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Research paper

Evolution of a physiological pH 6.8 bicarbonate buffer system: Application to the dissolution testing of enteric coated products

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ABSTRACT

The use of compendial pH 6.8 phosphate buffer to assess dissolution of enteric coated products gives rise to poor *in vitro–in vivo* correlations because of the inadequacy of the buffer to resemble small intestinal fluids. A more representative and physiological medium, pH 6.8 bicarbonate buffer, was developed to evaluate the dissolution behaviour of enteric coatings. The bicarbonate system was evolved from pH 7.4 Hanks balanced salt solution to produce a pH 6.8 bicarbonate buffer (modified Hanks buffer, *m*Hanks), which resembles the ionic composition and buffer capacity of intestinal milieu. Prednisolone tablets were coated with a range of enteric polymers: hypromellose phthalate (HP-50 and HP-55), cellulose acetate phthalate (CAP), hypromellose acetate succinate (HPMCAS-LF and HPMCAS-MF), methacrylic acid copolymers (EUDRAGIT® L100-55, EUDRAGIT® L30D-55 and EUDRAGIT® L100) and polyvinyl acetate phthalate (PVAP). Dissolution of coated tablets was carried out using USP-II apparatus in 0.1 M HCl for 2 h followed by pH 6.8 phosphate buffer or pH 6.8 *m*Hanks bicarbonate buffer. In pH 6.8 phosphate buffer, the various enteric polymer coated products displayed rapid and comparable dissolution profiles. In pH 6.8 *m*Hanks buffer, drug release was delayed and marked differences were observed between the various coated tablets, which is comparable to the delayed disintegration times reported in the literature for enteric coated products in the human small intestine. In summary, the use of pH 6.8 physiological bicarbonate buffer (*m*Hanks) provides more realistic and discriminative *in vitro* release assessment of enteric coated formulations compared to compendial phosphate buffer.

1. Introduction

The application of an enteric coating to a solid dosage form is an established approach to prevent drug release in the stomach and allow release in the small intestine. It is used to preclude the degradation of acid-labile actives in the gastric environment or to protect the stomach from irritant compounds [1]. The commonly used enteric coatings employ pH-dependent polymers which contain carboxylic acid groups. These remain un-ionized in the low pH environment of the stomach and become ionized in the higher pH conditions of the small intestine, thus initiating dissolution of the coating and allowing drug release.

The *in vitro* dissolution of enteric coated products is usually assessed in compendial pH 6.8 phosphate buffer. In this medium, drug release is normally rapid [2–5]. However, neither does this reflect the *in vivo* performance of enteric coated products, nor is it sufficient to discriminate the dissolution behaviour between different enteric coatings. *In vivo* gamma scintigraphy studies have shown that there is a substantial time delay (up to 2 h) for such products to disintegrate in the human small intestine post gastric emptying, with different enteric polymer coatings exhibiting varying disintegration times [6–11]. This *in vitro–in vivo* discrepancy is not surprising considering the inadequacy of the *in vitro* test method to simulate the gastrointestinal tract in many respects such as ionic composition, buffer capacity, pH, viscosity, fluid volume, coupled with further luminal factors including motility and hydrodynamics [12–17]. Moreover, the fact that these factors are subject to considerable inter- and intra-subject variability further adds to the complexity of developing an *in vitro* model to predict the gastrointestinal behaviour of complex dosage forms such as enteric coated products [16].

The constituent buffer salts, ionic strength and buffer capacity of the dissolution media have been reported to influence drug release from pH-responsive polymer coated dosage forms [16,18–20]. Notably, the luminal fluids of the small intestine are predominantly buffered by bicarbonate (as well as other ions and luminal constituents such as bile salts, proteins, carbohydrates and food components). Hence, bicarbonate buffers would more closely resemble the environment within the intestine and provide

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a more physiological medium for the *in vitro* assessment of products designed to release in the small bowel.

We have previously shown that a pH 7.4 bicarbonate system (Krebs buffer), which simulates the luminal environment of the distal small intestine, provided better *in vitro–in vivo* correlations for a series of enteric coated products for delivery of mesalazine to the ileo-colonic region of the gastrointestinal tract [21]. The normal pH of bicarbonate buffers (Krebs or Hanks) is 7.4; however, this pH is higher than the pH in the proximal small intestine [22]. The objective of this study was to develop a pH 6.8 bicarbonate system, based on Hanks buffer. This physiological medium was then employed to evaluate the dissolution behaviour of tablets coated with a series of enteric polymers from different chemical classes in comparison with compendial pH 6.8 phosphate buffer.

2. Materials and methods

2.1. Materials

The enteric polymers used in this study and their properties are presented in Table 1. Prednisolone was purchased from Aventis Pharma., Antony, France. Lactose (Pharmatose) was obtained from Ellis & Everard, Essex, UK. Cross-linked sodium carboxymethylcellulose was donated by FMC International, Cork, Ireland. Polyvinylpyrrolidone 40,000 was purchased from VWR International Ltd., Poole, UK. Magnesium stearate was purchased from Sigma–Aldrich Co. Ltd., Dorset, UK. Triethyl citrate was obtained from Lancaster Synthesis, Lancashire, UK. Sodium lauryl sulphate and triacetin were sourced from Sigma–Aldrich Co. Ltd., Dorset, UK. Talc (fine powder) was purchased from VWR International Ltd., Poole, UK. Organic solvents used were of analytical grade and were obtained from VWR International Ltd, Poole, UK (ethanol) and Fisher Scientific UK Ltd., Loughborough, UK (acetone and isopropanol). Salts for preparing buffer solutions were obtained from VWR International Ltd., Poole, UK.

2.2. Preparation of prednisolone tablets

Tablets were prepared containing 5% prednisolone, 88.5% lactose, 5% polyvinylpyrrolidone, 0.5% cross-linked sodium carboxymethylcellulose and 1% magnesium stearate. Tablets were prepared by wet granulation and were produced using a single punch tableting machine (Manesty F3, Liverpool, UK). Cross-linked sodium carboxymethylcellulose (disintegrant) was added both intra- and extra-granularly (50:50). A biconcave 8-mm punch and die set (Holland Ltd., Nottingham, UK) was used to obtain tablets of mass 200 mg (containing 10 mg drug) and crushing strength of 80 N.

2.3. Coating of prednisolone tablets

Enteric coating formulations were prepared from either aqueous polymer dispersions or organic solutions. The compositions of the aqueous and organic coating formulations are presented in Table 2.

Prednisolone tablets were coated using a Strea-1 bottom spray fluidised bed coater (Aeromatic AG, Bubendorf, Switzerland). The coating conditions were optimised for each polymer formulation and are summarized in Table 3, together with the coating levels of the polymers. Coating levels were determined by the amount of polymer applied per square centimetre of tablet surface (mg/cm^2), except for PVAP where percentage tablet weight gain (TWG %) was used. This is because the quantitative composition of the PVAP formulation is not in the public domain. After the coating process, the tablets were cured in an air-assisted oven at 40 °C for 2 h.

2.4. Dissolution of enteric coated tablets

2.4.1. Acid uptake

All enteric coating formulations at each coating level were evaluated for acid resistance and uptake. Six coated tablets of each formulation were weighed and subjected to dissolution conditions in 0.1 M HCl. After 2 h, the tablets were removed and excess medium was drained and blotted with filter paper from around the tablets. The tablets were weighed again, and the acid uptake by the tablet was calculated according to Eq. (1). Formulations were chosen for dissolution testing at the minimum coating level that met the criteria for acid protection, i.e., no more than 10% acid uptake and no visible signs of coat disruption after two hours acid treatment.

$$\text{Acid uptake } (\%) = \left(\frac{W_f - W_i}{W_i}\right) \times 100 \quad (1)$$

where W_f is the final tablet weight, W_i is the initial tablet weight.

2.4.2. Development of physiological bicarbonate buffer (mHanks)

Hanks balanced salt solution closely resembles the ionic composition of small intestinal milieu (Table 4); however, it has a pH of 7.4, which is too high, and a buffer capacity of 1 mmol/L/ΔpH, which is too low, compared to human jejunal fluids. Therefore, this buffer was modified to attain a pH of 6.8 and a higher buffer capacity. Hanks solution is primarily a bicarbonate buffer, in which bicarbonate (HCO_3^-) and carbonic acid (H_2CO_3) coexist, along with $CO_{2(aq)}$ resultant from the dissociation of the latter (Eq. (2)).

$$\begin{array}{c} CO_{2\,(g)} \\ \Big\downarrow\Big\uparrow \\ H_2O + CO_{2\,(aq)} \rightleftharpoons H_2CO_3 \rightleftharpoons H^+ + HCO_3^- \end{array} \quad (2)$$

The pH of the buffer system can be altered by adjusting the concentration of the acid (H_2CO_3) and its conjugate base (HCO_3^-) according to the Henderson–Hasselbalch equation (Eq. (3)). Purging

Table 1
Enteric polymers used in the study.

Polymer	Brand name	Abbreviation	Grade	Soluble at or above pH	Manufacturer/supplier
Methacrylic acid copolymer	EUDRAGIT®	–	L 100-55 L 30D-55 L100	5.5 5.5 6.0	Evonik GmbH, Darmstadt, Germany
Hypromellose acetate succinate	Aqoat®	HPMCAS	LF MF	5.0 6.0	Shin-Etsu Chemical Co., Ltd., Japan
Hypromellose phthalate	–	HPMCP	HP-50, HP-55	5.0, 5.5	Shin-Etsu Chemical Co., Ltd., Japan
Cellulose acetate phthalate	–	CAP	–	6.0	Eastman Chemical Company, USA
Polyvinyl acetate phthalate	Opadry® Enteric Sureteric®	PVAP	Organic Aqueous	5.0 5.0	Colorcon Ltd., USA.

Table 2
Composition of the coating formulations.

[A] Aqueous formulations	EUDRAGIT® L30 D-55	PVAP (aqueous)[a]	HPMCAS-LF	HPMCAS-MF
Polymer weight	20 g	20 g	20 g	20 g
Talc	10 g (50%[c])	–	6 g (30%[c])	6 g (30%[c])
Triethyl citrate	2 g (10%)[c]	–	4 g (20%[c])	4 g (20%[c])
Sodium lauryl sulphate	–	–	0.6 g (3%[c])	0.6 g (3%[c])
Water	128 g	113.3 g	352 g	352 g
Solid content of the spray suspension	20%	15%	8%	8%

[B] Organic formulations	EUDRAGIT® L100-55	EUDRAGIT® L100	PVAP (organic)[b]	CAP	HP-50	HP-55
Polymer weight	20 g	20 g	20 g	20 g	20 g	20 g
Talc	10 g (50%[c])	10 g (50%[c])	–	10 g (50%[c])	10 g (50%[c])	10 g (50%[c])
Triethyl citrate	2 g (10%[c])	2 g (10%[c])	–	–	2 g (10%[c])	2 g (10%[c])
Triacetin	–	–	–	5 g (25%[c])	–	–
Isopropanol	279.4 g (97%[d])	279.4 g (97%[d])	144 g (80%[d])	–	–	–
Ethanol	–	–	–	–	230.4 g (80%[d])	230.4 g (80%[d])
Acetone	–	–	–	249 g (97%[d])	–	–
Water	8.6 g (3%[d])	8.6 g (3%[d])	36 g (20%[d])	7.7 g (3%[d])	57.6 g (20%[d])	57.6 g (20%[d])
Solid content of the spray suspension	10%	10%	10%	12%	10%	10%

[a] Fully formulated coating system containing: PVAP, talc, polyethylene glycol 3350, sodium bicarbonate, triethyl citrate, purified stearic acid, sodium alginate and colloidal anhydrous silica.
[b] Fully formulated coating system containing: PVAP, titanium dioxide, triethyl citrate and stearic acid.
[c] Based on polymer weight.
[d] Based on solvent weight.

Table 3
Coating parameters for the different polymer systems.

Formulation	Inlet temp. (°C)	Outlet temp. (°C)	Fan capacity	Atomizing pressure (bar)	Flow rate (mL/min)	Coating level (mg/cm^2)
EUDRAGIT® L30D-55	46	34	17	0.2	4	5
EUDRAGIT® L100-55	40	32	17	0.2	3	5
EUDRAGIT® L100	40	32	17	0.2	3	5
HPMCAS-LF	46	43	17	0.2	7	5,6,7,8
HPMCAS-MF	56	60	17	0.2	7	5,6,7
PVAP(aqueous)	58	43	17	0.2	7	5, 6,7,8% TWG
PVAP (organic)	52	40	9	0.2	7	5% TWG
CAP	40	34	17	0.2	5	5
HP-50	46	38	17	0.2	4	5
HP-55	60	43	17	0.2	5	5

$CO_{2(g)}$ into Hanks buffer, in excess, increases the concentration of $CO_{2(aq)}$ which promotes the formation of carbonic acid and thus results in a decrease in the pH of the buffer system.

$$pH = pKa + \frac{[HCO_3^-]}{[H_2CO_3]} \quad (3)$$

In this experiment, sufficient $CO_{2(g)}$ is purged into the system to achieve the desired bicarbonate:carbonic acid ratio, resulting in a pH 6.8 bicarbonate buffer (H_2CO_3 p*K*a, 6.38) while keeping the concentration of bicarbonate unchanged, before and after modification. The concentration of bicarbonate in *m*Hanks was determined using a titration method. A known amount of hydrochloric acid was added into *m*Hanks buffer and the excess acid after neutralising bicarbonate is titrated with sodium hydroxide using an Autotitrator MPT-2 (Malvern Instruments Ltd., Worcestershire, UK), and the endpoint was determined by a titration curve. The molar concentration of the reacted acid is equal to that of the bicarbonate in the sample; a correction factor is applied for the available phosphate species in the solution. The final ionic composition and buffer capacity of the *m*Hanks buffer are compared with phosphate buffer and human jejunal fluid in Table 4.

The pH in the dissolution media was measured at periodic intervals during the dissolution experiments and was maintained at 6.8 ± 0.05 by continuously sparging CO_2 into the media. Six polyurethane flow tubes (Freshford Ltd., Manchester, UK), one for each dissolution vessel, were used to regulate the CO_2 flow via a manifold assembly.

Table 4
Comparison of the ionic composition (mM) and buffer capacity of small intestinal fluids [12,15,39] and phosphate and *m*Hanks media.

Composition	Human jejunal fluid	Phosphate buffer (0.05 M, pH 6.8)	*m*Hanks buffer (pH 6.8)
Bicarbonate	7.1	Not present	4.17
Phosphate	0.8	50	0.8
Potassium	5.1	50	5.8
Sodium	142	29	142
Chloride	131	Not present	143
Calcium	0.5	Not present	1.3
Magnesium		Not present	0.8
Buffer capacity (mmol/L/ΔpH)	3.2[a]	23.1 ± 0.3	3.1 ± 0.2

[a] Measured from luminal aspirates [40].

Buffer capacity (β) of the *m*Hanks was measured by adding aliquots of 0.1 M HCl to 100 mL of the buffer system. Buffer capacity was then calculated using Eq. (4). Buffer capacity was also measured at various time intervals while the medium was purged with

$CO_{2(g)}$ to maintain the pH at 6.8 over the period of the dissolution test. The reproducibility of the buffer capacity and pH was checked by experimental measurements on different occasions and on different days.

$$\beta = \frac{\Delta AB}{\Delta \mathrm{pH}} \quad (4)$$

where ΔAB is the small increment in mol/L of the amount of acid or base added to produce a pH change of ΔpH in the buffer. Buffer capacity was measured at a pH change of 0.5 units on addition of the acid.

2.4.3. Dissolution testing

Drug release from the coated prednisolone tablets was evaluated using a USP-II apparatus (Model PTWS, Pharmatest, Hainburg, Germany). The tests were conducted in triplicate, in 900 mL dissolution medium maintained at 37 ± 0.5 °C. A paddle speed of 50 rpm was employed. The tests were conducted under sink conditions. Tablets were tested for 2 h in 0.1 M HCl, followed by pH 6.8 phosphate buffer (composition: 50 mM KH_2PO_4 and 23.5 mM NaOH; pH adjusted with 1 M HCl / NaOH solutions) or pH 6.8 *m*Hanks buffer (composition: 136.9 mM NaCl, 5.37 mM KCl, 0.812 mM $MgSO_4 \cdot 7H_2O$, 1.26 mM $CaCl_2$, 0.337 mM $Na_2HPO_4 \cdot 2H_2O$, 0.441 mM KH_2PO_4, 4.17 mM $NaHCO_3$, $CO_{2(g)}$ quantity sufficient to reach pH 6.8) (Table 4). The pH of the *m*Hanks buffer was maintained by sparging CO_2 into the medium during the dissolution studies as described in Section 2.4.2.

The quantity of prednisolone released from tablets coated with HPMCAS-LF, HPMCAS-MF, EUDRAGIT® L30 D-55, L100-55 and L100 was determined using an in-line UV spectrophotometer (Cecil 2020, Cecil Instruments Ltd., Cambridge, UK) at a wavelength 247 nm. Data were processed using Icalis software (Icalis Data Systems Ltd, Berkshire, UK). In the case of the tablets coated with CAP, HP-50, HP-55 and PVAP, drug release was determined using HPLC-UV due to the interference of UV absorbance of the polymers at the peak wavelength of prednisolone.

The HPLC-UV system used was a Hewlett Packard 1050 Series HPLC system (Agilent Technologies, UK). Dissolution samples were filtered through 0.22-μm filters (Millipore Ltd, Ireland), and 10 μL was then injected into a reverse phase C8 (5 μm particle size) column (Waters, Massachusetts, USA). The chromatographic conditions were as follows: column temperature of 40 °C, a pressure of 1800 psi, mobile phase consisting of water:tetrahydrofuran:methanol (68.8:25:6.2 v/v), and a flow rate of 1.0 mL/min. Prednisolone was detected at a wavelength of 254 nm.

The dissolution data were analysed by two-way ANOVA followed by a Tukey post hoc analysis with 99.8% confidence interval using Univariate General Linear Model tool in PASW Statistics 18 (SPSS Inc., Illinois, USA).

3. Results and discussion

3.1. Acid resistance

The acid uptake results for the various enteric formulations after exposure to 0.1 M HCl for 2 h are shown in Table 5. All organic-based enteric coating formulations showed low acid uptake values at a coating level of 5 mg/cm^2 (PVAP organic at 5% TWG), indicating good acid resistance. Notably, the aqueous dispersion of the methacrylic acid copolymer, EUDRAGIT® L30D-55, showed comparable acid-resistant properties to its organic counterpart EUDRAGIT® L100-55, at the same coating level of 5 mg/cm^2. The aforementioned formulations remained intact and showed no physical changes after acid treatment.

The aqueous cellulose-based polymer HPMCAS-LF showed poor acid resistance at 5 mg/cm^2; acid uptake values were high, and the tablets swelled in acid (Table 5). Therefore, higher coating levels (6, 7 and 8 mg/cm^2) were investigated and a coating of 7 mg/cm^2 was required for sufficient acid protection. This was also the case with HPMCAS-MF, which required a 7 mg/cm^2 coating level to achieve acid protection (Table 5). The aqueous coating formulation of PVAP (Sureteric®) also required a higher coating level (7% TWG) to achieve acid resistance compared to its organic-based counterpart (Opadry®) (5% TWG). The film forming mechanisms of aqueous polymer dispersions are distinct from that of the organic solutions and require complete particle coalescence to obtain film coatings with the desired properties [23,24]; this often results in the need for a higher coating level for the aqueous formulations to achieve adequate acid resistance compared to organic formulations.

The organic formulations with a coating level of 5 mg/cm^2 and/or 5% TWG were further subjected to dissolution testing. The aqueous EUDRAGIT® L30D-55 formulation was also tested at a coating level of 5 mg/cm^2. The remaining aqueous formulations were tested at 7 mg/cm^2 (HPMCAS-LF and HPMCAS-MF) and 7% TWG (PVAP aqueous/Sureteric®).

3.2. Evolution of pH 6.8 bicarbonate buffer (mHanks)

In the present study, pH 6.8 physiological bicarbonate buffer (*m*Hanks buffer) was successfully developed by modification of Hanks buffer (pH 7.4). An attempt has been made previously to develop pH 6.8 physiological bicarbonate buffers by sparging $CO_{2(g)}$ into 0.9 % sodium chloride solution, and the pH was achieved and maintained by titration using 1 N sodium hydroxide [25]. Several issues with this approach were raised including the effect of thermal equilibrium on pH and buffer capacity, loss of $CO_{2(g)}$ during transfer, cost, long set-up time and more profoundly bubble formation and changes in the hydrodynamics of the system, resulting in high variability and poor reproducibility of the dissolution profiles [26]. These authors subsequently proposed a modified

Table 5
Acid uptake of enteric coated prednisolone tablets.

[A] Organic formulations						
	CAP	EUDRAGIT® L 100	EUDRAGIT® L100-55	HP-50	HP-55	PVAP[a] (Opadry®)
Coating Level (mg/cm^2)	5[b]	5[b]	5[b]	5[b]	5[b]	5%[b]
Acid uptake (%)	2.6	4.2	2.7	2.8	2.1	1.2

[B] Aqueous formulations													
	EUDRAGIT® L30D-55	HPMCAS-LF				HPMCAS-MF			PVAP[a] (Sureteric®)			
Coating level (mg/cm^2)	5[b]	5	6	7[b]	8	5	6	7[b]	5%	6%	7%[b]	8%
Acid uptake (%)	3.1	19.4	7.8	7.5	6	9.9	7.8	5.5	30.3	14	8.3	7.2

[a] Coating level based on tablet weight gain (%).
[b] Coating levels selected for dissolution testing in buffers.

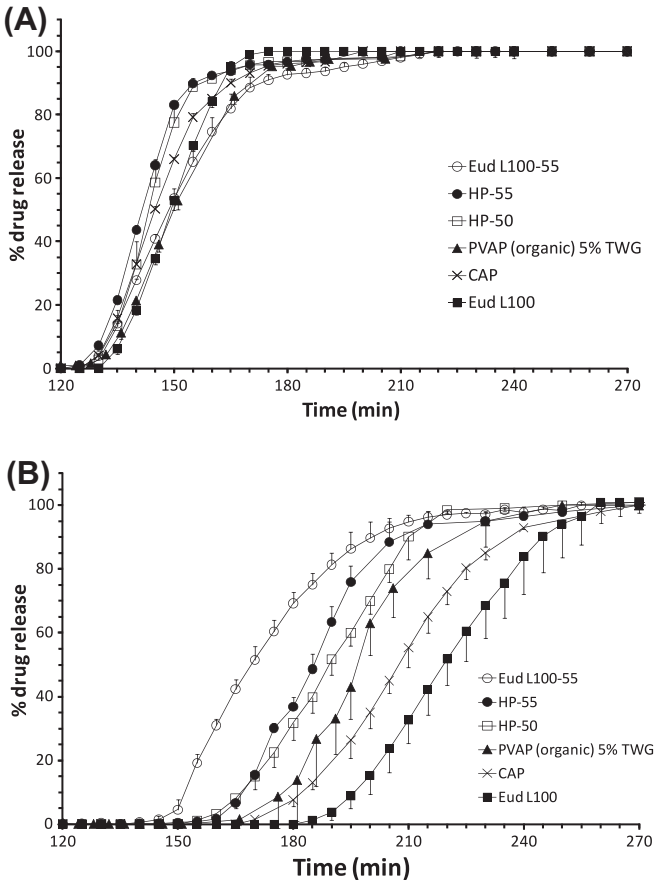

Fig. 1. Drug release for organic solution coated (5 mg/cm^2 unless otherwise indicated) prednisolone tablets in 0.1 M HCl for 2 h (data not shown) followed by pH 6.8 [A] phosphate buffer and [B] *m*Hanks bicarbonate buffer.

methodology to overcome these issues by keeping sodium hydroxide at a constant concentration and maintaining the pH using $CO_{2(g)}$ [26]. However, maintaining the pH was problematic due to poor resistance of the buffer to pH change and consequently a much higher buffer capacity medium (30 mmoles/L/ΔpH) was used in these tests. In the proposed *m*Hanks physiological bicarbonate buffer used in the present study, the desired pH was achieved and maintained by sparging $CO_{2(g)}$ into the medium just 2 cm below the liquid surface at a very low flow rate compared to what has been used previously, hence avoiding significant effects on hydrodynamics and changes in buffer capacity of the medium throughout the duration of the experiment. The buffer capacity and pH were reproducible when tested on different occasions and on different days of experiments. In addition, the *m*Hanks closely resembles the ionic composition and buffer capacity of the luminal contents (Table 4), compared to the previously proposed medium.

3.3. Dissolution testing

Drug release profiles for tablets coated with the organic and aqueous formulations in pH 6.8 phosphate buffer post-exposure to 0.1 M HCl for 2 h are shown in Figs. 1A and 2A. All enteric coated formulations, organic and aqueous showed rapid and comparable drug release profiles in pH 6.8 phosphate buffer. This buffer cannot therefore discriminate between the various polymer coatings.

Drug release from prednisolone tablets coated with the organic and aqueous formulations in pH 6.8 *m*Hanks buffer post-exposure to 0.1 M HCl for 2 h is shown in Figs. 1B and 2B. Drug release was delayed from all enteric coated tablets in pH 6.8 *m*Hanks buffer, compared to that in pH 6.8 phosphate buffer. Moreover, distinctive dissolution profiles were observed for the various enteric coated formulations in this bicarbonate buffer. The dissolution profiles of the organic polymers in *m*Hanks buffer (Fig. 1B) are significantly different from each other, except for HP-50 and HP-55. The dissolution rank order for the enteric coating formulations was EUDRAGIT® L100-55 > HP-55 ≈ HP-50 > PVAP > CAP > EUDRAGIT® L100.

The enteric formulations based on aqueous polymer dispersions also showed slower drug release and longer lag times in bicarbonate buffer when compared to compendial phosphate buffer. In contrast to the aqueous formulations of PVAP and HPMCAS, the methacrylic acid copolymer EUDRAGIT® L30D-55 showed comparable acid-resistant properties to its organic form (EUDRAGIT® L100-55) at 5 mg/cm^2 coating level. Drug release from this aqueous coating system was similar to that from the organic coating system in pH 6.8 *m*Hanks buffer (Figs. 1B and 2B). Films formed from this aqueous polymeric dispersion has a minimum film formation temperature lower than room temperature (<23 °C), and the particles in the latex EUDRAGIT® L30D-55 dispersion have relatively small size (mean particle size of 0.2 μm) [27]. These properties of the dispersion provide ease of particle coalescence during film formation and ensured comparable properties of the aqueous film coating to the organic coating.

The bicarbonate buffer (*m*Hanks) not only discriminated between the different enteric polymer coatings, but is indeed better reflective of the reported delayed disintegration times of enteric coated products in the human intestine [6–10]. For example, the EUDRAGIT® L30 D-55 coated tablet tested in this study has also been evaluated in a scintigraphic study in humans [11]. The mean disintegration time of the formulation (post gastric emptying) was 66 min. The delay in disintegration observed under *in vivo* conditions is more closely reflected under *in vitro* dissolution using bicarbonate media (*m*Hanks) than in compendial phosphate buffers (Fig. 2).

Differences in the release profiles of the various enteric formulations in *m*Hanks also confirms that most of the enteric coating systems are not interchangeable in terms of drug release as would be suggested by the release data in phosphate buffer, and therefore testing these enteric formulation in *m*Hanks also provides a rank order for these systems in terms of dissolution.

3.4. Explanation for the distinct behaviour of enteric formulations in physiological bicarbonate media

The mechanism of carboxylic acid polymer dissolution in aqueous solutions is different to that of non-ionic polymers [28] because it involves an additional ionization step that stabilizes the polymer chains. The process of dissolution consists of five steps: (i) diffusion of water and hydroxyl ions into the polymer matrix to form a gel layer, (ii) ionization of polymer chains in the gel layer, (iii) disentanglement of polymer chains out of the gel layer to the polymer–solution interface, (iv) further ionization of polymer chains at the polymer interface, and (v) diffusion of disentangled polymer chains away from the interface towards the bulk solution [29]. The rank order in the dissolution of the organic polymer formulations observed in *m*Hanks buffer can be explained by the determinant factors for enteric coating dissolution: polymer p*K*a and chemical structure [30]. Polymers with higher p*K*a values reflected by higher dissolution pH thresholds, such as EUDRAGIT® L100, showed slower drug release. Apart from p*K*a, structure of the polymer backbone is also an important factor controlling the dissolution of acidic polymers. For instance, CAP has a water-insoluble back bone and dissolves slower than HPMCP (HP-50/HP-55) and PVAP which have water-soluble backbones [31].

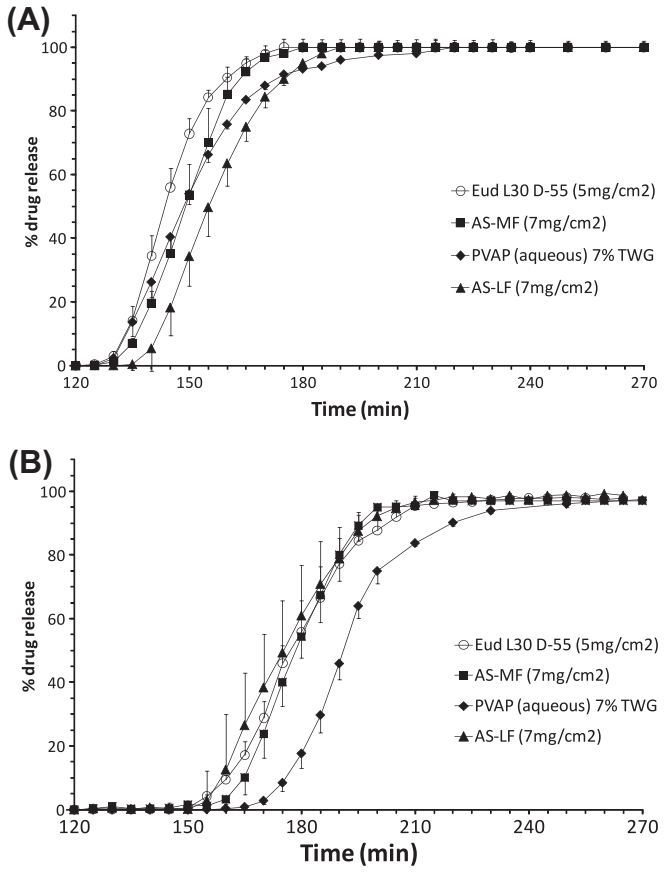

Fig. 2. Drug release for aqueous dispersion coated prednisolone tablets in 0.1 M HCl for 2 h (data not shown) followed by pH 6.8 [A] phosphate buffer and [B] *m*Hanks bicarbonate buffer.

It is also important to understand the interaction of the functional groups on the polymer chain with various ions and buffer species present in the dissolution media. There have been several reports in the literature on the extremely low rates of dissolution of enteric polymers when tested in normal saline or very weak buffer solutions at pH values well above the dissolution thresholds, whereas fast dissolution was observed at the same pH but in relatively higher strength buffers [19,30,32,33]. This suggests that pH is not the only factor controlling drug release from enteric polymers and other factors also play an important role in the dissociation of the polymer chains. The composition of the dissolution medium, especially the buffer salt, profoundly influences the dissolution rate of enteric polymers. The influence of salts on the dissolution of enteric polymers can be explained by general base catalysis. The acid polymers dissociate through proton transfer to the Brönsted base, resulting in the formation of the conjugate base of the polymer and hydronium ions. In the presence of a basic salt, the rate of proton transfer is increased by the higher affinity of the water in accepting the proton and consequently the dissolution rate is increased. By obeying the Brönsted catalysis law, the dissolution rate was directly proportional to the p*K*a and the concentration of the salts present in the solution [32].

It was also explained that apart from p*K*a, buffer capacity of the salts also affects acidic polymer dissolution [34]. Hydrogen ions are generated at the polymer–solution interface during polymer dissociation [29] and contribute to a pH drop near the surface of the dissolving carboxylic polymer [35]. Removal of these hydrogen ions at the interface increases the polymer dissolution rate and can be facilitated by reacting with proton acceptors (buffer species), depending on their buffer capacities which directly link to the p*K*a of the buffer salt. Phosphate buffer has an effective p*K*a of 7.19, and a resultant higher buffer capacity (23.1 ± 0.3 mmol/L/ΔpH) thus provides greater driving force for the acidic polymer dissolution than that of bicarbonate with a p*K*a of 6.31 and a much lower buffer capacity (3.1 ± 0.2 mmol/L/ΔpH). Although the above difference in buffer capacity can also be attributed to the difference in buffer concentration, Sheng et al. reported that phosphate buffer had a 23% higher buffer capacity at the solid liquid interface compared to bicarbonate buffer, at the same pH and buffer concentration [36]. Besides buffer capacity, the ionic composition and strength of the dissolution media also has a profound effect on the reaction rate between the polymer film and the basic buffer species; a drastic change in the dissolution rate has been reported from enteric coated formulations with change in ionic strengths of the media [19,37,38]. When we evaluated the enteric coated formulations in a weaker phosphate buffer (9 mM), having the same pH and buffer capacity as the *m*Hanks buffer, drug release was further delayed than in *m*Hanks (data not shown). This reiterates the crucial importance of buffer species and ionic composition of the media and suggests that how the buffer capacity is generated is also important in addition to buffer capacity itself. This will form the basis of future investigations.

4. Conclusions

Drug release from the different enteric coated formulations was rapid and comparable in the commonly used compendial pH 6.8 phosphate buffer, failing to reflect the general literature values on *in vivo* variation and delayed release of enteric coated products in the human small intestine. A pH 6.8 physiological bicarbonate buffer (*m*Hanks) was developed as a dissolution medium to better simulate small intestinal luminal fluid. This buffer was able to discriminate between the different enteric polymer coated systems, providing a rank dissolution order, and is likely to improve *in vitro–in vivo* correlations. This new knowledge can also be useful in the rational design of enteric coated products designed to target different sites in the small bowel.

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