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Research Paper

Estimation of Intragastric Solubility of Drugs: In What Medium?

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Purpose. To measure the solubility of four drugs in human gastric aspirates, canine gastric aspirates (CGF) and simulated gastric fluids in order to propose a medium for estimating intragastric drug solubility relevant to a bioavailability study in the fasted state.
Materials and Methods. Intragastric environment after administration of water to healthy fasted adults and to healthy fasted dogs (this study) was initially characterized. Solubilities were then measured with the shake-flask method in gastric fluid aspirated after the administration of water to healthy fasted adults and to healthy fasted dogs, in various simulated gastric fluids, i.e. SGF_{SLS}, SGF_{Triton}, FaSSGF, $FaSSGF_{NaCl}$, and in various HCl solutions with pH values ranging from 1.2 to 2.9.
Results. In all cases, FaSSGF performed better than canine aspirates, SGF_{SLS}, SGF_{Triton}, or $FaSSGF_{NaCl}$ in predicting solubility in HGF. However, its superiority over HCl pH 1.6 was not clear. For ketoconazole, dipyridamole, miconazole, and felodipine deviations of solubility data in FaSSGF from solubility data in HGF were non-significant, 34, −39 and 252%, respectively, whereas the corresponding deviations of data in HCl pH 1.6 from data in HGF were non-significant, 24, 70, and 130%, respectively.
Conclusions. Combining data in FaSSGF and HCl pH 1.6 is comparatively the most efficient way to get an estimate of drug solubility in the fasting gastric contents during a bioavailability study. However, accurate estimation of intragastric solubility is limited by the changing environment during intragastric residence of solid particles and the degree of simulation of intragastric composition.

KEY WORDS: canine gastric contents; dipyridamole; felodipine; human gastric contents; ketoconazole; miconazole; simulated gastric fluid; solubility.

INTRODUCTION

Intragastric drug concentrations are crucial for compounds acting in the stomach whereas they determine the potential for precipitation of a highly dosed weak base upon entering the upper small intestine. Also, since net water flux is minimal in the proximal fasted small intestine (e.g. 1), intragastric concentrations should in many cases reflect intraduodenal concentrations and, therefore, they should provide a basis for deciding what concentrations to use in *in vitro* or *in situ* intestinal permeability studies. Direct assessment of intragastric concentrations in humans using imaging techniques are costly and still in their early infancy (2) whereas techniques involving direct sampling could affect the data and are limited by high costs and ethical issues.

Drug concentrations in the gastric contents depend on the administered dose or, in case of highly dosed compounds, the intragastric solubility and, also, on the intragastric dissolution kinetics (e.g. 3). The use of human aspirates in the *in vitro* estimation of intragastric solubility or dissolution kinetics is limited by high costs and ethical issues. Although, various media have been proposed for simulating the fasting physicochemical characteristics and/or intragastric composition (4–9), literature data on the usefulness of these media are very limited. The only published solubility data in human gastric fluids refer to danazol (a non-ionizable compound) and the difference of solubility data in HCl pH 1.2 from data in human gastric aspirates was −70% whereas the difference of solubility data in HCl pH 1.2 containing 0.25% (w/v) sodium lauryl sulfate and 34.2 mM NaCl [SGF_{SLS}, (4)] from data in human aspirates was 6,500% (10). In line with these findings, it was recently reported that media containing non-physiologically relevant surfactants may lead to high overestimation of intragastric dissolution rates (9). In contrast, a physiologically relevant medium appeared to be much more useful in predicting the amounts of a lipophilic weak base absorbed after oral administration (9).

The purpose of the present study was to measure the solubility of model drugs in human gastric aspirates, canine gastric aspirates and various simulated gastric fluids in order to propose a medium for estimating intragastric drug solubility. If an immediate release tablet is administered with a glass of water (~250 ml) solid particles of disintegrated dosage forms are expected to empty with rates similar to that

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Table I. Physicochemical Characteristics and Single Doses of the Compounds Used in this Study

	Ketoconazole (e.g. 12,13)	Dipyridamole (e.g. 12–14)	Miconazole (13,15)	Felodipine (13,16)
Molecular weight	531.4	504.6	416.1	384.3
ClogP	4.45	2.74, 4.90	6.25	4.46
PKa	2.94 and 6.51 (both alkaline)	5.7–6.4 (alkaline)	6.7 (alkaline)	Not applicable
Aqueous solubility (μg/ml, 37°C)	6.9 (pH 6.5, phosphate buffer)	5 (pH 7, phosphate buffer)	<1.03 (water)	1 (water)
Dose (mg)	200–400	300–600	250	5–10

of the co-administered water (11). Therefore, initially, the intragastric environments of humans (1) and dogs (this study) were characterized. Then, solubility experiments were performed in aspirates collected under conditions that guaranteed the presence of part of the orally administered water in the stomach and data were compared with those collected in various simulated gastric fluids. Since intragastric dissolution can be crucial for the absorption of lipophilic weakly alkaline compounds (9), the present study was performed using three lipophilic weak bases, i.e. ketoconazole, dipyridamole and miconazole and one lipophilic non-ionizable compound, i.e. felodipine (Table I). In order to facilitate the discussion of the data, the pH-solubility profiles of the three bases at biorelevant pH values were also constructed.

MATERIALS AND METHODS

Materials

Ketoconazole was from Recordati Espana S.L. (lot # 03000051, Beniel, Spain). Dipyridamole was from Boehringer Ingelheim GmbH (lot # 02116, Ingelheim, Germany). Miconazole was from Janssen Pharmaceutical (Cork, Ireland, lot# 0310002753) and felodipine was from AstraZeneca (Mölndal, Sweeden, lot # 85-01). Pepsin from porcine stomach mucosa (0.064 mg pepsin/mg solid), taurocholic acid (sodium salt, >99% pure), Triton X-100 and sodium lauryl sulfate were from Sigma Chemical (St-Louis, USA). Egg- lecithin (Lipoid E PC, >98% phosphatidylcholine) was from Lipoid GmbH (Ludwigshafen, Germany). Methanol and acetonitrile of HPLC grade were from E. Merck (Darmstadt, Germany). Water purified with Labconco water pro ps system (Kansas City, Missouri, USA) was used in all procedures. All other chemicals were of analytical grade.

Characterization of the Human and Canine Fasting Gastric Contents

The human gastric contents after administration of 250 ml of water have been recently characterized (1).

Canine gastric contents were characterized using four healthy female mongrel dogs (28–32 kg) that are accommodated in an animal facility operating according to the European Union regulations for the maintenance and experimentation on animals and has been approved by the Veterinary Directorate of the Municipality of Athens. Dogs were fasted from the afternoon prior to the experimental day. As in humans (1), the objective was to assess the canine gastric environment as a function of time after water administration. Knowing that gastric emptying rates in fasted dogs state are faster than in fasted humans (17) and in order to be able to aspirate gastric contents, on the experimental day, each dog was administered 400 ml of water containing 10 mg/ml PEG 4000 (non-absorbable marker) using a sterile disposable tube (Levin #14). At 5, 10, and 20 min after water administration, the tube was again inserted into the dog's stomach and aspiration of gastric contents was performed manually. No medication was given to the dogs prior to or during the aspiration period.

Solubility Media

The human gastric fluid, in which solubility measurements were performed, was created from samples collected for the characterization of the human gastric contents in the fasted state (1) and kept at −70°C. Assuming that during a bioavailability study the particles of an immediate release dosage form will empty from the stomach together with the co-administered water (11) and in order to aspirate contents that reflect the average gastric composition, samples aspirated between 20 and 40 min post water administration were used. On the day of the solubility experiment, samples were brought to room temperature and pooled so that from each individual a total of two samples were obtained and each sample had a volume of approximately 3 ml. The pooled sample (HGF), after centrifuging at a low speed to remove possible small solid particles, was used immediately for the solubility experiments of all drugs tested in this study. The pH of HGF was 1.8.

The procedure that was followed for the characterization of the canine gastric fluid (please see previous section) was applied again to three healthy female mongrel dogs (27–29 kg). Assuming that during a bioavailability study the particles of an immediate release dosage form will empty from the stomach together with the co-administered water (11) and in order to aspirate contents that reflect the average gastric composition, 10 min after the administration of 400 ml water, about 25 ml of gastric contents were aspirated from each dog and kept at −70°C. On the day of the solubility experiment, the three samples were brought to room temperature and pooled. The pooled sample (CGF), after centrifuging at a low speed, was used immediately for the solubility experiments of all drugs tested in this study. The pH of CGF was 2.9.

Four media that simulate the gastric fluids were prepared on the day of the solubility experiments. Simulated gastric fluid containing sodium lauryl sulfate (SGF_{SLS}) consisted of HCl pH 1.2 containing 0.25% (w/v) sodium lauryl sulfate and 34.2 mM NaCl (4). Simulated gastric fluid containing Triton (SGF_{Triton}) consisted of HCl pH 1.2

Table II. The Chromatographic Conditions Used for the Analysis of Drugs in the Present Study

Chromatographic conditions	Ketoconazole	Dipyridamole	Miconazole	Felodipine
Column	BDS RP-C_{18} (250×4.6 mm, 5 μm)	ODS RP-C_{18} (250×4.6 mm, 5 μm)	BDS RP-C_{18} (150×4.6 mm, 5 μm)	BDS RP-C_{18} (150×4.6 mm, 5 μm)
Mobile phase v/v/v	methanol:water: diethylamine 74:26:0.1	acetonitrile:water: diethylamine 50:50:0.2	acetonitrile:methanol: buffer phosphate (pH 7) 36:36:28	acetonitrile:methanol: water 40:20:40
Flow rate (ml/min)	1	1	2	1.5
Detection wavelength (nm)	240	280	230	362
Injection volume (μl)	20	20	50	50

containing 0.1% (w/v) Triton X-100 and 34.2 mM NaCl (5). Fasted state simulating gastric fluid (FaSSGF) consisted of HCl pH 1.6 containing 0.1 mg/ml pepsin, 80 μM sodium taurocholate, 20 μM phosphatidylcholine, and 34.2 mM NaCl (9). $FaSSGF_{NaCl}$ differed from FaSSGF only in that it contained 68 mM NaCl [to reflect the actual intragastric chloride levels (e.g. 18)].

Solubilities of the three weak bases were also measured in simple HCl solutions having pH values of 1.2, 1.4, 1.6, 1.8, 2.4, and 2.9 in order to assess the pH-solubility profile of these compounds at biorelevant pH values.

The resistance of HGF, CGF and simulated gastric fluids to pH increase by the dropwise addition of 0.01 M NaOH was tested in comparison with the resistance of simple HCl solutions having identical pH values.

Solubility Studies

Solubilities were measured in triplicate using the shake-flask method. Six milliliters of the medium and pure drug powder in excess (1 g for ketoconazole, 400 mg for dipyridamole, 50 mg for miconazole, and 10 mg for felodipine) were transferred into Erlenmeyer flasks (ca: 25 ml). Flasks were then covered with parafilm and put in a shaking water bath (160 vibrations/min, 37°C). Equilibration times were measured in SGF_{SLS}, SGF_{Triton} and FaSSGF. Samples were drawn at 2, 4, 6, 8, and 24 h after the initiation of shaking. All samples were immediately filtered with regenerated cellulose filters (17 mm, 0.45 μm Titan®, Wilmington, USA), after discarding the first 0.2 ml. After measuring its pH, the filtrate was subjected to HPLC assay.

Since HGF contained PEG, solubility of all drugs was additionally tested in HCl (pH 1.8) in presence of 5 mg/ml PEG (1,12).

Analysis of Samples

The analytical procedures used for the assay of PEG, pepsin, and total bile salt content as well as the procedures for measuring pH, buffer capacity, osmolality and surface tension of the canine fasting gastric contents were identical to those used for the characterization of the human gastric contents (1).

All four drugs were assayed with HPLC-UV methods that were based in previously published methods (16,19–21). The chromatographic conditions are presented in Table II. For all drugs, quantification of solubility data was made according to standard curves prepared in the corresponding solubility medium.

Apart from ketoconazole in SGF_{SLS}, all drugs were stable in all solubility media. Results of a separate study, in which ketoconazole's solutions (50 μg/ml) in SGF_{SLS}, in SGF_{Triton} and in FaSSGF were incubated at 37°C, revealed that in SGF_{SLS}, unlike in the other two media, ketoconazole's concentration decreases with time according to zero-order kinetics. The half-life of the process was estimated to be 38.4±5.9 h (data not shown). The decrease could be related to the products of hydrolysis of SLS in highly acidic solutions (22) and/or the formation of salt(s) with varying dissolution/crystallization rates (23).

Data Analysis

The statistical significance of a difference between two solubility data sets was assessed with unpaired *t* test at the 0.05 level using SigmaStat® (Sigma-Stat® 2.03, SPSS INC., USA).

The percent difference of solubility in a medium from solubility in HGF was estimated with the following formula:

$$\%\ \text{difference} = \left[\frac{(\text{solubility in a medium}) - (\text{solubility in HGF})}{\text{solubility in HGF}}\right] \times 100$$

RESULTS

Physicochemical Characteristics of the Canine Fasting Gastric Contents After Administration of Water

The physicochemical characteristics of the canine fasting gastric contents estimated in four dogs at various time points after the administration of 400 ml of water are presented in Table III.

Median PEG concentration 5–10 min after administration of water indicates that more than 40% of the contents consist of secretions (Table III). At 20 min after water administration the concentration was lower than the limit quantification of the method (3.3 mg/ml) in all dogs. Since basal gastric output is low in dogs (17), the low PEG concentrations immediately after administration of water should be primarily related to the fast (compared to humans) gastric emptying of non-caloric liquids in the fasted dog (17).

Table III. Median [Range] Values of Various Physicochemical Parameters of the Canine Gastric Contents After Administration of 400 ml of Water to 4 Fasted Mongrel Dogs[a]

Time (min) after administration	PEG (mg/ml)	pH	Buffer capacity (mmol/l/ΔpH)	Pepsin (mg/ml)	Osmolality (mOsmol/kg)	Surface tension (mN/m)	Total bile salt content (mM)
5	5.7 [5.3–6.7]	6.3 [3.2–7.2]	4.0 [0.6–6.6]	<0.01	25 [23–50]	37.3 [33.3–43.3]	<0.5
10	5.8 [4.2–6.2]	2.4 [1.8–6.6]	4.6 [3.7–18]	0.09 [0.04–0.10]	77 [48–88]	36.0 [34.2–41.6]	<0.5
20	<3.3	1.9 [1.9–2.3]	14 [1.0–14]	<0.01	NM	NM	<0.5

[a] When the values were lower than the limit of quantification, data are presented as "< limit of quantification." NM means Not Measured.

At 10 min after water administration, when most of the administered water should have been emptied from the stomach [(24) and according to PEG data above], median pH value was 2.4 (Table III), i.e. higher than the value observed in the fasted human stomach at time when most of the water had been emptied from the stomach [40 min after administration of water, median pH value was 1.7 (1)]. In dogs, fluctuations in intragastric pH have been observed and values are thought to be higher than that of humans (17).

Compared to human data [where pepsin increases from about 0.1 to about 0.2 mg/ml after emptying of most of the administered water (1)], pepsin levels in the canine stomach are practically non-existent (Table III).

Osmolality was about half of that in the human stomach (1).

Interestingly, surface tension of the canine gastric contents was lower than that of humans (1) indicating the presence of more surface active agents than in humans. However, whether or not this relates to the higher bile salt content could not be confirmed due to the high limit of quantification of analytical method used in this study (Table III).

Equilibration Times in Solubility Experiments

For ketoconazole, dipyridamole and felodipine, equilibration times were found to be less than 4 h in all cases. Therefore, any effect of SLS on ketoconazole's concentration (please see "Materials and Methods", "Analysis of Samples") should be minimal.

With miconazole a supersaturated solution was formed at 2 h in SGF_{SLS}, in HCl pH 1.2, and in HCl pH 1.4 leading to the dissolution of almost all of the solid miconazole in the flask. However, at 4 h the presence of the solid in the flask

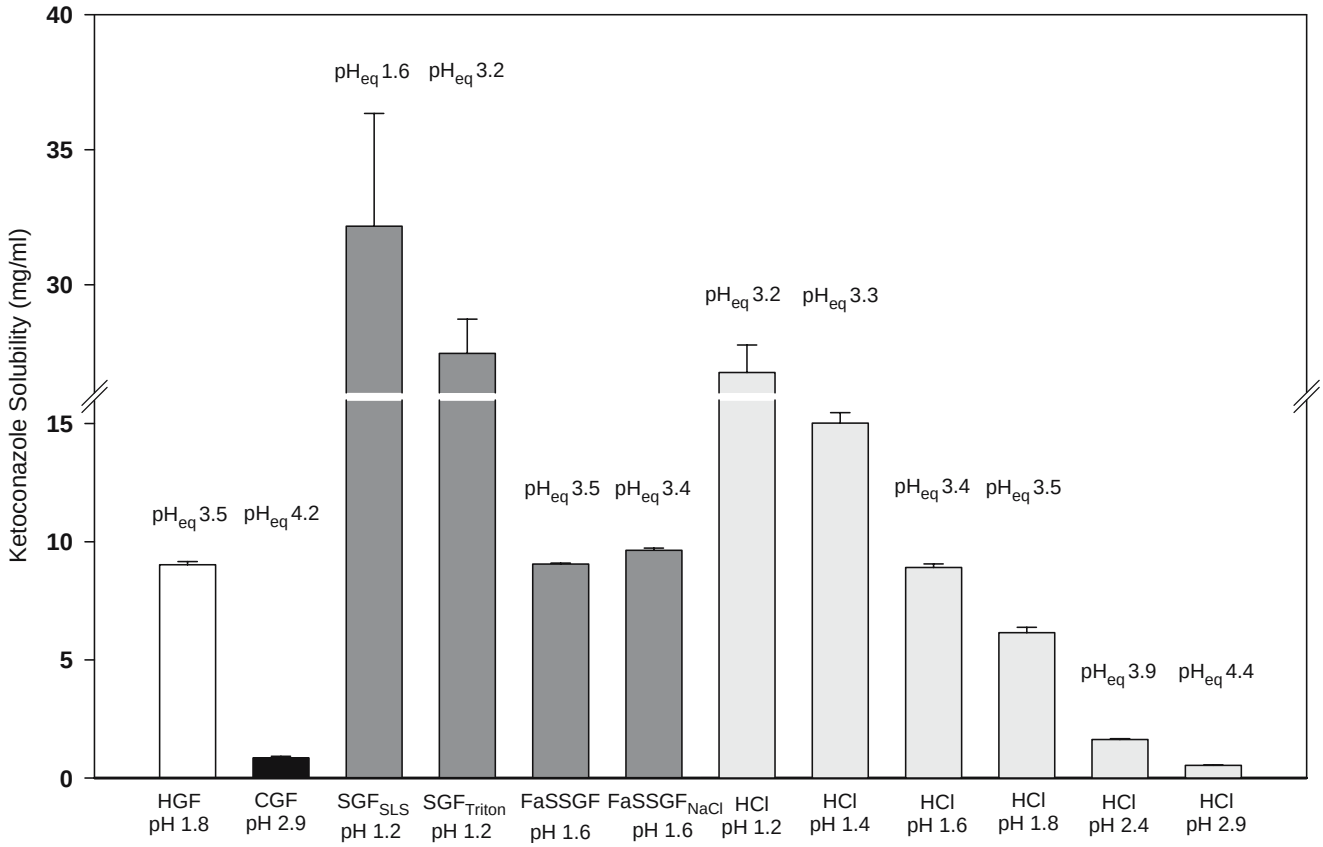

Fig. 1. Mean+SD solubility data of ketoconazole in human gastric fluid (HGF), in canine gastric fluid (CGF), in various simulated gastric fluids, i.e. SGF_{SLS}, SGF_{Triton}, FaSSGF, and $FaSSGF_{NaCl}$, and in HCl solutions with initial pH values 1.2, 1.4, 1.6, 1.8, 2.4 and 2.9. Abbreviations are explained in the text. pH_{eq} is the pH of the medium at equilibrium.

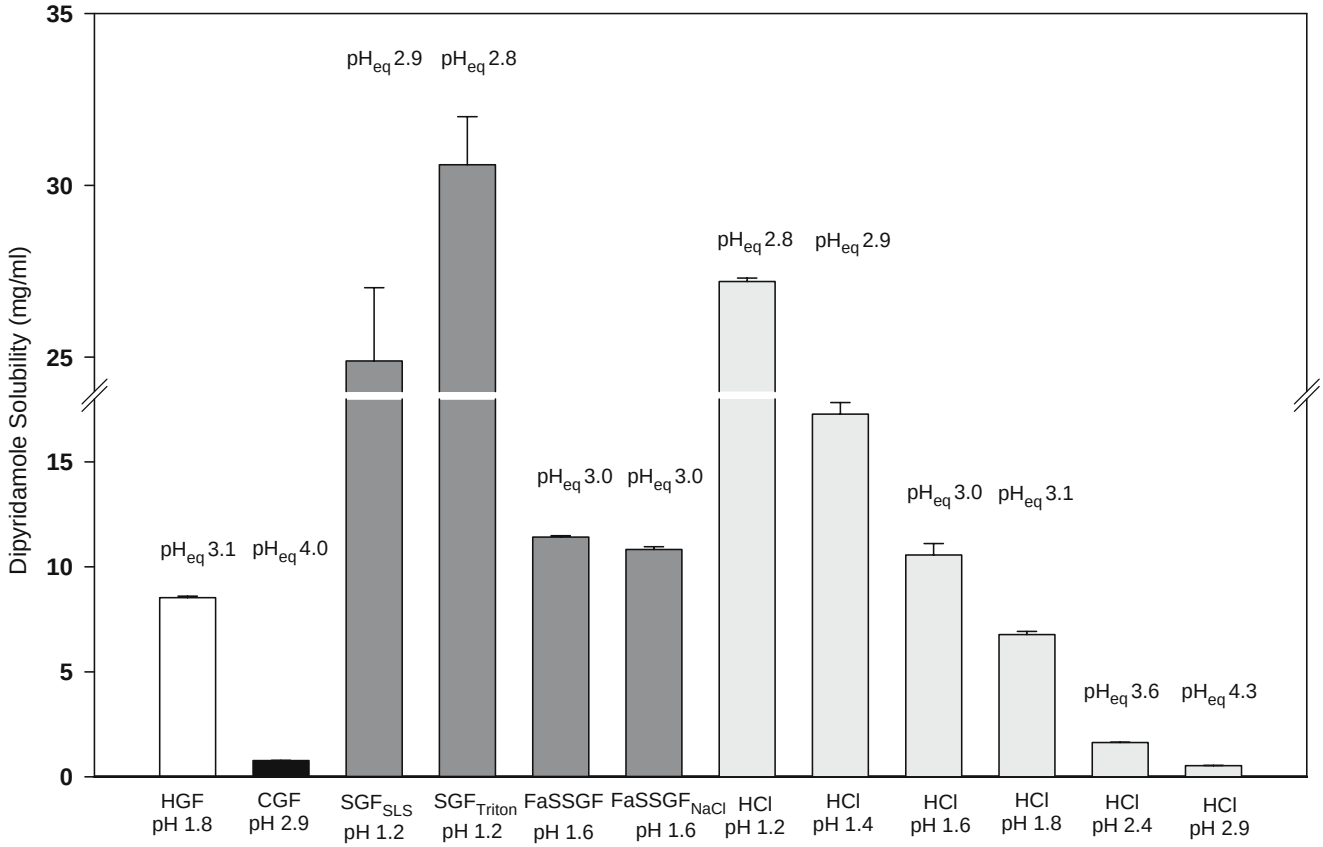

Fig. 2. Mean+SD solubility data of dipyridamole in human gastric fluid (HGF), in canine gastric fluid (CGF), in various simulated gastric fluids, i.e. SGF_{SLS}, SGF_{Triton}, FaSSGF, and $FaSSGF_{NaCl}$, and in HCl solutions with initial pH values 1.2, 1.4, 1.6, 1.8, 2.4 and 2.9. Abbreviations are explained in the text. pH_{eq} is the pH of the medium at equilibrium.

increased, the concentration decreased and remained constant at least up to 24 h. For example, at 2 h in SGF_{SLS} the concentration was 0.46±0.18 mg/ml and the solubility value at 4 h (and up to 24 h) decreased to 0.29±0.02 mg/ml. Miconazole is known to form polymorphs (25,26); the melting point of solid miconazole in SGF_{SLS} at 2 h was not distinct (63–81°C) whereas the melting point of the powder used in solubility studies was 95°C. Temporary formation of supersaturated solutions of carbamazepine (27) and testosterone (28) in presence of SLS has also been reported.

Based on the above, 4 h were considered adequate for the solubility experiments of all drugs in all media.

PEG Effects on Solubility Data

Solubility of felodipine in HCl pH 1.8 was increased from 0.98 μg/ml to 1.4 μg/ml, i.e. by 42%, when 5 mg/ml PEG 4000 was added in the solution. Solubility data of all other compounds were not affected by the presence of PEG.

Solubility Data

Solubility data are graphically presented in Figs. 1, 2, 3, 4.

Media containing ionized compounds (Figs. 1, 2, 3) at equilibrium had higher than the initial pH. Fig. 5 shows that the resistance of each medium to pH increase by the dropwise addition of 0.01 M NaOH is, at least partly, related to the acidity of the medium. The total concentration of sodium hydroxide in a medium that resulted in increasing the pH of the medium by one unit was 57–74 mM in SGF_{SLS}, SGF_{Triton} and HCl pH 1.2, 23–25 mM in FaSSGF and HCl pH 1.6, 14 mM in HGF and HCl pH 1.8, and 1.2 mM in CGF and HCl pH 2.9 (Fig. 5). However, since the three weak bases have similar pKa values and molecular weights (Table I), the degree of pH increase is also a function of the concentration of the dissolved ionized species; indeed, in all solubility media, at equilibrium the pH increased more in the ketoconazole or dipyridamole experiments than in the miconazole experiments (Figs. 1 and 2 vs. Fig. 3).

The difference of the solubility data in CGF from the solubility data in HGF was −91, −91, −56 and 142% for ketoconazole, dipyridamole, miconazole, and felodipine, respectively (Figs. 1, 2, 3, 4, $p \leq 0.001$ in all cases). For the three ionized compounds relevant differences are partly attributable to the pH difference of the two media whereas for felodipine the difference could be partly attributed to the presence of ions with salting out properties (29) in HGF.

The difference of the solubility data in SGF_{SLS} from the solubility data in HGF was 256, 192, −73, and 24,204% for ketoconazole, dipyridamole, miconazole and felodipine, respectively (Figs. 1, 2, 3, 4, $p<0.001$ in all cases). The corresponding numbers for the data in SGF_{Triton} were 204, 259, −57, and 5,659%, respectively (Figs. 1, 2, 3, 4, $p<0.001$ in all cases). Ketoconazole, dipyridamole and felodipine data are in qualitative agreement with previous data of danazol in SGF_{SLS} (10) and confirm that SGF_{SLS} and

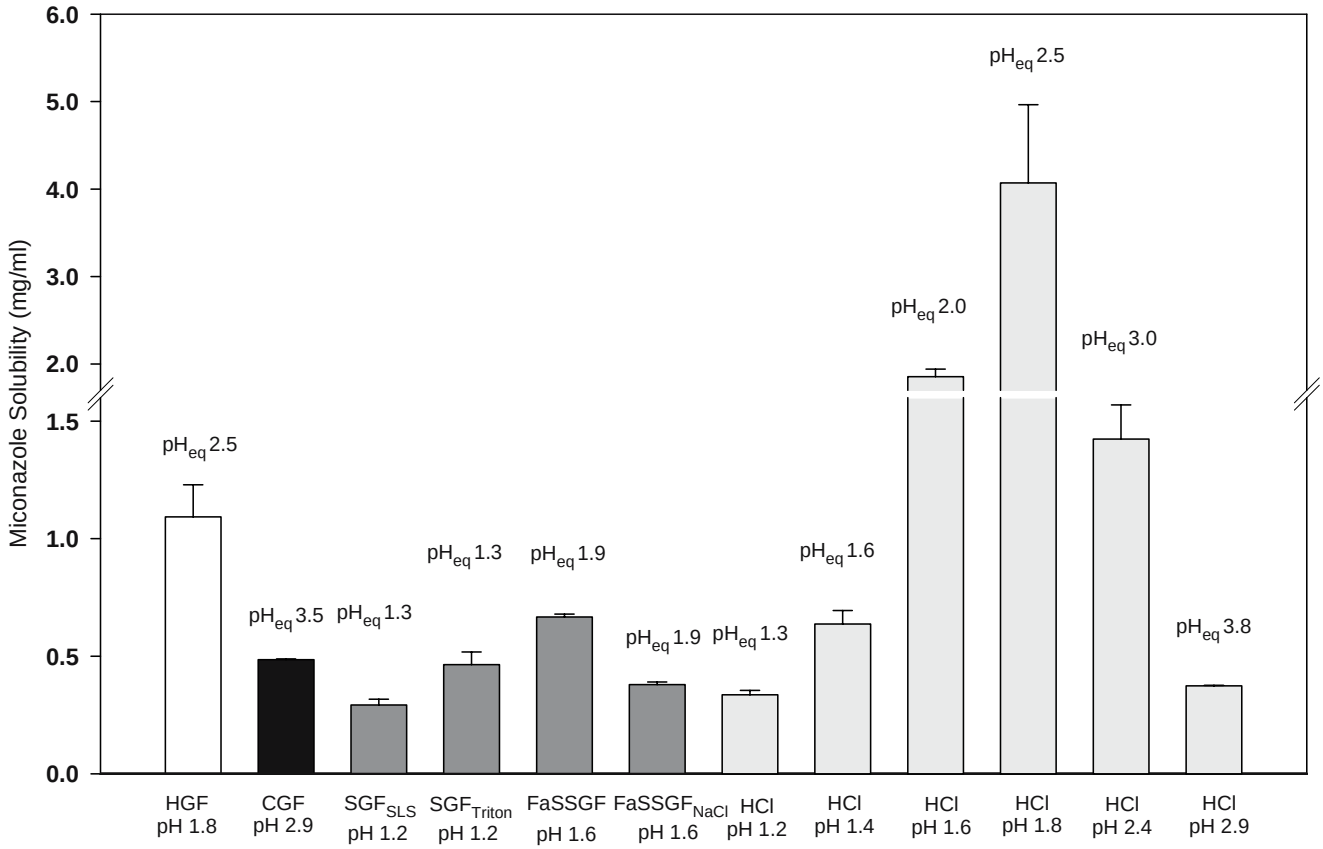

Fig. 3. Mean+SD solubility data of miconazole in human gastric fluid (HGF), in canine gastric fluid (CGF), in various simulated gastric fluids, i.e. SGF_{SLS}, SGF_{Triton}, FaSSGF, and $FaSSGF_{NaCl}$, and in HCl solutions with initial pH values 1.2, 1.4, 1.6, 1.8, 2.4 and 2.9. Abbreviations are explained in the text. pH_{eq} is the pH of the medium at equilibrium.

SGF_{Triton} are not appropriate for estimating intragastric solubility due to their low pH and/or the solubilizing effects of SLS [CMC_{SLS}<0.2% (w/v) (e.g. 30)] and Triton X-100 [CMC_{Triton}~0.01% (w/v) (e.g. 31)]. In contrast, miconazole's data in SGF_{SLS} and SLS_{Triton} are lower than data in HGF and indistinguishable from the pH-solubility profile of this weak base (Fig. 3). Based on the pH-solubility profile of miconazole (Fig. 3), the pH of maximum solubility (pH_{max}) is around pH 2.5. Below this pH, the solubility of miconazole is limited by the solubility of its hydrochloride salt and decreases as the pH is lowered due to the common ion effect (e.g. 32). Therefore, any solubilization by SLS or Triton on miconazole is not observable. A pH_{max} was not observed for ketoconazole or dipyridamole and hence their solubility is limited by the unionized form throughout the pH range tested (Figs. 1 and 2 – pH-solubility profiles).

The difference of the solubility data in FaSSGF from the solubility data in HGF was non-significant (p=0.728), 34% (p<0.001), −39% (p=0.006), and 252% (p<0.001) for ketoconazole, dipyridamole, miconazole, and felodipine, respectively (Figs. 1, 2, 3, 4). The corresponding values for the data in $FaSSGF_{NaCl}$ were 7, 27, −65, and 283% (Figs. 1, 2, 3, 4, $p\leq$0.003 in all cases). In hydrochloric acid solutions of a base at pHs where solubility is limited by that of the hydrochloride salt, the solubility product decreases with increasing the concentration of NaCl (common ion effect, e.g. 32). Indeed, the increase of NaCl from 34.2 mM in FaSSGF to 68 mM in $FaSSGF_{NaCl}$ significantly decreased the solubility of miconazole by −43% (p<0.001). This decrease led to worse prediction of solubility in HGF. Solubility of ketoconazole and dipyridamole remained statistically unaffected because their solubility is limited by their unionized forms throughout the pH range tested.

The difference of the solubility data in HCl pH 1.6 from the solubility data in HGF (pH 1.8) was non-significant (p=0.387), 24% (p=0.003), 70% (p=0.001), and 130% (p<0.001) for ketoconazole, dipyridamole, miconazole, and felodipine, respectively (Figs. 1, 2, 3, 4). The difference of the solubility data in HCl pH 1.8 from the solubility data in HGF (pH 1.8) (Fig. 1) was −32, −21, 272, and 155%, for ketoconazole, dipyridamole, miconazole, and felodipine, respectively ($p\leq$0.005 in all cases), i.e. higher than the difference of data in HCl pH 1.6 from data in HGF (pH 1.8). These findings confirm that drug solubility in HGF is not an exclusively pH-dependent parameter.

The solubility of miconazole and felodipine in HGF was overestimated by data in HCl pH 1.6 or in HCl pH 1.8. For miconazole, this could be attributed to the common ion effect (rather than salting out) due to the higher concentration of chloride ions in the HGF. This is supported by the lower solubility observed in the FaSSGF and FaSSGF with higher sodium chloride content. For felodipine, the higher solubility in HCl solutions compared to HGF (despite the solubilizing effect of PEG in HGF) could have been attributed to the salting out effect. However, this mechanism is rather difficult to reconcile with the FaSSGF data which also showed higher

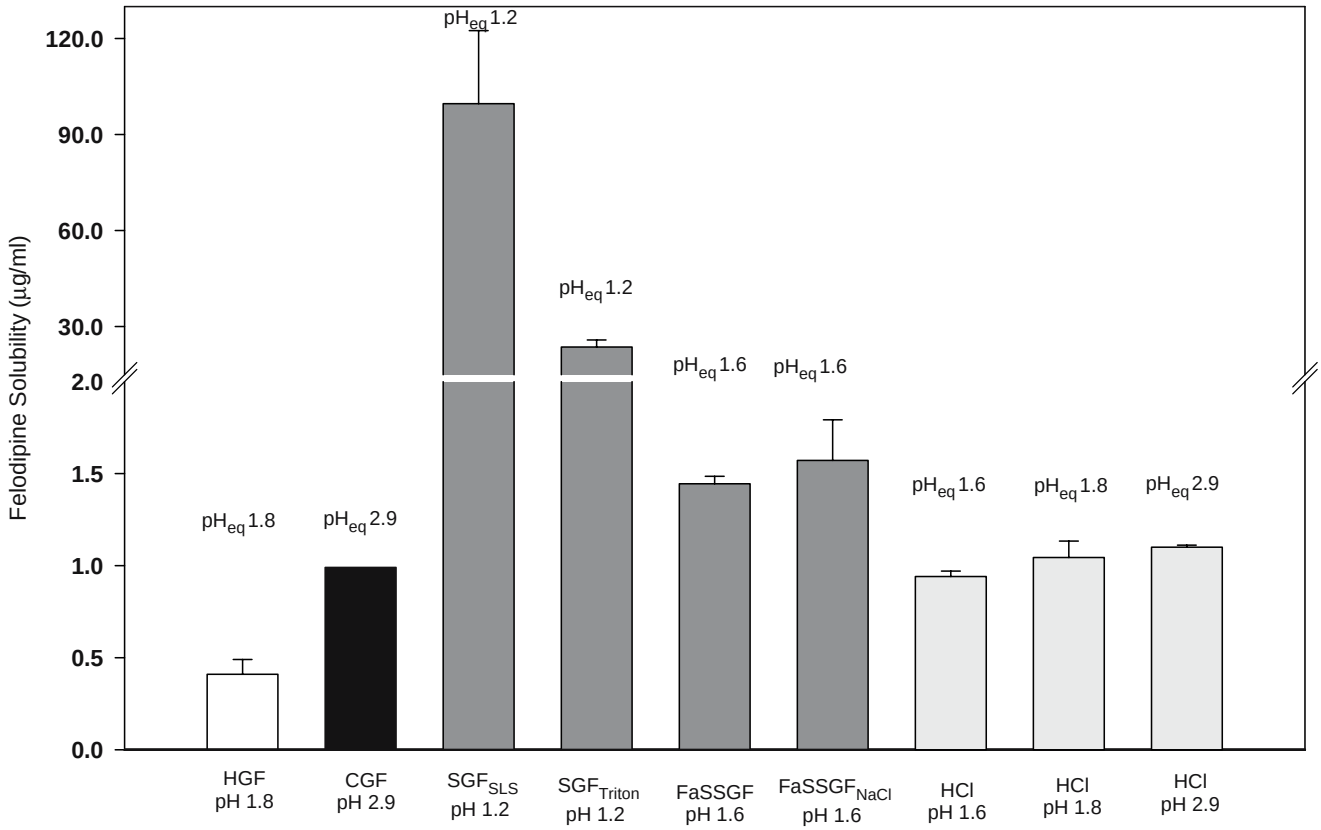

Fig. 4. Mean+SD solubility data of felodipine in human gastric fluid (HGF), in canine gastric fluid (CGF), in various simulated gastric fluids, i.e. SGF_{SLS}, SGF_{Triton}, FaSSGF, and $FaSSGF_{NaCl}$, and in HCl solutions with initial pH values 1.6, 1.8, and 2.9. Abbreviations are explained in the text. pH_{eq} is the pH of the medium at equilibrium.

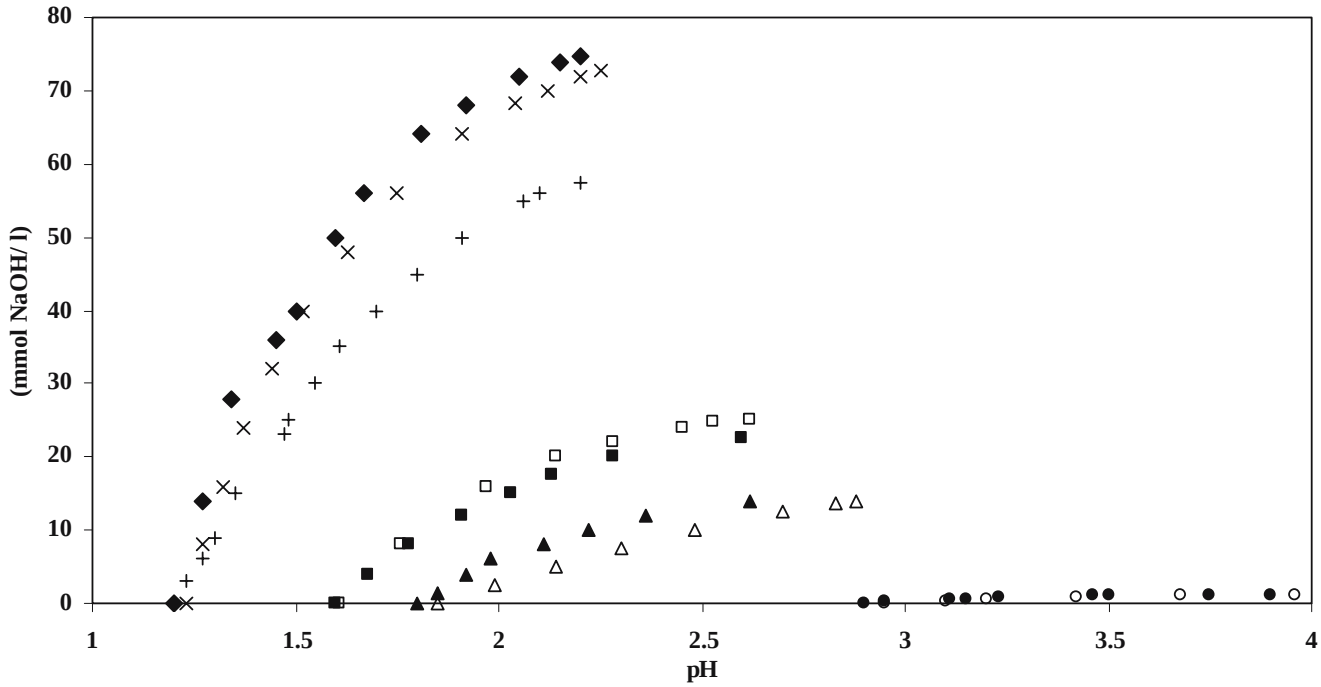

Fig. 5. The pH change of solubility media by the dropwise addition of 0.01M NaOH. Key: (*filled diamond*) SGF_{SLS}; (*multiplication sign*) SGF_{Triton}; (*plus sign*) HCl pH 1.2; (*open square*) FaSSGF; (*filled square*) HCl pH 1.6; (*open triangle*) HGF; (*filled triangle*) HCl pH 1.8; (*open circle*) CGF; (*filled circle*) HCl pH 2.9.

solubility than HGF despite its sodium chloride content. In contrast to felodipine's data of this study, in a previous study (10) the solubility of danazol [non-ionizable, logP 4.5 (10)] in HGF was found to be underestimated (by 70%) by data in HCl pH 1.2. However, in that study, intragastric bile salt concentrations were at levels that could create solubilization effects [~0.82 mM] and higher than values measured in this study (<0.5 mM, Table III). Also, in the relevant publication, no information on the volume of water administered prior to aspirations was given (10).

The difference between solubility data in CGF and solubility data in HCl pH 2.9 was −37, −31, −23 and 11% for ketoconazole, dipyridamole, miconazole, and felodipine, respectively (Figs. 1, 2, 3, 4, $p<0.001$ in all cases). For ketoconazole and dipyridamole these differences are similar to the differences between solubility in HGF and solubility in HCl pH 1.8. In contrast, for the more lipophilic miconazole and felodipine they were smaller than differences between solubility in HGF and solubility in HCl pH 1.8, because, based on Table III, CGF should have lower osmolality, and contain less HCl and pepsin than HGF (1) and, presumably, less salting out ions than HGF.

DISCUSSION

One characteristic of the shake-flask method in estimating solubility of ionized compounds is the possible alteration of the final pH of the medium. However, for highly dosed compounds [i.e. for those compounds for which solubility constitutes a parameter of interest (e.g. 3)], similar alterations may also occur intralumenally, especially in the fasted small intestine where buffering capacity is very low (1).

Depending on the dose, the ratios *Dose To Solubility-in-HGF* of ketoconazole, dipyridamole and miconazole (Figs. 1, 2, 3 and Table I) are 22–44, 35–70, and 229 ml, respectively. Therefore, a dose of miconazole requires about all intragastric contents to dissolve (about 250 ml after the administration of a glass of water) and, since intragastric concentrations are expected to reach values of the order of solubility levels, elevation of gastric pH should be expected (according to data in Fig. 3), i.e. in the fasted stomach, dissolution of a highly dosed base should elevate the intragastric pH. In contrast, the data of ketoconazole and dipyridamole collected in this study may not have physiological relevance for these specific compounds, because the administered doses can dissolve in few milliliters, and, the intragastric pH would not be elevated. However, ketoconazole and dipyridamole data collected in this study are useful for understanding parameters that can affect intragastric solubility of highly dosed weak bases for which HCl salts do not crash out of the solution at biorelevant pHs. Obviously, for felodipine, a highly dosed drug in the fasting gastric environment (*Dose To Solubility-in-HGF*=12,200–24,400 ml, Table I and Fig. 4), pH effects are irrelevant.

During a bioavailability study in the fasting small intestine, unlike in the fasted stomach, the concentration of potential solubilizing species is clearly higher than their critical micellar concentration. This decreases the importance of other components and makes accurate predictions of intraintestinal solubility of lipophilic compounds easier (e.g. 12). In addition, the secretion of numerous substances at minute concentrations in stomach (e.g. 33–35) and incoming saliva, make the design of a medium for estimating intragastric drug solubility harder than the design of simulated intestinal fluids. Accordingly, this study shows that, although combining data in FaSSGF and in HCl pH 1.6 seems to be comparatively the most efficient way to get an estimate of intragastric solubility, estimations may still be not accurate.

Although FaSSGF is not clearly more useful than HCl pH 1.6 for intragastric solubility estimations, dissolution rates may also be dependent on the wetting ability of the particles. In this case, it would be expected that the lower surface tension of FaSSGF would facilitate dissolution of poorly wettable compounds better than HCl pH 1.6. However, due to the small aspirate volumes generated, dissolution rate studies were not conducted.

Finally, it has been reported that in media simulating the contents of the small intestine and/or media containing solubilizing agents with high molecular weight (36,37), dissolution rates may not be proportional to solubility due to the low diffusivity of the vehicle that carries the drug into the bulk solution. Although relevant work still needs to be done, initial data with another weak base suggest that this may not be the case in the media simulating the fasted gastric contents [increased solubilization leads to increased dissolution rates (9)].

CONCLUSION

Compared with data in other frequently used media, solubility data in FaSSGF and/or HCl pH 1.6 provide a better basis for the assessment of intragastric solubility during a bioavailability study in the fasted state. However, accurate estimation of intragastric solubility remains problematic.

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