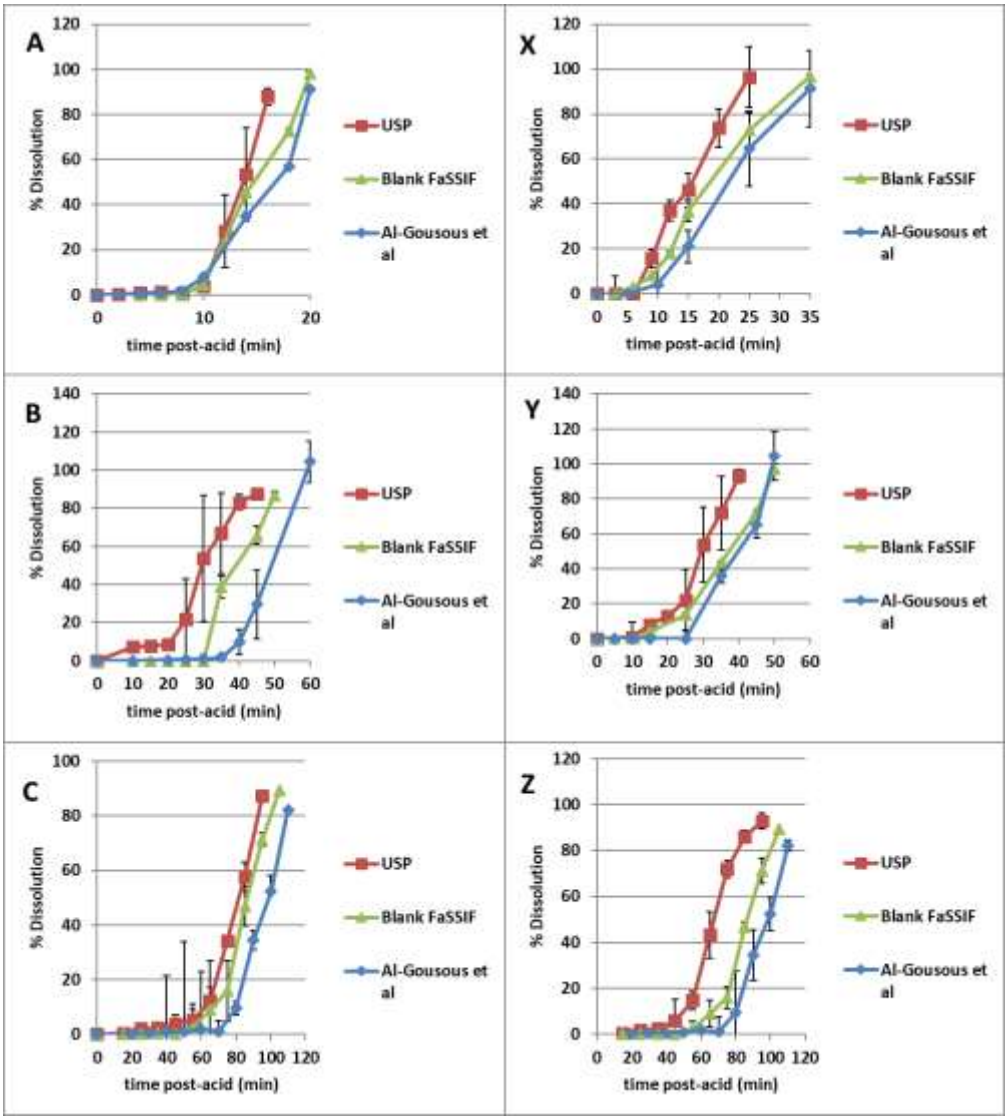


Figure 22: Dissolution test results (mean ± SD) of the formulations tested (n = 6). Panels A, B, and C represents DRcaps®, HP-50, and HPMCAS-H in basket apparatus, while the panels X, Y, and Z represent DRcaps®, HP-50 and HPMCAS-H in paddle apparatus.

4.2.3. Correlation between disintegration and dissolution results

Disintegration times have been correlated with the times required to gain 10, 50 and 80% release (t10%, t50% and t80% respectively) representing the early, middle and late quantities of the dissolution profiles. When all the dissolution times were correlated with their respective disintegration times, strong correlations were acquired for all dissolution profile quantities (Figure 23).

A more detailed analysis was achieved by way of making three point correlations for formulation effects (Figure 24 and Figure 25) and medium effects (Figure 26 and Figure 27). When the results of different formulations tested in the identical medium were correlated, good r^2 and p-values had been almost invariably received. However, the situation was different whilst correlating outcomes of the identical system in unique media (Figure 27 and Figure 28), where the differences tended to be smaller than the inter-formulation variations.

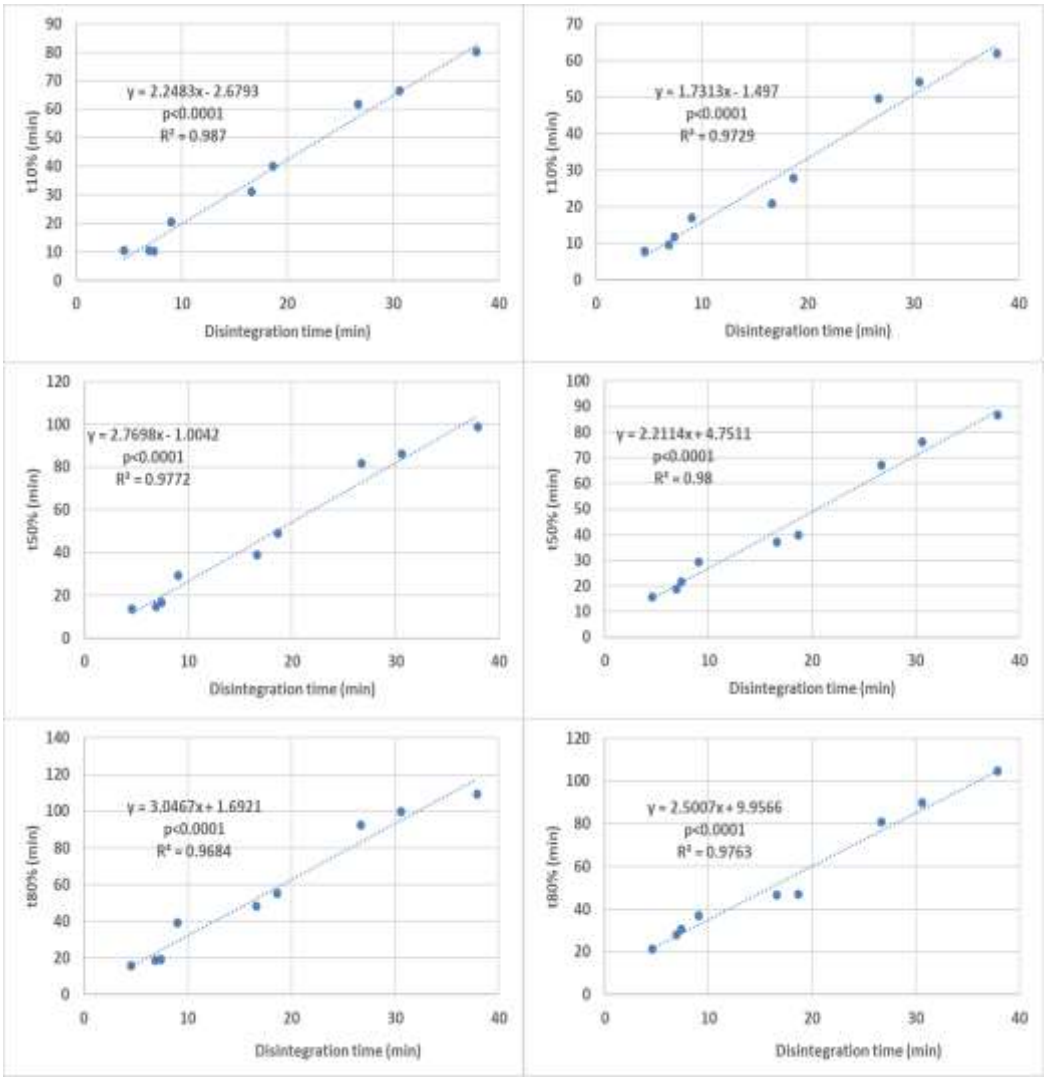


Figure 23: The overall correlation of all disintegration results with their respective dissolution results (all formulations in all media are available in every diagram) in the basket device (right side) and the paddle device (left side). The p-value is a one-sided value for a t-test applied to the slope.

Poor correlations were normally received for DRcaps® (with the notable exception of the t80% case). A feasible rationalization for that might be that the weakened capsule shell structure made the preliminary rupture more associated with random mechanical events and much less with the enteric-polymer dissolution promoting capabilities of the buffer. The complete shell dissolution/disintegration was much less confounded by such random effects resulting in better correlations for t80%. As for the weak correlations received for the t10% and t80% parameters for the HP-50-coated capsules, they seem to be caused by the close disintegration times within the blank FaSSIF and Al-Gousous et al. media. This is most likely associated with the unique discriminative abilities of the disintegration tester vs. paddle and basket apparatuses (owing to the differing hydrodynamics). HPMCAS-HG gave the finest correlations likely because of the sluggish capsule shell disintegration relative to drug dissolution. Anyway, despite multiple occurrences of weak correlations, while correlating the results of one formulation in different media, each correlation shows at the least one instance with $p < 0.05$.

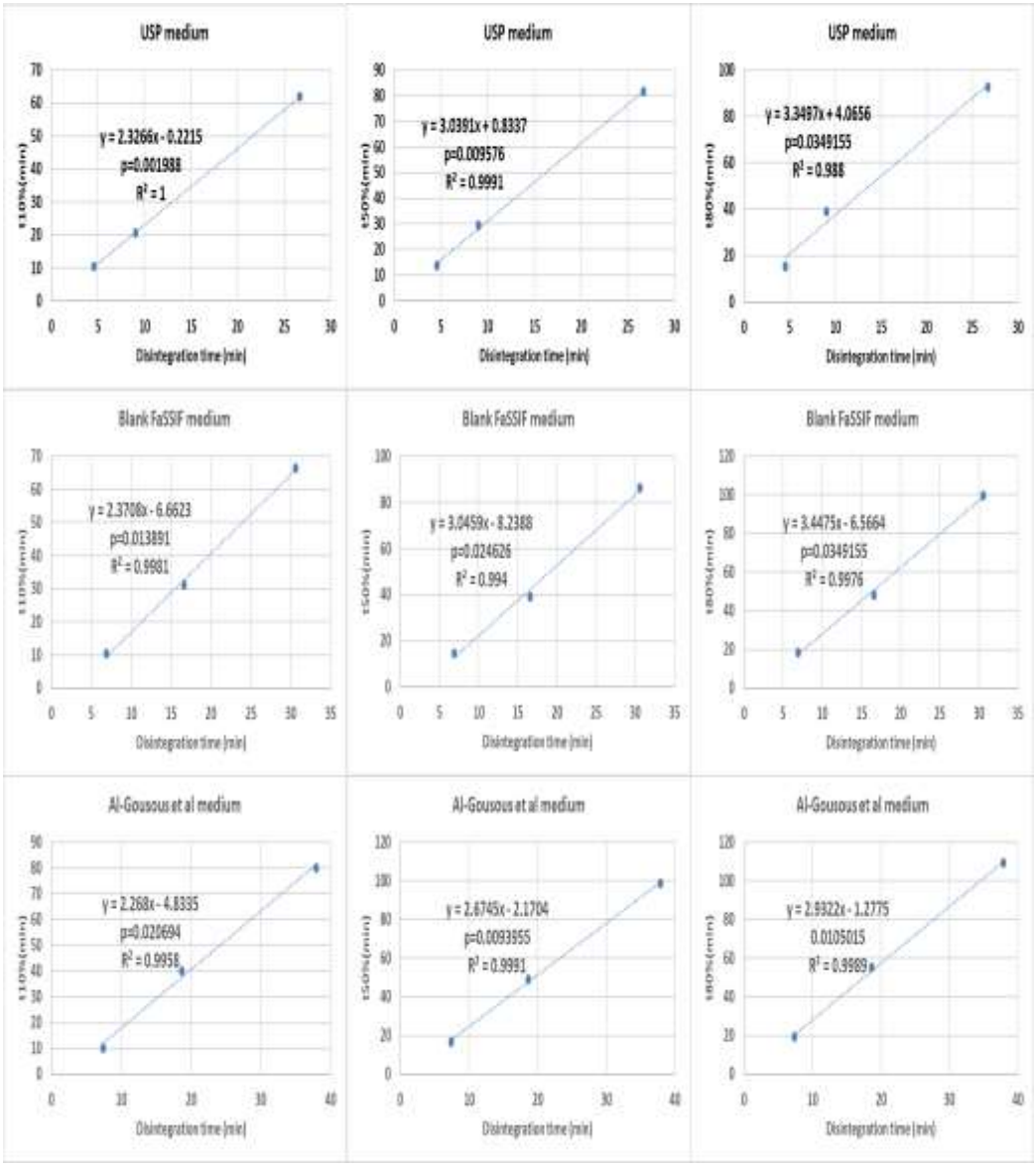


Figure 24: Correlation of disintegration time with different dissolution parameters (basket apparatus) for different formulations tested in a medium. The p-value is a one-sided value for a t-test applied to the slope.

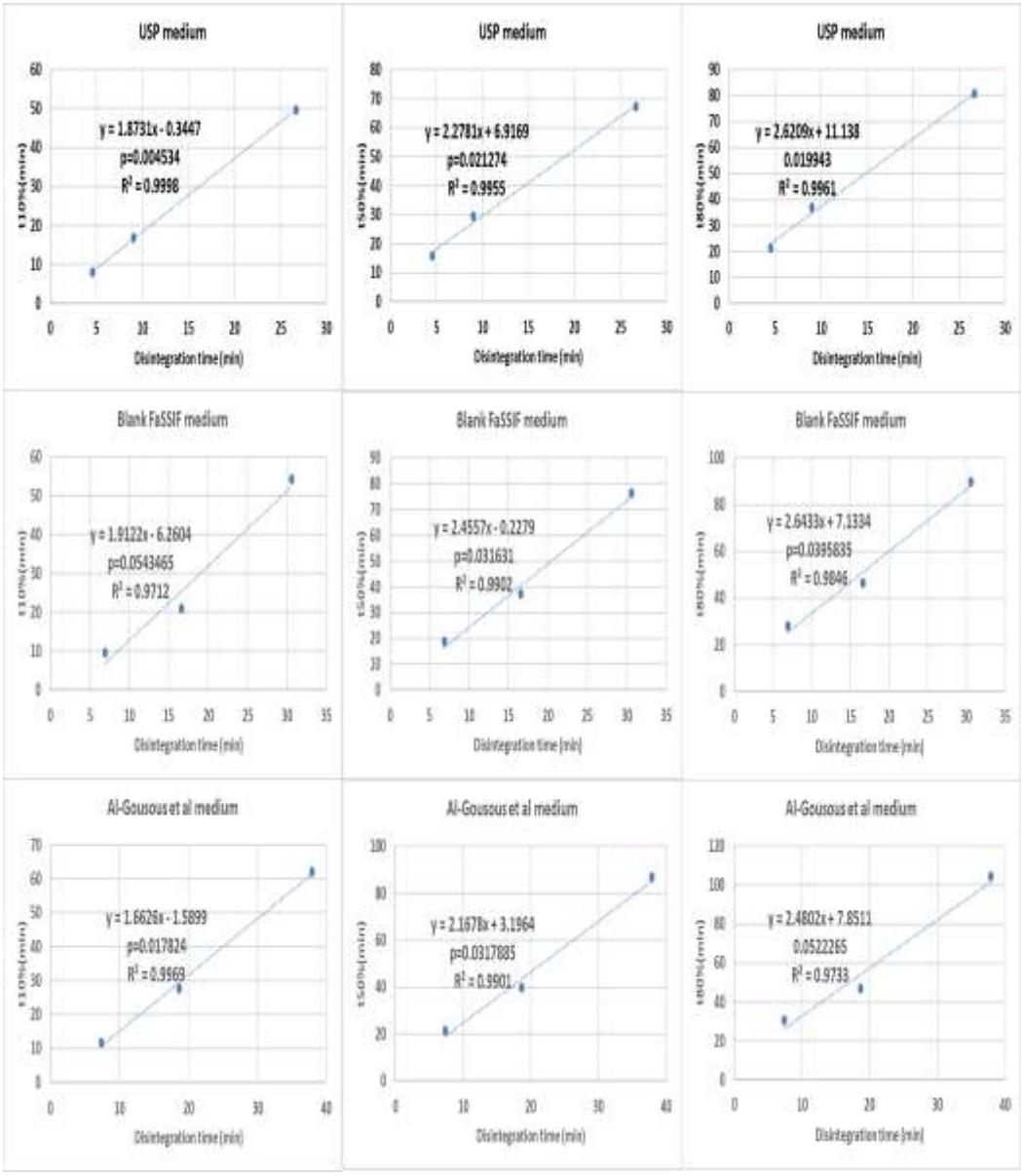


Figure 25: Correlation of disintegration time with different dissolution parameters (paddle equipment) for different formulations tested in a medium. The p-value is a one-sided value for a t-test applied to the slope.

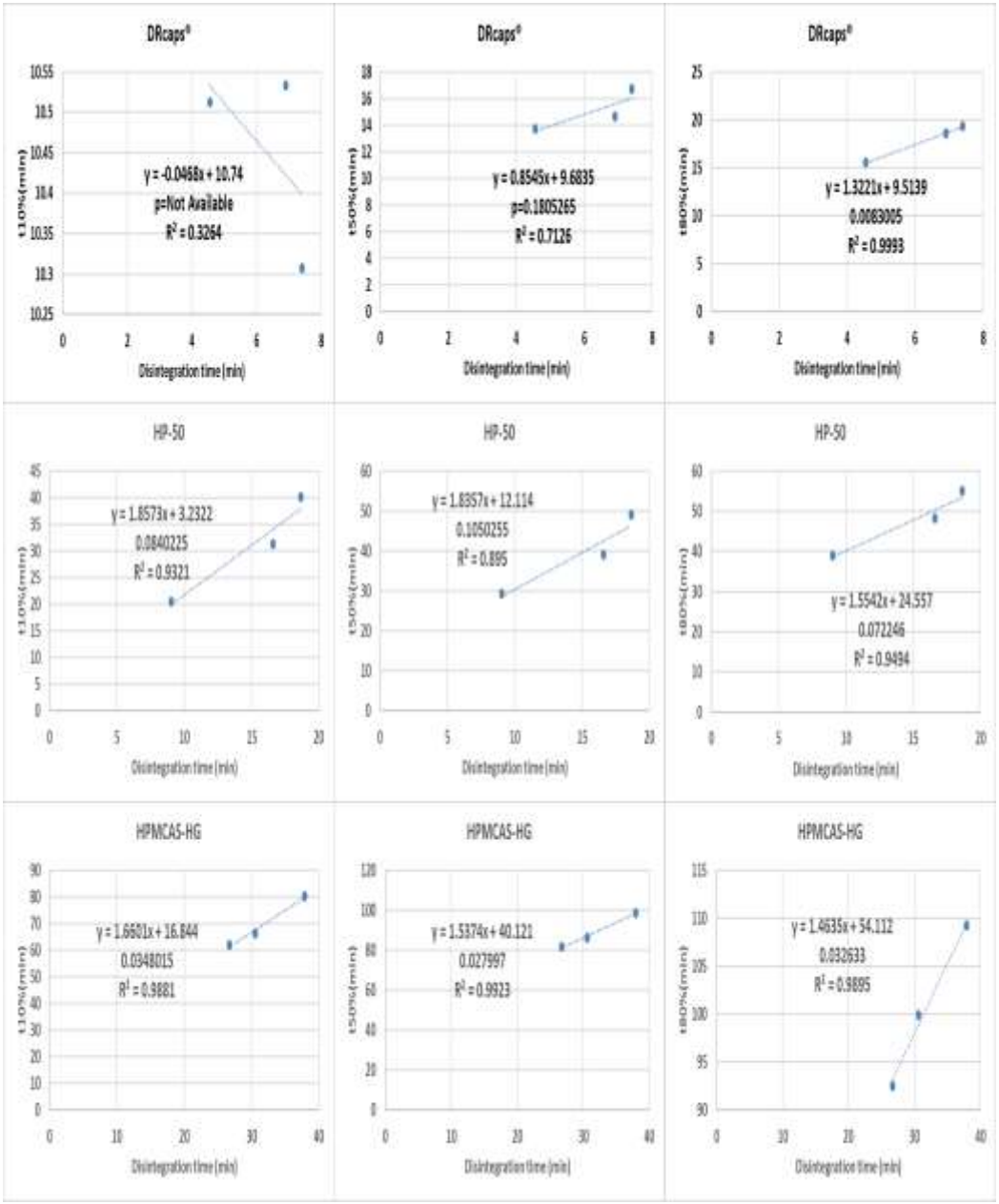


Figure 26: Correlation of disintegration time with different dissolution parameters (basket apparatus) for a formulation in different media. The p-value is a one-sided value for a t-test applied to the slope.

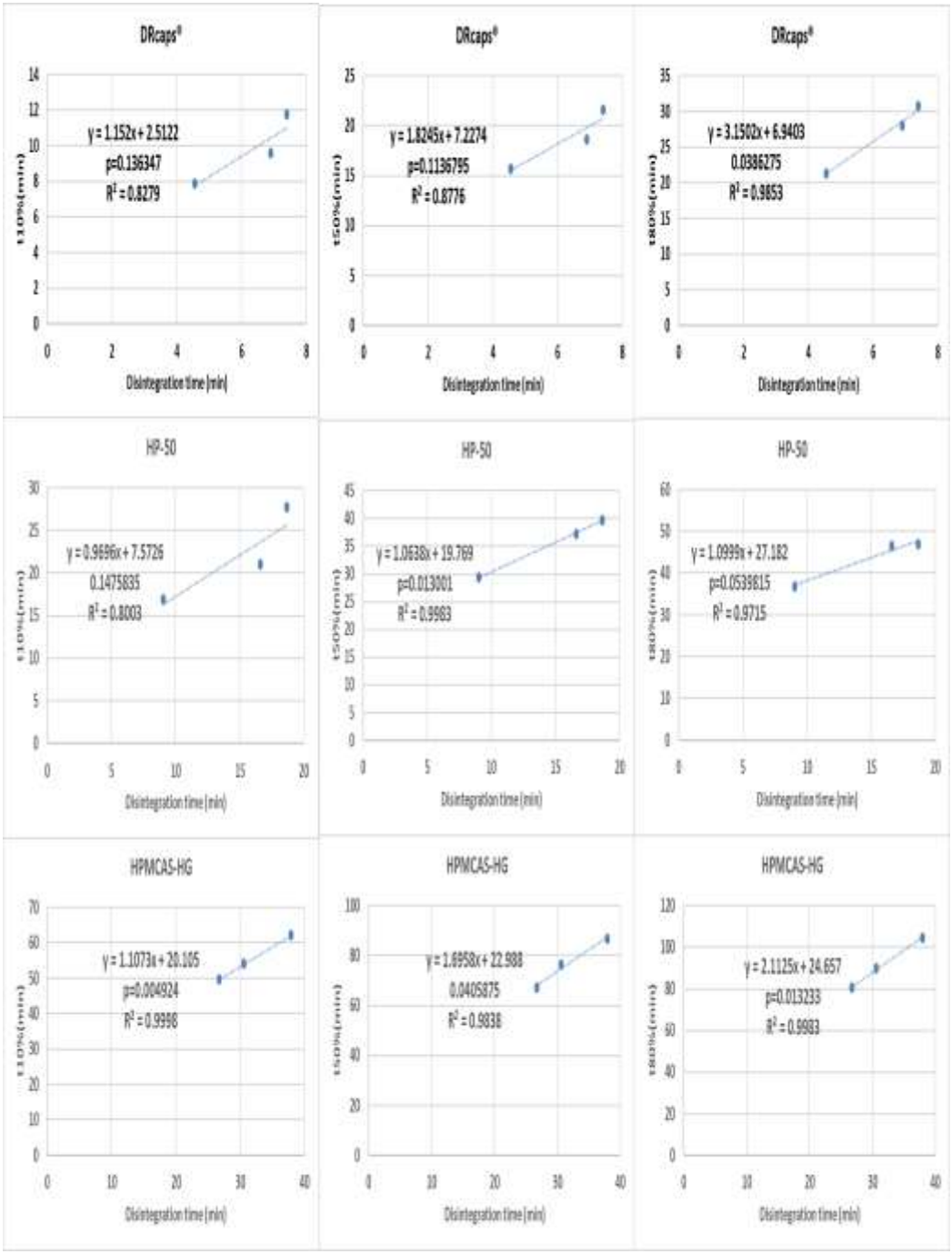


Figure 27: Correlation of the disintegration time with different dissolution parameters (paddle apparatus) for a formulation in different media. The p-value is a one-sided value for a t-test applied to the slope.

Additionally, correlations for the different dissolution time points display not only different intercepts but also unique slopes. This suggests that the disintegration times do not correlate with the different dissolution times solely because of the profiles being shifted due to distinct coat rupture times, but also because of the impact of disintegration on the general post-capsule rupture release kinetics. This is shown by using the fair to robust typical correlations obtained for the difference between t80% and t10% (corresponding to the time required for % release to rise from 10% to 80%).

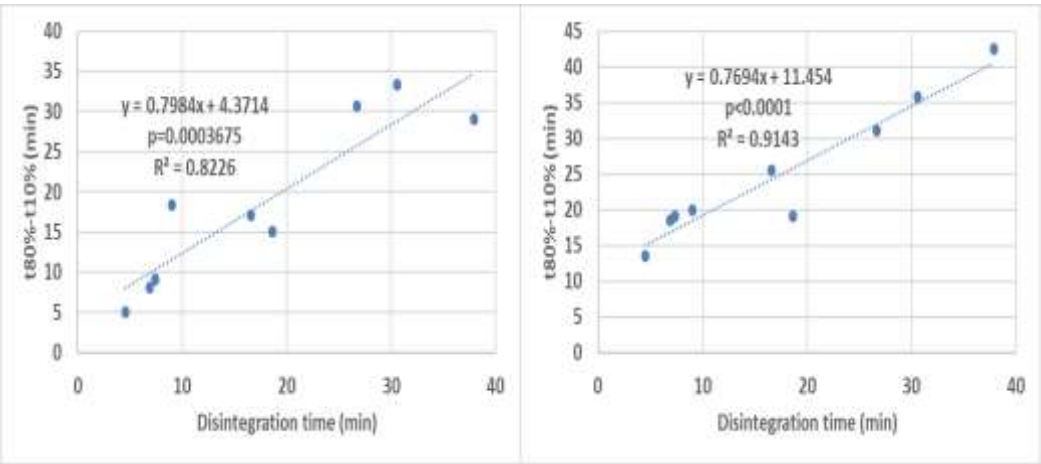


Figure 28: The total correlation of all disintegration times with their t80-t10% values Results (all formulations in all media are available in every diagram) in the basket apparatus (left side) and the paddle apparatus (right side). The p-value is a one-sided value for a t-test applied to the slope.

Figure 28 indicates stronger correlation for the paddle apparatus. A possible reason might be that variation in the floating capsule orientation within the basket, collectively with the extra variable fluid velocities inside the upper place of the basket [90], leads to the greater statistical scatter found for the basket apparatus. All in all, the acquired set of correlations indicates that enteric-coated formulations seem promising in regard to the use of disintegration tests as dissolution surrogates. This suggests that the usage of disintegration tests as dissolution tests surrogate may not only be restricted to immediate release dosage forms. However, in addition investigations on further EC dosage forms need to be performed earlier than creating a definitive judgment in this matter.

In end-effect, these results justify the use of disintegration as a dissolution surrogate in the previous sections of this work.

5. Summary

An investigation was performed on the utility of various enteric polymers for use in coating hard shell capsules. This type of preparations is of particular importance during early clinical trials, as it reduces the burden associated with formulation development for API's in need of enteric coating. A wide range of different polymers was investigated, and it was found that those polymers lead to differences in the performance parameters.

HP-55S was found (even without adding plasticizers) to provide by far the best coating in terms of closing the gap between capsule cap and body as well as providing a smooth surface as shown by scanning electron microscopy images. This was reflected by the clearly superior performance of this polymer in terms of the acid resistance of coated capsules in both 0.1 and 0.01 M HCl. Addition of talc was generally beneficial in terms of coat quality because of its bulking up effect on the coat, which helps in closing the gap.

Delayed release capsule shells showed poor performance in terms of resisting acid uptake into the capsule even though they could prevent the contents from being released in the medium. This indicates that while possibly useful for preventing gastric release of irritating substances, they are not dependable for the protection of acid-labile compounds against gastric juice, and therefore incapable of obviating the need for capsule coating.

In terms of disintegration performance, HP-50 was found to provide the fastest disintegration. This might be related to the low interfacial pH the polymer needs to dissolve. TEC had limited effect on disintegration except for HPMCAS-MG were higher TEC content led to longer disintegration times, most probably owing to increased film flexibility and continuity.

A further significant observation was the biorelevant medium of Al-Gousous et al consistently giving longer disintegration times compared to the compendial and blank FaSSIF media. This, in addition to instance of TEC level effect being detected by this medium but not by the compendial one, shows that proper selection of test media is important particularly when dealing with API's that have a preferential absorption site in the proximal small intestine.

Whether disintegration results can be considered reflective of API release was also investigated. It was found that disintegration performance correlated well to early, middle and late stages of the dissolution process both in paddle and basket devices. This indicates that using disintegration testing as a quick screening tool during enteric formulation development is justified.

6. Zusammenfassung

Eine Untersuchung zur Verwendung verschiedener Magensaft-resistenter Polymere zur Beschichtung von Hartkapseln wurde durchgeführt. Diese Art von Präparaten ist in frühen klinischen Studien von besonderer Bedeutung, da sie den Entwicklungsaufwand für Wirkstoffe zu festen Formulierungen verringert, die eine magensaft-resistente Beschichtung benötigen. Eine breite Palette verschiedener Polymere wurde untersucht, und es wurde festgestellt, dass diese Polymere zu Unterschieden in den Leistungsparametern der so hergestellten Arzneiformen führen.

Es wurde beobachtet, dass HP-55S (auch ohne Zugabe von Weichmachern) bei weitem die beste Beschichtung hinsichtlich des Schließens der Lücke zwischen Kapselkappe und Boden sowie einer glatten Oberfläche liefert, wie durch Rasterelektronenmikroskop-Aufnahmen gezeigt werden konnte. Dies spiegelte sich in der deutlich überlegenen Leistung dieses Polymers hinsichtlich der Säurebeständigkeit von beschichteten Kapseln sowohl in 0,1 als auch in 0,01 M HCl wieder. Die Zugabe von Talkum zur Coatingdispersion war im Allgemeinen in Bezug auf die Qualität des Überzugs vorteilhaft, da es sich auf die Volumenvergrösserung des Films auswirkt, was beim Schließen der Stufe Vorteile bietet.

Kapselhüllen mit verzögerter Freisetzung (ohne Beschichtung) zeigten eine mangelnde Qualität hinsichtlich ihres Widerstandes gegen die Säureaufnahme in die Kapsel, obwohl sie verhindern konnten, dass der Kapselinhalt in das Medium freigesetzt wurde. Dies weist darauf hin, dass diese Kapselhüllen zwar möglicherweise zur Verhinderung der Freisetzung von reizenden Substanzen im Magen nützlich sind, sich jedoch nicht unbedingt für den Schutz säurelabiler Verbindungen gegen Magensaft eignen und daher nicht in der Lage sind, die Notwendigkeit einer Kapselbeschichtung zu vermeiden.

In Bezug auf den Kapselzerfall wurde festgestellt, dass HP-50 den schnellsten Zerfall sicherstellt. Dies könnte mit dem niedrigen Grenzflächen-pH zusammenhängen, den das Polymer zum Auflösen benötigt. TEC hatte eine begrenzte Wirkung auf den Zerfall, außer dass HPMCAS-MG mit einem höheren TEC-Gehalt längere Zerfallszeiten zur Folge hatte, höchstwahrscheinlich aufgrund der daraus resultierenden erhöhten Filmflexibilität und -kontinuität.

Eine weitere signifikante Beobachtung war, dass das biorelevante Medium von Al-Gousous et al. im Vergleich zu den von Ph.Eur. und USP vorgeschlagenen sowie blank FaSSIF-Medien durchweg längere Zerfallszeiten ergab. Dies, zusammen mit einem Fall, wo der beobachtete TEC-Effekt in diesem Medium aber nicht in Medien der Arzneibücher nachgewiesen wurde, zeigt dass die richtige Auswahl der Auflösungsmedien wichtig ist, insbesondere wenn es sich um Wirkstoffe handelt, die eine bevorzugte Absorptionsregion im proximalen Dünndarm aufweisen.

Es wurde auch untersucht, ob Zerfallsergebnisse die Messung der Wirkstofffreisetzung spiegeln können. Dabei wurde gefunden, dass die Zerfallsgeschwindigkeit gut mit frühen,

mittleren und späten Stadien des Auflösungsprozesses sowohl in Paddel- als auch in Drehkörbchen-Apparaturen korrelierte. Dies weist darauf hin, dass die Verwendung von Zerfallsprüfungen als schnelle Screening-Tools während der Entwicklung magensaftresistenter Formulierungen gerechtfertigt ist.

7. References

[1] B. Karolewicz, A review of polymers as multifunctional excipients in drug dosage form technology, Saudi Pharm. J. (2016). https://doi.org/10.1016/j.jsps.2015.02.025.

[2] M. Nitz, O.P. Taranto, Film coating of theophylline pellets in a pulsed fluid bed coater, Chem. Eng. Process. Process Intensif. (2008). https://doi.org/10.1016/j.cep.2007.10.007.

[3] T. Ogura, HPMC capsules: an alternative to gelatin, Pharm. Tech. Eur. 10 (1998).

[4] E.T. Cole, R.A. Scott, A.L. Connor, I.R. Wilding, H.U. Petereit, C. Schminke, T. Beckert, D. Cadé, Enteric coated HPMC capsules designed to achieve intestinal targeting, Int. J. Pharm. (2002). https://doi.org/10.1016/S0378-5173(01)00871-7.

[5] K. Thoma, K. Bechtold, Enteric coated hard gelatin capsules, Capsugel Tech. Bull. (1992).

[6] R. Albanez, M. Nitz, O.P. Taranto, Enteric coating process of diclofenac sodium pellets in a fluid bed coater with a wurster insert: Influence of process variables on coating performance and release profile, Adv. Powder Technol. (2013). https://doi.org/10.1016/j.apt.2012.12.003.

[7] J. Al-Gousous, M. Penning, P. Langguth, Molecular insights into shellac film coats from different aqueous shellac salt solutions and effect on disintegration of enteric-coated soft gelatin capsules, Int. J. Pharm. 484 (2015) 283–291. https://doi.org/10.1016/j.ijpharm.2014.12.060.

[8] Shin-Etsu Chemical Co. Ltd., Shin-Etsu Pharmaceutical Excipients, (n.d.).

[9] P.J. Tarcha, Polymers for Controlled Drug Delivery, CRC Press, 1990.

[10] Shin-Etsu Chemical Co. Ltd., USP Hypromellose Phthalate, n.d.

[11] R.C. Rowe, P.J. Sheskey, S.C. Owen, Cellulose acetate phthalate, in: Handb. Pharm. Excipients, Pharmaceutical Press ; American Pharmacists Association, London; Greyslake, IL; Washington, DC, 2006.

[12] M. Aulton, G. Cole, J. Hogan, Film-coating materials and their properties, in: Pharm. Coat. Technol., Taylor & Francis, 1995.

[13] K.J. Edgar, Cellulose esters in drug delivery, Cellulose. (2007). https://doi.org/10.1007/s10570-006-9087-7.

[14] S. Wu, D. Wyatt, M. Adams, Chemistry and applications of cellulosic polymers for enteric coatings of solid dosage forms, in: Aqueous Polym. Coatings Pharm. Dos. Forms, Third Ed., Third Edit, CRC Press, 2008.

[15] J. Wang, J. Hemenway, W. Chen, D. Desai, W. Early, S. Paruchuri, S.Y. Chang, H. Stamato, S. Varia, An evaluation of process parameters to improve coating efficiency of an active tablet film-coating process, Int. J. Pharm. (2012). https://doi.org/10.1016/j.ijpharm.2012.01.033.

[16] J. Al-Gousous, P. Langguth, Oral Solid Dosage Form Disintegration Testing - The Forgotten Test, J. Pharm. Sci. 104 (2015) 2664–2675. https://doi.org/10.1002/jps.24303.

[17] ICH, INTERNATIONAL CONFERENCE ON HARMONISATION, (1999).

[18] A. Gupta, R.L. Hunt, R.B. Shah, V.A. Sayeed, M.A. Khan, Disintegration of highly soluble immediate release tablets: A surrogate for dissolution, AAPS PharmSciTech. 10 (2009) 495–499. https://doi.org/10.1208/s12249-009-9227-0.

[19] A. Radwan, G.L. Amidon, P. Langguth, Mechanistic investigation of food effect on disintegration and dissolution of BCS class III compound solid formulations: The importance of viscosity, Biopharm. Drug Dispos. (2012). https://doi.org/10.1002/bdd.1798.

[20] B. Nickerson, A. Kong, P. Gerst, S. Kao, Correlation of dissolution and disintegration results for an immediate-release tablet, J. Pharm. Biomed. Anal. (2018). https://doi.org/10.1016/j.jpba.2017.12.017.

[21] I. Locatelli, N. Nagelj Kovai, A. Mrhar, M. Bogataj, Gastric emptying of non-disintegrating solid drug delivery systems in fasted state: Relevance to drug dissolution, Expert Opin. Drug Deliv. (2010). https://doi.org/10.1517/17425247.2010.495982.

[22] F. Podczeck, N.C. Course, J.M. Newton, M.B. Short, The influence of non-disintegrating tablet dimensions and density on their gastric emptying in fasted volunteers, J. Pharm. Pharmacol. 59 (2007) 23–27. https://doi.org/10.1211/jpp.59.1.0004.

[23] F. Podczeck, N. Course, J.M. Newton, M.B. Short, Gastrointestinal transit of model mini-tablet controlled release oral dosage forms in fasted human volunteers, J. Pharm. Pharmacol. (2007). https://doi.org/10.1211/jpp.59.7.0005.

[24] A.T. Florence, E.G. Salole, Physical properties of formulations and adverse reactions to medicines, in: Formul. Factors Advers. React., Elsevier, 1990: pp. 96–120. https://doi.org/10.1016/b978-0-7236-0923-0.50010-x.

[25] I. Locatelli, A. Mrhar, M. Bogataj, Gastric emptying of pellets under fasting conditions: A mathematical model, Pharm. Res. (2009). https://doi.org/10.1007/s11095-009-9869-3.

[26] S.Y. Choe, B.L. Neudeck, L.S. Welage, G.E. Amidon, J.L. Barnett, G.L. Amidon, Novel method to assess gastric emptying in humans: The Pellet Gastric Emptying Test, Eur. J. Pharm. Sci. (2001). https://doi.org/10.1016/S0928-0987(01)00196-8.

[27] J.M. Newton, Gastric emptying of multi-particulate dosage forms, Int. J. Pharm. (2010). https://doi.org/10.1016/j.ijpharm.2010.04.047.

[28] G.M. Clarke, J.M. Newton, M.B. Short, Comparative gastrointestinal transit of pellet systems of varying density, Int. J. Pharm. (1995). https://doi.org/10.1016/0378-5173(94)00200-O.

[29] P.J. Tarcha, Polymers for Controlled Drug Delivery, Taylor & Francis, 1990. https://books.google.de/books?id=A73NI1TmS7wC.

[30] D.M. Mudie, G.L. Amidon, G.E. Amidon, Physiological parameters for oral delivery and in vitro testing, Mol. Pharm. (2010). https://doi.org/10.1021/mp100149j.

[31] A. Kambayashi, H. Blume, J. Dressman, Understanding the in vivo performance of enteric coated tablets using an in vitro-in silico-in vivo approach: Case example diclofenac, Eur. J. Pharm. Biopharm. (2013). https://doi.org/10.1016/j.ejpb.2013.09.009.

[32] M. Štefanič, I. Locatelli, F. Vrečer, T. Sever, A. Mrhar, M. Bogataj, The influence of gastric emptying kinetics on the drug release from enteric coated pellets in fasted state: An in vitro/in vivo correlation, Eur. J. Pharm. Biopharm. (2012). https://doi.org/10.1016/j.ejpb.2012.07.011.

[33] F. BENDTSEN, L. OVESEN, B. ROSENKILDE-GRAM, S.J. RUNE, Effect of omeprazole on intragastric and duodenal bulb acidity in duodenal ulcer patients, Aliment. Pharmacol. Ther. (2007). https://doi.org/10.1111/j.1365-2036.1989.tb00201.x.

[34] K. Matsui, Y. Tsume, G.E. Amidon, G.L. Amidon, In Vitro Dissolution of Fluconazole and Dipyridamole in Gastrointestinal Simulator (GIS), Predicting in Vivo Dissolution and Drug-Drug Interaction Caused by Acid-Reducing Agents, Mol. Pharm. (2015). https://doi.org/10.1021/acs.molpharmaceut.5b00135.

[35] P. Morrison, H. Rogers, I. Bradbrook, C. Parsons, Concurrent administration of cimetidine and enteric-coated prednisolone: effect on plasma levels of prednisolone., Br. J. Clin. Pharmacol. (1980). https://doi.org/10.1111/j.1365-2125.1980.tb00506.x.

[36] S. Kofler, C. Wolf, N. Shvets, Z. Sisic, T. Mller, J. Behr, H.Y. Sohn, M. Vogeser, M. Shipkova, B. Meiser, G. Steinbeck, B. Reichart, I. Kaczmarek, The proton pump inhibitor pantoprazole and its interaction with enteric-coated mycophenolate sodium in transplant recipients, J. Hear. Lung Transplant. (2011). https://doi.org/10.1016/j.healun.2010.12.003.

[37] C.E. Staatz, S.E. Tett, Clinical pharmacokinetics and pharmacodynamics of mycophenolate in solid organ transplant recipients, Clin. Pharmacokinet. (2007). https://doi.org/10.2165/00003088-200746010-00002.

[38] M. Kamba, Y. Seta, N. Takeda, T. Hamaura, A. Kusai, H. Nakane, K. Nishimura, Measurement of agitation force in dissolution test and mechanical destructive force in disintegration test, Int. J. Pharm. (2003). https://doi.org/10.1016/S0378-5173(02)00535-5.

[39] M. Gibaldi, Introduction to Biopharmaceutics, Lea & Febiger, 1994. https://doi.org/10.1016/B978-0-444-81871-3.50006-0.

[40] J.G. Wagner, P.K. Wilkinson, A.J. Sedman, R.G. Stoll, Failure of usp tablet disintegration test to predict performance in man, J. Pharm. Sci. (1973). https://doi.org/10.1002/jps.2600620548.

[41] O.N. Fernando, J. Moorhead, H.F. West, Absorption of enteric-coated prednisolone, Br. Med. J. (1979). https://doi.org/10.1136/bmj.1.6180.1795-c.

[42] L. Rudinskas, T.W. Paton, S.E. Walker, D.A. Dotten, D.H. Cowan, Poor clinical response to enteric-coated iron preparations, CMAJ. (1989).

[43] I.R. Wilding, J.G. Hardy, R.A. Sparrow, S.S. Davis, P.B. Daly, J.R. English, In Vivo Evaluation of Enteric-Coated Naproxen Tablets Using Gamma Scintigraphy, Pharm. Res. An Off. J. Am. Assoc. Pharm. Sci. (1992). https://doi.org/10.1023/A:1015858829187.

[44] J.P. Ebel, M. Jay, R.M. Beihn, An in Vitro/in Vivo Correlation for the Disintegration and Onset of Drug Release from Enteric-Coated Pellets, Pharm. Res. An Off. J. Am. Assoc. Pharm. Sci. 10 (1993) 233–238. https://doi.org/10.1023/A:1018986827350.

[45] J. Al-Gousous, G.L. Amidon, P. Langguth, Toward Biopredictive Dissolution for Enteric Coated Dosage Forms, Mol. Pharm. 13 (2016) 1927–1936. https://doi.org/10.1021/acs.molpharmaceut.6b00077.

[46] EUDRAGIT® functional polymers – Evonik Health Care, (n.d.). https://healthcare.evonik.com/product/health-care/en/products/pharmaceutical-excipients/EUDRAGIT/ (accessed May 9, 2020).

[47] J. Al-Gousous, P. Langguth, European versus United States Pharmacopeia disintegration testing methods for enteric-coated soft gelatin capsules, Dissolution Technol. (2015). https://doi.org/10.14227/DT220315P6.

[48] H.A. Merchant, A. Goyanes, N. Parashar, A.W. Basit, Predicting the gastrointestinal behaviour of modified-release products: Utility of a novel dynamic dissolution test apparatus involving the use of bicarbonate buffers, Int. J. Pharm. (2014). https://doi.org/10.1016/j.ijpharm.2014.09.003.

[49] G. Garbacz, B. Kołodziej, M. Koziolek, W. Weitschies, S. Klein, A dynamic system for the simulation of fasting luminal pH-gradients using hydrogen carbonate buffers for dissolution testing of ionisable compounds, Eur. J. Pharm. Sci. (2014). https://doi.org/10.1016/j.ejps.2013.09.020.

[50] J.J. Sheng, D.P. McNamara, G.L. Amidon, Toward an In Vivo dissolution methodology: A comparison of phosphate and bicarbonate buffers, Mol. Pharm. (2009). https://doi.org/10.1021/mp800148u.

[51] M. Štefanič, F. Vrečer, P. Rizmal, A. Mrhar, M. Bogataj, Prediction of the in vivo performance of enteric coated pellets in the fasted state under selected biorelevant dissolution conditions, Eur. J. Pharm. Sci. (2014). https://doi.org/10.1016/j.ejps.2014.05.007.

[52] C. Schiller, C.P. Fröhlich, T. Giessmann, W. Siegmund, H. Mönnikes, N. Hosten, W. Weitschies, Intestinal fluid volumes and transit of dosage forms as assessed by magnetic resonance imaging, Aliment. Pharmacol. Ther. (2005). https://doi.org/10.1111/j.1365-2036.2005.02683.x.

[53] D.M. Mudie, K. Murray, C.L. Hoad, S.E. Pritchard, M.C. Garnett, G.L. Amidon, P.A. Gowland, R.C. Spiller, G.E. Amidon, L. Marciani, Quantification of gastrointestinal liquid volumes and distribution following a 240 mL dose of water in the fasted state, Mol. Pharm. (2014). https://doi.org/10.1021/mp500210c.

[54] F. Liu, R. Lizio, C. Meier, H.U. Petereit, P. Blakey, A.W. Basit, A novel concept in enteric coating: A double-coating system providing rapid drug release in the proximal

small intestine, J. Control. Release. (2009). https://doi.org/10.1016/j.jconrel.2008.09.083.

[55] F. Liu, A.W. Basit, A paradigm shift in enteric coating: Achieving rapid release in the proximal small intestine of man, J. Control. Release. (2010). https://doi.org/10.1016/j.jconrel.2010.07.105.

[56] K. Nollenberger, J. Albers, Poly(meth)acrylate-based coatings, Int. J. Pharm. 457 (2013) 461–469. https://doi.org/10.1016/j.ijpharm.2013.09.029.

[57] S.K. Dutta, R.M. Russell, F.L. Iber, Influence of exocrine pancreatic insufficiency on the intraluminal pH of the proximal small intestine, Dig. Dis. Sci. (1979). https://doi.org/10.1007/BF01489321.

[58] P. Langguth, G. Breves, A. Stökli, H.P. Merkle, S. Wolffram, Colonic Absorption and Bioavailability of the Pentapeptide Metkephamid in the Rat, Pharm. Res. An Off. J. Am. Assoc. Pharm. Sci. 11 (1994) 1640–1645. https://doi.org/10.1023/A:1018974107844.

[59] Dissolution Methods, (n.d.). https://www.accessdata.fda.gov/scripts/cder/dissolution/index.cfm (accessed May 9, 2020).

[60] ON THE MEASUREMENT OF BUFFER VALUES AND ON THE RELATIONSHIP OF BUFFER VALUE TO THE DISSOCIATION CONSTANT OF THE BUFFER AND THE CONCENTRATION AND REACTION OF THE BUFFER SOLUTION, J. Biol. Chem. (1922).

[61] G. Garbacz, G.M. Rappen, M. Koziolek, W. Weitschies, Dissolution of mesalazine modified release tablets under standard and bio-relevant test conditions, J. Pharm. Pharmacol. (2015). https://doi.org/10.1111/jphp.12332.

[62] F.J.O. Varum, G.B. Hatton, A.C. Freire, A.W. Basit, A novel coating concept for ileo-colonic drug targeting: Proof of concept in humans using scintigraphy, Eur. J. Pharm. Biopharm. (2013). https://doi.org/10.1016/j.ejpb.2013.01.002.

[63] CENTER FOR DRUG EVALUATION AND RESEARCH APPLICATION NUMBER: 204412Orig1s000 CLINICAL PHARMACOLOGY AND BIOPHARMACEUTICS REVIEW(S), n.d.

[64] fda, cder, label, n.d. www.fda.gov/medwatch (accessed May 9, 2020).

[65] A. Goyanes, G.B. Hatton, H.A. Merchant, A.W. Basit, Gastrointestinal release behaviour of modified-release drug products: Dynamic dissolution testing of mesalazine formulations, Int. J. Pharm. (2015). https://doi.org/10.1016/j.ijpharm.2015.02.051.

[66] S.G. Nugent, D. Kumar, D.S. Rampton, D.F. Evans, Intestinal luminal pH in inflammatory bowel disease: Possible determinants and implications for therapy with aminosalicylates and other drugs, Gut. (2001). https://doi.org/10.1136/gut.48.4.571.

[67] N. Washington, C. Washington, C.G. Wilson, C.G. Wilson, Physiological pharmaceutics : barriers to drug absorption, Taylor & Francis, 2001.

[68] M. Koziolek, F. Schneider, M. Grimm, C. Mode, A. Seekamp, T. Roustom, W. Siegmund, W. Weitschies, Intragastric pH and pressure profiles after intake of the high-caloric, high-fat meal as used for food effect studies, J. Control. Release. (2015). https://doi.org/10.1016/j.jconrel.2015.10.022.

[69] A.J. Coupe, S.S. Davis, D.F. Evans, I.R. Wilding, Do pellet formulations empty from the stomach with food?, Int. J. Pharm. (1993). https://doi.org/10.1016/0378-5173(93)90276-L.

[70] F. Schneider, M. Grimm, M. Koziolek, C. Modeß, A. Dokter, T. Roustom, W. Siegmund, W. Weitschies, Resolving the physiological conditions in bioavailability and bioequivalence studies: Comparison of fasted and fed state, Eur. J. Pharm. Biopharm. (2016). https://doi.org/10.1016/j.ejpb.2016.09.009.

[71] S.S. Davis, J.G. Hardy, J.W. Fara, Transit of pharmaceutical dosage forms through the small intestine, Gut. (1986). https://doi.org/10.1136/gut.27.8.886.

[72] G. Digenis, In Vivo Behavior of Multiparticulate Versus Single-Unit Dose Formulations, in: 1994: pp. 333–355. https://doi.org/10.1201/b14855-14.

[73] E. Jantratid, N. Janssen, C. Reppas, J.B. Dressman, Dissolution media simulating conditions in the proximal human gastrointestinal tract: An update, Pharm. Res. (2008). https://doi.org/10.1007/s11095-008-9569-4.

[74] J. Mössner, V. Keim, Therapie mit pankreasenzymen, Dtsch. Arztebl. 108 (2011) 578–582. https://doi.org/10.3238/arztebl.2011.0578.

[75] Guidance, Compliance, & Regulatory Information | FDA, (n.d.). https://www.fda.gov/drugs/guidance-compliance-regulatory-information (accessed May 10, 2020).

[76] J. Rubbens, J. Brouwers, K. Wolfs, E. Adams, J. Tack, P. Augustijns, Ethanol concentrations in the human gastrointestinal tract after intake of alcoholic beverages, Eur. J. Pharm. Sci. (2016). https://doi.org/10.1016/j.ejps.2016.02.009.

[77] J. Al-Gousous, P. Langguth, Effect of Calcium Ions on the Disintegration of Enteric-Coated Solid Dosage Forms, J. Pharm. Sci. (2016). https://doi.org/10.1002/jps.24700.

[78] Shin-Etsu Chemical Co. Ltd., Shin-Etsu AQOAT® - For Pharmaceutical : Shin-Etsu Cellulose, (n.d.).

[79] Shin-Etsu Chemical Co. Ltd., HPMCP - For Pharmaceutical : Shin-Etsu Cellulose, (n.d.).

[80] M.M. Al-Tabakha, A.I. Arida, K.M.S. Fahelelbom, B. Sadek, R.A.A. Jarad, Performances of new generation of delayed release capsules, J. Young Pharm. (2015). https://doi.org/10.5530/jyp.2015.1.7.

[81] Vassarstats, vassarstats website, (n.d.). http://vassarstats.net/.

[82] Norman E. Dowling, Mechanical Behavior of Materials, 2013.

[83] S. Missaghi, C. Young, K. Fegely, A.R. Rajabi-Siahboomi, Delayed release film coating applications on oral solid dosage forms of proton pump inhibitors: Case studies

Delayed release solid dosage forms of proton pump inhibitors, Drug Dev. Ind. Pharm. (2010). https://doi.org/10.3109/03639040903468811.

[84] J. Al-Gousous, Y. Tsume, M. Fu, I.I. Salem, P. Langguth, Unpredictable Performance of pH-Dependent Coatings Accentuates the Need for Improved Predictive in Vitro Test Systems, Mol. Pharm. 14 (2017) 4209–4219. https://doi.org/10.1021/acs.molpharmaceut.6b00877.

[85] L.A. Felton, J.W. McGinity, Aqueous Polymeric Coatings for Pharmaceutical Dosage Forms, Third Edition, Third Edit, CRC Press, 2008.

[86] Ashland Chemical Co. Ltd., AquaSolve HPMCAS Physical Chemical Properties Handbook, (n.d.). https://www.ashland.com/file_source/Ashland/Industries/Pharmaceutical/Links/PC-12624.6_AquaSolve_HPMCAS_Physical_Chemical_Properties.pdf (accessed May 10, 2020).

[87] A.T. Florence, D. Attwood, Physicochemical principles of pharmacy: In manufacture, formulation and clinical use, Pharmaceutical press, 2015.

[88] FDA, SCOGS (Select Committee on GRAS Substances), SCOGS Database. (n.d.).

[89] Evonik, EUDRAGIT® Application Guidelines, n.d.

[90] D.M. D'Arcy, O.I. Corrigan, A.M. Healy, Evaluation of hydrodynamics in the basket dissolution apparatus using computational fluid dynamics - Dissolution rate implications, Eur. J. Pharm. Sci. (2006). https://doi.org/10.1016/j.ejps.2005.10.007.

8. List of Figures

9. List of Tables

10. Publication list

Journal articles

Maoqi Fu, Jozef Al-Gousous, Johannes Blechar, Peter Langguth. Enteric hard capsules for targeting the small intestine: Positive correlation between disintegration and dissolution times. [J] Pharmaceutics, 2020, 12(2), 123. (IF:4.77)

Maoqi Fu, Johannes Blechar, Andreas Sauer, Jozef Al-Gousous, Peter Langguth. Comparison of organic enteric coatings for HPMC capsules. [J] Pharmaceutics. (Submitted) (IF:4.77)

Al Gousous J, Tsume Y, **Fu M**, Salem II, Langguth P. Unpredictable performance of pH dependent coatings accentuates the need for improved predictive in vitro test systems. [J] Molecular Pharmaceutics, 2017, 14(12):42094219 (IF:4.396)

Poster presentation:

Fu,M.Q.; Blechar,J.A.; Al-Gousous,J.; Sauer,A.; Lesser,I.; Eggers,T., Langguth,P. Relationship between Disintegration and Dissolution of Enteric-Coated Caffeine Capsules. Annual Meeting of the German Pharmaceutical Socienty in Heidelberg (2019)

11. Curriculum Vitae