Use of a Physiologic Bicarbonate Buffer System for Dissolution Characterization of Ionizable Drugs

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Purpose. The aim of this study was to examine if sparging with CO_2(g) could be used to establish stable biorelevant bicarbonate buffers, in aqueous medium, for use in dissolution characterization of low-solubility ionizable drugs.

Methods. Preparation of the bicarbonate-containing dissolution medium was monitored by use of a commercially available fiberoptic probe to measure the concentration of dissolved CO_2(aq). Intrinsic dissolution measurements at 100 rpm, 37°C for indomethacin and dipyridamole were performed using a rotating disk and UV detection at pH 6.8 and 5.0 in a USP dissolution vessel apparatus.

Results. Indomethacin dissolution at pH 6.8 was significantly impacted by the concentration of CO_2(g) in the sparging gas. Dipyridamole flux at pH 6.8 was independent of buffer species or buffer concentrations studied. However, dipyridamole dissolution at pH 5 was also a strong function of the concentration of CO_2(g) in the sparging gas.

Conclusions. Stable bicarbonate biorelevant buffers could be established to perform intrinsic dissolution rate determinations for indomethacin and dipyridamole as long a continuous gas sparging of CO_2(g) was used. Depending of the pH of the dissolution medium, the intrinsic dissolution rates of both indomethacin and dipyridamole were affected by the bicarbonate concentration. Sparging with CO_2(g) to create physiologic buffers has a unique advantage over conventional buffers in that gas sparging serves as a continuous source of bicarbonate buffer species. This advantage was demonstrated by performing dissolution experiments at pH values typically associated with the fed state (pH 5) and applying relatively low CO_2(g) pressures, resulting in bicarbonate concentrations less than 0.5 mM. It was demonstrated that CO_2(g) sparging at a pH consistent with the fed state created an *in-situ* bicarbonate buffer at low concentrations, which had a significant impact on the dissolution of a basic drug such as dipyridamole.

KEY WORDS: physiologic dissolution media; biorelevant dissolution media; bicarbonate buffer; gas sparging; intrinsic dissolution.

INTRODUCTION

Acceptance of the Biopharmaceutics Classification System (BCS) as a tool to categorize new chemical entities (NCEs) has refocused attention on those classes of NCEs for which *in vitro–in vivo* correlations may be expected (1). A consequence of this attention has been renewed examination of how *in vitro* dissolution evaluations are performed. It is commonly agreed that in trying to develop an *in vitro–in vivo* correlation for an NCE, the dissolution method used to assess *in vitro* formulation performance should, as closely as possible, mimic *in vivo* conditions (2). In order to establish a biorelevant dissolution test, the investigator must design an appropriate dissolution medium by choosing an appropriate pH and buffer system. One seemingly obvious choice, which has not been used often by pharmaceutical researchers, is to use the same buffer that predominates in human physiology, namely, a bicarbonate buffer. Bicarbonate is a ubiquitous component of human extracellular fluids and is actively secreted by the pancreas to neutralize gastric secretions in the gastrointestinal lumen. It has also been shown that the epithelial cells of the duodenum secrete bicarbonate and that the duodenal secretion of bicarbonate is an important mechanism to protect the duodenal epithelium against acid discharged from the stomach (3).

The influence of dissolution medium on accurate characterization of dissolution rate of ionizable drugs has been examined. The buffer system composition of the dissolution medium is especially critical for low-solubility ionizable drugs, where it has been demonstrated that the concentration and identity of buffer species themselves significantly impact dissolution, even if pH is held constant (4–7). Aunins *et al.* demonstrated that for a low-solubility acidic drug such as indomethacin (intrinsic solubility of 0.9 μg/mL), use of a 25 mM phosphate buffer in the dissolution medium at pH 7 accelerates the dissolution by 20 times over the rate at pH 7 in unbuffered medium (5). In this report we describe intrinsic dissolution rates for indomethacin and dipyridamole in bicarbonate buffers at pH 5 and 6.8 and compare these results with those from other suggested dissolution media that do not contain bicarbonate buffers.

The physical chemistry of the carbon dioxide and aqueous bicarbonate species is reviewed schematically in Fig. 1. Figure 1 shows the relationship between carbon dioxide [CO_2(g)] in the headspace above an aqueous solution and dissolved carbon dioxide [CO_2(aq)] in the solution. Once dissolved, carbon dioxide in the aqueous medium hydrates to form carbonic acid (H_2CO_3). Finally, carbonic acid (pK_{a1} = 6.1, 25°C, and ionic strength 0.1) will dissociate to yield a proton and bicarbonate (HCO_3^-). The ionization of bicarbonate to yield carbonate (CO_3^{-2}) can occur, but the pK_{a2} of bicarbonate is so high (pK_{a2} = 9.9, 25°C, and ionic strength 0.1) that it is of little consequence in most physiologically relevant systems (8). In order to maintain a stable bicarbonate buffer concentration for dissolution testing, one must maintain a constant partial pressure of carbon dioxide (P_{CO_2}) over the solution or continuously sparge CO_2(g) into the medium to saturate it with dissolved carbon dioxide [CO_2(aq)].

Table I is a summary list of the bicarbonate concentrations that have been reported for luminal gastrointestinal fluids in man and dog. The dynamic range of the data reported in Table I is remarkable. These data underscore the dynamic complexity of the gastrointestinal fluid luminal environment. There is also a substantial historical gastroenterology literature regarding the direct *in vivo* measurement of gastrointestinal P_{CO_2}. Note, as shown in Fig. 1, if the P_{CO_2} and pH of the gastrointestinal fluid is known, then the equilibrium concen-

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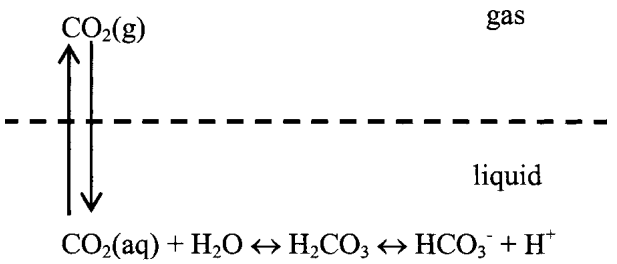


Fig. 1. Schematic for gas/liquid equilibria for CO_2/bicarbonate buffer system.

trations of dissolved CO_2(aq) and bicarbonate can be calculated (8). Physicians and gastroenterologists have been measuring *in vivo* P_{CO_2} via gastrointestinal tonometry since the 1950s. Initially gastrointestinal tonometry was used to assess the ventilatory status of pediatric patients during mechanical ventilation; however, more recently, physicians have begun using this technique to assess metabolism and splanchnic tissue perfusion of critically ill patients (24). The *in vivo* P_{CO_2} values that have been reported using tonometry can depend on many factors, including gastrointestinal location (gastric or duodenal), local splanchnic tissue perfusion, pancreaticobiliary secretory status, and fed or fasted status (24). Typically, reported tonometry P_{CO_2} values range from 4 CO_2 %atm, which is comparable to P_{CO_2} in arterial blood, to 37 CO_2 %atm, reported postprandially from duodenal samples in man (14,24). At an extreme, Rune and Henricksen reported mean postprandial duodenal canine P_{CO_2} values of 66 CO_2 %atm (19). In these investigations, we confined our range of experimental P_{CO_2} concentrations from 5 to 20 CO_2 %atm.

MATERIALS AND METHODS

To establish the bicarbonate buffer, our experimental procedure included continuous sparging with CO_2(g) of a fixed partial pressure into 1 L of 0.9%(w/v) NaCl, pH 6.8 or 5.0, aqueous solution with magnetic stirring. The solution pH was fixed at 6.8 so that comparisons could be made to dissolution experiments done in USP simulated intestinal fluid (SIF) or at pH 5.0 so that comparisons could be made to fed state simulated intestinal fluid (FeSSIF) (25,26). Carbon dioxide sparging was accomplished using compressed gas cylinders (primary standards containing 5, 10, 15, and 20 CO_2 %atm in air, Airgas, Cheshire, CT) and gas regulators (Airgas model UPE 25) with fluoropolymer tubing (Chemfluor, VWR) and a stainless steel frit (Upchurch Scientific, Oak Harbor, WA; 10-μm solvent inlet filter A310) at a flow rate (flowmeters; Gilmont Instruments, model GF-4540) of approximately 300 ml/min. While the solution was equilibrating, the pH and dissolved carbon dioxide [CO_2(aq)] content were monitored. The pH was monitored with a standard pH meter and electrode (Corning model 307 pH meter, Corning 476600 general purpose combination electrode), and the concentration of CO_2(aq) was followed with a fiberoptic probe (YSI Biovision 8500 CO_2 monitor). Because protons are produced as carbon dioxide dissolves, the pH of the solution was continuously adjusted to maintain a pH of 6.8 or 5.0 by addition of 1 N NaOH. Fig. 2 shows a typical concentration vs. time profile for dissolved carbon dioxide using 15 CO_2 %atm as the sparging gas at pH 6.8 in 1 L of 0.9% (w/v) NaCl. After 30–40 min of stirring and sparging, the 1-L solution was equilibrated [saturated with CO_2(aq)] and was ready for use as dissolution testing medium. Also shown in Fig. 2 is the concentration of CO_2(aq) after the sparging was stopped and the medium was allowed to equilibrate at room temperature with continued stirring. If CO_2(g) is not continuously sparged into the solution, the concentration of dissolved carbon dioxide CO_2(aq) falls rapidly with continued stirring, and the pH of the solution will increase. Note that bicarbonate concentration, and hence the bicarbonate buffer, is stable as long as continuous sparging occurs.

During dissolution experiments, 800 ml of the 1 L

Table I. Summary of Reported Bicarbonate Luminal Concentrations (Range or Mean Values)

GI location	$[HCO_3^-]$ (mEq/L)	Species
Stomach	5–33	Dog[9]
	7.3	Man[10]
	9–20	Man[11]
Duodenum	14–22	Dog[9]
	2.7	Man[12]
	6.7	Man[13]
	10	Man (fed)[14]
	15	Man[15]
Jejunum	1–4	Dog[16]
	14	Dog[17]
	5–30	Dog[9]
	2–20	Man[18]
	5–10	Man[19]
	6–20	Man[20]
	17	Man[12]
	30	Man[15]
	30	Man[21]
Ileum	9–37	Dog[16]
	67	Dog[17]
	70–114	Dog[9]
	40	Man[20]
	50	Man[22]
	70	Man[15]
	74	Man[23]
	75	Man[21]

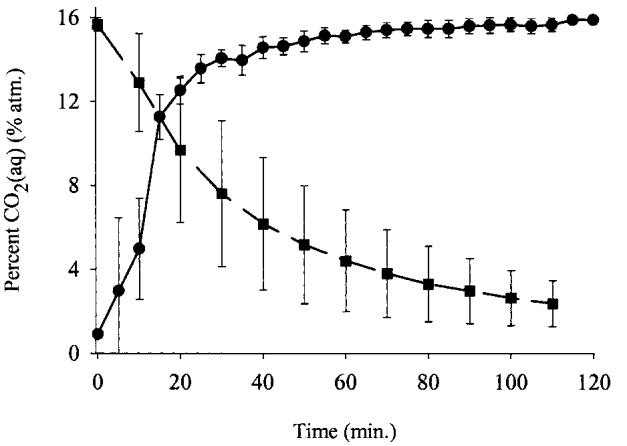


Fig. 2. Uptake and loss of dissolved CO_2(aq) in 1 L pH 6.8, 0.9% NaCl (w/v) aqueous solution at 25°C sparged with 15 CO_2 %atm. (●) uptake experiments, (■) loss (no sparging). Error bars represent ±1 standard deviation.

sparged equilibrated pH 6.8 or pH 5.0 solution was transferred to a USP apparatus 2 dissolution vessel (VanKel 7010) at 37°C. The solution was allowed to thermally equilibrate, as it was continuously sparged with carbon dioxide (stainless steel 10-μm solvent inlet filter frit or ceramic frit, at 250–500 ml/min.). At the start of a dissolution experiment, a Wood's die apparatus (rotating disk) containing a compressed plug of indomethacin (350 mg compressed at 6000 lb for 60 s in a Carver press) or dipyridamole (350 mg compressed at 20,000 lb for 90 s in a Carver press) was lowered into the solution and stirred at 100 rpm. Filtered samples were automatically withdrawn periodically, with fluid recirculation, from the dissolution vessel and assayed via UV spectrophotometry (Cary 50 UV-Vis) using flow-through cells for absorbance. Absorbance values at $\lambda = 265$ nm for indomethacin and at $\lambda = 290$ nm for dipyridamole were converted to concentration using standard indomethacin or dipyridamole solutions of known concentration, respectively. Dissolution flux (mass/time·area) of indomethacin and dipyridamole were calculated using the slope of the concentration vs. time plot, volume of dissolution medium, and area of the exposed disk (2.84 cm^2). Individual dissolution experiments typically were carried out for several hours.

Buffer capacity of the dissolution medium was also checked at the end of the dissolution run during several of the experiments to ensure that the bicarbonate concentration was maintained during the dissolution experiments. Samples of the dissolution medium (25–50 ml) were immediately withdrawn from the dissolution vessel and covered with a layer (20–50 ml) of paraffin oil The paraffin oil layer, which floats above the aqueous sample, slows the loss of carbon dioxide from the sample. This procedure was used successfully in studies where the concentration of CO_2(aq) and buffer capacity of collected human saliva was of interest (27). Once the paraffin oil was added, the samples were then immediately titrated with 0.1 N NaOH. The pH and titrant volumes were recorded, and buffer capacities were calculated.

For comparative purposes, dissolution flux of indomethacin and dipyridamole from a rotating disk at 100 rpm into the following media were determined: simulated intestinal fluid (SIF) without pancreatin and fasted-state simulated intestinal fluid (FaSSIF). Dipyridamole dissolution flux was also determined in fed-state simulated intestinal fluid (FeSSIF). Because FeSSIF contains 144 mM acetic acid as the primary buffer in addition to sodium taurocholate and lecithin, dissolution in 144 mM acetic acid at pH 5 was also determined for dipyridamole to show the buffer species effect. To define the effect of pH on dissolution, without added buffer species, dissolution at pH 6.8 and pH 5 with 0.9% (w/v) NaCl corresponding to the pHs of FaSSIF and FeSSIF, respectively, were determined. SIF was prepared according to USP XXIV (25). FaSSIF and FeSSIF were prepared according to a recent publication (26).

KCl, NaCl, NaOH, and paraffin oil were all received from EM Science; indomethacin (lot 117-0595) and taurocholic acid (sodium salt) from Sigma Chemical Company; and dipyridamole (lot 11502AO) from Aldrich Chemical Company Inc. L-α-Phosphatidylcholine (lecithin) was received from Avanti Polar Lipids, Inc. KH_2PO_4 was from Fisher Scientific. Glacial acetic acid was from J. T. Baker Chemical Company.

RESULTS

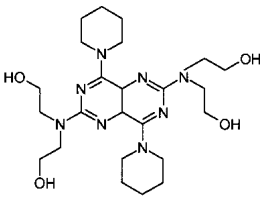
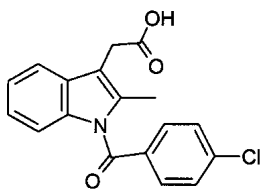
In these investigations, the dissolution of acidic and basic drugs with low intrinsic solubilities from a rotating disk were studied. The aim of these investigations was to establish *in situ* bicarbonate buffers by sparging with CO_2(g) and to study the impact of bicarbonate buffers on dissolution relative to other biorelevant dissolution media containing phosphate and acetate buffers. Table II lists the intrinsic solubility and pK_a of each compound. Both compounds have characteristically low intrinsic solubilities. Fig. 3 shows the flux of indomethacin at pH 6.8 using four different concentrations of carbon dioxide (5, 10, 15, and 20 CO_2 %atm) as sparging gas. The theoretical concentrations of bicarbonate that are established at 5, 10, 15, and 20% CO_2 %atm are 6.4, 12.9, 19.3, and 25.8 mM HCO_3^-, respectively (8). Sparging with 5 CO_2 %atm develops an aqueous solution that contains 6.4 mM HCO_3^-, whereas sparging with 20 CO_2% atm develops an aqueous solution that contains 25.8 mM HCO_3^-. Higher partial pressures of CO_2(g) in the sparging gas lead to higher concentrations of dissolved CO_2(aq) and, hence, to higher concentrations of bicarbonate in the dissolution medium.

The dissolution rate of a solid free acid such as indomethacin is directly impacted by the concentration of basic bicarbonate species because of the ionization equilibrium of the acid that drives the solid dissolution. Indomethacin has a low intrinsic solubility (0.9 μg/mL) and is thus unable to effectively buffer the microenvironmental pH associated near the solid surface in the boundary layer (4–6). For indomethacin dissolving in a bicarbonate buffer system, the dissolution equilibrium that drives dissolution is specified in Eq. (1).

$$HA + HCO_3^- \leftrightarrow H_2CO_3 + A^- \leftrightarrow A^- + CO_2(aq) + H_2O \quad (1)$$

where HA represents the free acid of indomethacin and A^- represents the conjugate base of indomethacin. As expected,

Table II. Physicochemical Properties of Compounds Tested with Carbon Dioxide Sparging during Dissolution

Compound	Intrinsic solubility at 25°C (μg/ml)	pK_a
 Dipyridamole	5.8†	6.05,†6.1[28]
 Indomethacin	0.9§[,29]	4.17§[,29]

† Internal Boehringer Ingelheim Pharm. Inc. report, $\mu = 0.1$ with NaCl.

§ pH 2, $\mu = 0.5$ with KCl.

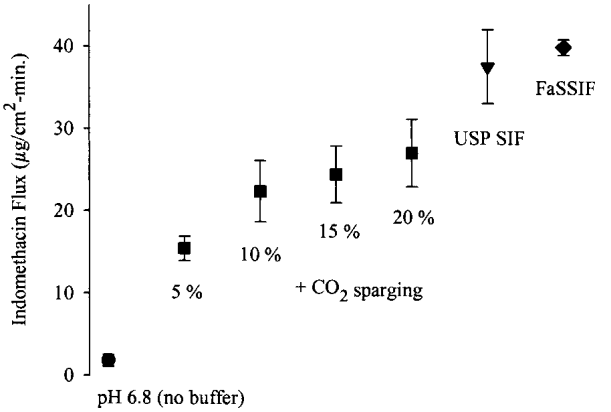


Fig. 3. Indomethacin flux at pH 6.8, 0.9% NaCl, 37°C 100 rpm. (●) pH 6.8, no buffer, (■) pH 6.8 plus CO_2 sparging, (▼) USP SIF, 50 mM KH_2PO_4, (♦) FaSSIF, pH 6.5, 29 mM KH_2PO_4, 3 mM Na taurocholate, 0.75 mM lecithin, 103.3 mM KCl. Error bars represent ±1 standard deviation.

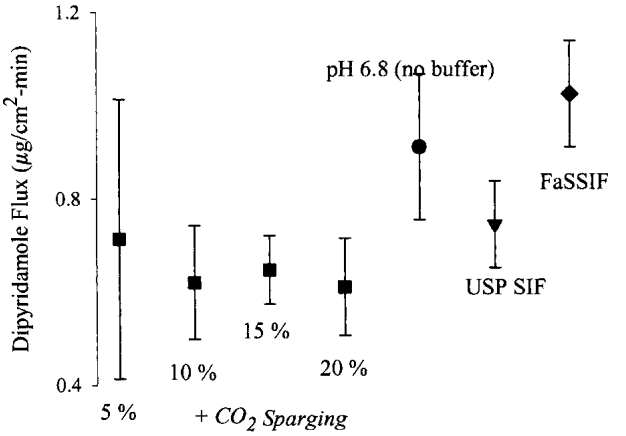


Fig. 4. Dipyridamole flux at pH 6.8, 0.9% NaCl, 37°C 100 rpm. (●) pH 6.8, no buffer, (■) pH 6.8 plus CO_2 sparging, (▼) USP SIF, 50 mM KH_2PO_4, (♦) FaSSIF, pH 6.5, 29 mM KH_2PO_4, 3 mM Na taurocholate, 0.75 mM lecithin, 103.3 mM KCl. Error bars represent ±1 standard deviation.

the higher the partial pressure of carbon dioxide (P_{CO_2}) used to sparge the dissolution media, the more bicarbonate is formed in the aqueous medium, which forces the equilibrium described in Eq. (1) further to the right. Therefore, the dissolution rate of the free acid (HA) increases as more conjugate base (A^-) is produced, and the dissolution rate of indomethacin increases with increasing bicarbonate concentration. For comparison purposes, the dissolution of indomethacin in other biorelevant dissolution media at pH 6.8 and FaSSIF (pH 6.5) is also shown in Fig. 3. In SIF (50 mM KH_2PO_4 at pH 6.8), the free acid of indomethacin reacts with the basic species in the buffer ($H_2PO_4^-$), and the reaction of indomethacin with monobasic phosphate drives the solid dissolution. When no buffer is present (aqueous solution 37°C, pH 6.8, 0.9% NaCl), the dissolution flux for indomethacin drops significantly by a factor of 20 as compared to the flux in SIF. Even though FaSSIF contains lesser amounts of phosphate buffer (29 mM KH_2PO_4) and a slight change in pH (pH 6.5) as compared to SIF, the measured dissolution rate of indomethacin in FaSSIF is comparable to the rate in SIF. The reason that dissolution in FaSSIF is comparable to that in SIF, even though FaSSIF has a lower pH and a lower phosphate buffer concentration, is most likely the solubilizing effect of taurocholate and lecithin contained in FaSSIF. Taurocholate and lecithin increase the intrinsic solubility of un-ionized indomethacin in the medium.

The dipyridamole flux data show the impact of buffer species and pH on the dissolution behavior of a basic drug with low intrinsic solubility. At fasting state pHs (pH 6.8 or 6.5), the concentration of acidic species (H^+, $H_2PO_4^-$, or H_2CO_3) in the buffers tested is so low that dissolution is not accelerated by protonation of the free base (B) to produce the conjugate acid (BH^+), as shown in Eq. (2).

$$B + H^+ \leftrightarrow BH^+ \quad (2)$$

At pH 6.8 or 6.5, dipyridamole is soluble enough to maintain a high pH at the dissolving solid surface so that there is no significant ionization of the basic drug in the boundary layer and, hence, no increase in dissolution regardless of buffer concentration or species (see Fig. 4). The concentration of CO_2(g) in the sparging gas has no impact, and similarly, the presence or absence of $H_2PO_4^-$ has no effect on dipyridamole dissolution. There is some small increase in intrinsic dissolution when FaSSIF is used as dissolution medium. This is likely related to the solubilizing effect of taurocholate and lecithin (30).

The dissolution data obtained for dipyridamole in the fed state (pH 5) are notable (see Fig. 5). At pH 5, the dissolution medium is acidic enough that protonation of the basic drug accelerates dissolution. The dissolution rate at pH 5 without buffer species present is twice the rate determined at pH 6.8. Given the solubility of dipyridamole, the dissolved dipyridamole at the solid surface cannot maintain as basic a pH in the boundary layer at pH 5. Addition of more acidic species, from the buffer species (HOAc and H_2CO_3) in the dissolution media, significantly impacts dissolution. Even though the equilibrium concentrations of carbonic acid and bicarbonate at pH 5 are low (<1 mM) at all sparging CO_2(g) concentrations, there is still a significant impact of sparging on the dissolution rate. Flux results using 20 %atm CO_2(g) sparged media are comparable to 144 mM acetic acid. Results for dissolution in

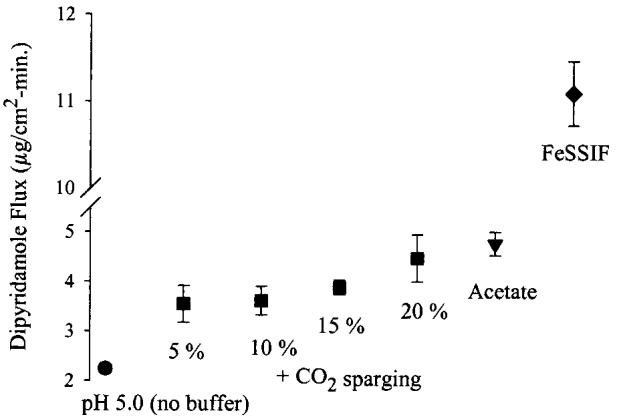


Fig. 5. Dipyridamole flux at pH 5.0, 0.9% NaCl, 37°C 100 rpm. (●) pH 5.0, no buffer, (■) pH 5.0 plus CO_2 sparging, (▼) 144 mM acetic acid, (♦) FeSSIF, pH 5.0, 144 mM acetic acid, 15 mM Na taurocholate, 3.75 mM lecithin, 204 mM KCl. Error bars represent ±1 standard deviation.

FeSSIF (144 mM acetic acid plus taurocholate and lecithin) also show an increased rate from the solubilizing effect of taurocholate and lecithin (30).

As further evidence that a bicarbonate buffer system has been established *in situ* in the dissolution medium by sparging with carbon dioxide gas, samples from the dissolution vessel were titrated with NaOH, and buffer capacities were determined. Average relative error (|exp. – theoretical|/exp. × 100) calculated for low (5% CO_2 atm) and high (20% CO_2 atm) content sparging experiments was less than 18%. The experimental buffer capacity data agree well with theoretical predictions calculated using standard physical constants for the bicarbonate system at 25°C (8).

DISCUSSION

Pharmaceutical scientists have long known that dissolution of ionizable drugs, specifically acidic drugs, can be changed by addition of basic species to the dissolution medium. Given the continued interest in how to mimic *in vivo* dissolution using biorelevant dissolution media, experimentalists should consider how easily a stable bicarbonate buffer (true physiologic buffer system) can be established in aqueous solutions with relatively simple gas sparging equipment (frits, flow meters). In this report, we have shown that sparging with gases of varying carbon dioxide concentrations can be used to establish physiologic bicarbonate buffer systems that influence the dissolution flux of acidic or basic drugs with low intrinsic solubilities such as indomethacin or dipyridamole. Both compounds studied are examples of class II BCS (low solubility, high permeability) type drugs. According to the BCS, one would expect that an *in vitro–in vivo* correlation may exist for these compounds if the *in vitro* dissolution rate, which has been shown to be strongly influenced by pH and buffer composition, is comparable to the *in vivo* dissolution rate (1).

The bicarbonate buffers featured in this report are stable and equilibrate relatively quickly (30–40 min) with stirring, using fluid volumes and gas sparging flow rates that are amenable to normal laboratory practice. A 30- to 40-min time scale is consistent with reports in the literature to establish bicarbonate equilibria for well-stirred aqueous systems (31). Recent advances in fiberoptic probe technology, which have made rugged responsive probes designed specifically for monitoring dissolved CO_2(aq) commercially available, have made this process easier to monitor and confirm.

Our data show that indomethacin dissolution in pH-controlled buffers such as SIF yield dissolution fluxes that are 20 times higher than what can be expected at the same physiologic pH (pH 6.8). Dissolution flux for indomethacin determined in SIF may overestimate what occurs *in vivo* in a bicarbonate buffer system because of the high concentration of phosphate that is specified in SIF. Even though the phosphate concentration is lower in FaSSIF, as compared to SIF, and the pH is slightly reduced, the dissolution flux of indomethacin in FaSSIF is still higher than the results generated in any of the bicarbonate buffers examined in these studies. Though the pH of SIF and FaSSIF may mimic the pH of physiologic intestinal fluids, the buffer composition and concentrations have impacts on dissolution of ionizable compounds that may not be physiologic.

Dissolution results generated using a basic drug such as dipyridamole show that at pH values consistent with the fasted state (pH 6.8 or 6.5), there is no impact of dissolution medium buffer composition. These results are consistent with theoretical expectations, as no significant ionization of the basic drug (pK_a 6.05) occurs under fasted-state pH conditions in lightly buffered media. Dipyridamole is soluble enough to self-buffer the boundary layer at bulk pH 6.8 under these conditions. However, the dipyridamole dissolution results at pH 5 representing the fed state are remarkable and demonstrate a unique advantage of employing a bicarbonate buffer created by sparging. Fig. 6 shows the bicarbonate concentrations that theoretically are in equilibrium with aqueous solutions at 35°C over a range of pH at several gas sparging pressures (P_{CO_2}s). At fed-state pH (pH 5), less than 1 mM bicarbonate equilibrates in the dissolution medium no matter how high the P_{CO_2} is in the sparging gas. However, continuous sparging of the dissolution medium provides a continuous source of bicarbonate and carbonic acid, so that even though the equilibrium concentration of bicarbonate is low, and the bicarbonate and carbonic acid are consumed by dissolution of dipyridamole, more bicarbonate species are always being supplied via the sparging gas. The dynamic behavior of continuous replacement of bicarbonate demonstrates a unique advantage of employing a physiologic bicarbonate buffer via sparging. It would take considerable experimental development to duplicate the dynamic nature of sparged bicarbonate buffers with acetate or phosphate buffers. Even though the concentration of bicarbonate species in equilibrium at pH 5 in 37°C with 5 to 20 CO_2(g) %atm sparging is of the range of 0.1–0.4 mM, there is a significant impact of gas sparging on dissolution flux that is concentration dependent (Fig. 5). These results suggest that high P_{CO_2}s reported postprandially in man [37 CO_2 %atm (14)] may significantly impact dissolution of basic drugs even though the acidic pHs of fed-state luminal fluids are consistent with bicarbonate concentrations that are low (<1 mM).

These preliminary investigations have demonstrated that sparging with carbon dioxide is a viable method to establish bicarbonate buffers for use in dissolution characterization in a USP dissolution vessel. We have taken advantage of a new commercially available fiberoptic probe to monitor dissolved CO_2(aq) during preparation of the dissolution media. We have further corroborated the *in situ* bicarbonate buffer formation by measuring buffer capacities of samples of the dissolution media after dissolution has been completed. One unique advantage to employing gas sparging to create bicarbonate buffers to mimic *in vivo* luminal fluids is that sparging

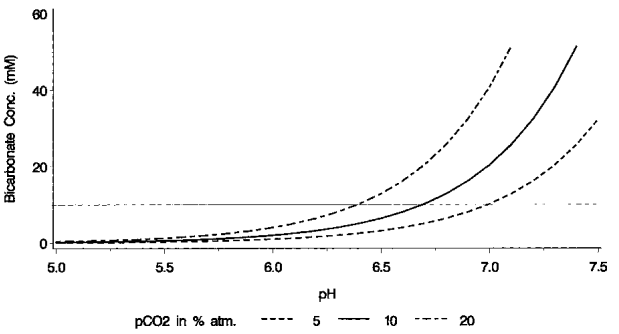


Fig. 6. Bicarbonate concentration with pH and P_{CO_2} at 35°C. Calculated using constants from Butler (8).

serves to continuously supply bicarbonate ions even at pHs where the solubility of bicarbonate is characteristically low. This may have implications for defining buffer effects on dissolution of ionizable drugs in fed-state conditions.

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