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Does the Intake of Ethanol Affect Oral Absorption of Poorly Soluble Drugs?

Janneke Keemink [1,*], Erik Sjögren [1,2], René Holm [3], Christel A.S. Bergström [1]

[1] Department of Pharmacy, Uppsala University, BMC P.O. Box 580, SE-751 23 Uppsala, Sweden
[2] Pharmeteus, Dag Hammarskjölds Väg 52B, 752 37 Uppsala, Sweden
[3] Drug Product Development, Janssen R&D, Johnson & Johnson, Turnhoutseweg 30, 2340 Beerse, Belgium

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ABSTRACT

The presence of ethanol in gastrointestinal (GI) fluids may increase the solubility of poorly water-soluble drugs. This suggests that intake of ethanol with such compounds could result in increased drug absorption in the stomach and duodenum because of the greater concentration gradient present. To test this hypothesis, *in vitro* dissolution of 2 poorly soluble compounds (indomethacin and felodipine) was studied in simulated GI rat fluids in the presence or absence of ethanol. Results were used to predict plasma exposure of the compounds using the software PK-Sim. Finally, *in vivo* plasma exposure in rats was investigated after oral dosing followed by immediate administration of water or ethanol. Despite increased solubility in GI fluids in the presence of ethanol, simulations predicted a negligible effect on absorption. This was confirmed in the rat study where oral intake of indomethacin or felodipine with ethanol did not increase *in vivo* plasma exposure. A possible explanation for the lack of an effect may be that dilution, absorption, and transfer of ethanol upon arrival in the stomach resulted in intragastric and intraduodenal ethanol concentrations that did not reach the levels required to affect local solubility.

Introduction

The concomitant intake of ethanol has been shown to affect the bioavailability of several commercially available drug products.[1] Ethanol in the gastrointestinal (GI) tract can result in the premature disintegration of modified-release dosage forms, referred to as "dose dumping," leading to (1) rapid drug dissolution, (2) an increased rate of drug absorption, and (3) higher plasma concentrations. In the case of Pallodone XL™, a once-daily formulation of hydromorphone hydrochloride, the potential for ethanol-induced dose dumping even prompted market withdrawal.[1,2]

Alternatively, ethanol in the GI tract may affect absorption by increasing drug dissolution and solubility in GI fluids, because ethanol can act as a cosolvent. We have previously shown that the presence of 20% ethanol in simulated human gastric and intestinal fluids significantly increased the apparent solubility (S_{app}) of poorly water-soluble compounds; moreover, *in silico* simulations based on these solubility measurements predicted increased plasma concentrations of non-ionizable lipophilic compounds.[3,4] Similarly, increased solubility has been observed when lipophilic drugs are taken together with meals rich in lipids. For the latter, higher plasma concentration have been reached because of the enhanced drug solubility in the GI tract.[5,6]

Despite the fact that increased bioavailability can have serious and negative consequences (especially for compounds with a narrow therapeutic window), limited systematic studies are available on the *in vivo* effects of ethanol. In addition to performing *in vitro* dissolution studies and *in silico* absorption simulations, we evaluated the *in vivo* effect of ethanol as a cosolvent on the intestinal absorption of lipophilic drugs in rats. Felodipine was used as the model drug for which an effect of ethanol was expected. The solubility of felodipine in human gastric and intestinal simulated fluids has previously been reported to increase 14- and 1.9-fold in the presence of 20% v/v ethanol, which could enhance absorption.[3,4] Indomethacin was used as a control drug. Although the solubility of indomethacin in gastric and intestinal simulated fluids

Abbreviations used: AUC, area under the curve; C_{max}, maximum plasma concentration; CYP, cytochrome P450; FaSSIF, fasted state simulated intestinal fluid; FaSSGF, fasted state simulated gastric fluid; GI, gastrointestinal; rFaSSIF, rat fasted state simulated intestinal fluid; S_{app}, apparent solubility; t_{max}, time to reach maximum plasma concentration.

Declaration of interest: none.

* *Correspondence to:* Janneke Keemink (Telephone: +46 18-471 4371).
E-mail address: Janneke.keemink@farmaci.uu.se (J. Keemink).

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increased 13- and 3.5-fold in the presence of 20% v/v ethanol,[3,4] a negligible effect on indomethacin absorption was expected in humans.[4] The reason for this may be that indomethacin (p*Ka* 3.91) gets ionized at the pH of the small intestine, which results in a solubility that allows the maximum dose (100 mg) to become completely dissolved already in the absence of ethanol. Indeed, the dose number for indomethacin (considering solubility in fasted state simulated intestinal fluids [FaSSIF]) was <1.[4,7] This means that at the pH of the small intestine, indomethacin performs as a biopharmaceutics classification system class I compound with high solubility and high permeability.

To exclude processes other than dissolution and solubility, we evaluated the effect of ethanol on oral absorption of felodipine and indomethacin from both drug solutions and suspensions.

Materials and Methods

Materials

Methylcellulose United States Pharmacopeia grade, sodium hydroxide, monobasic sodium phosphate monohydrate, sodium chloride, warfarin, and formic acid were purchased from Sigma-Aldrich (St. Louis, MO). PEG400 was from Fluka (Buchs, Switzerland), ethanol (96%) from De Danske Spritfabrikker (Aalborg, Denmark), and acetonitrile from VWR (Stockholm, Sweden). Felodipine was a kind gift from AstraZeneca (Cambridge, UK). Indomethacin was obtained from Hawkins, Inc. Pharmaceutical Group (Roseville, MN). FaSSIF/fasted state simulated gastric fluid (FaSSGF) powder was purchased from biorelevant.com (London, UK).

Preparation of Formulations

The intravenous formulations were prepared by dissolving the compound in pure ethanol and adding isotonic saline to the ethanol solution to produce a concentration of totally 20% (v/v) ethanol solution. pH was adjusted to 7.0 and the solution was filtrated through a sterile 0.22-μm filter before use. Oral solutions of felodipine and indomethacin were prepared by weighing the compounds and subsequently adding PEG400 to a final concentration of 3.3 mg/mL. After overnight stirring, compounds were completely dissolved. Aqueous suspensions containing 33.3 mg/mL of felodipine or indomethacin were prepared by mixing the compounds with a 0.5% (w/v) methylcellulose solution. The compounds were used as supplied with no further processing other than stirring on a magnetic stirrer.

Particle Size of Suspensions

The particle size of the suspensions was measured by laser diffraction using the Fraunhofer theory on a HELOS from Sympatec GmbH (Clausthal-Zellerfeld, Germany). The suspensions were diluted in purified water immediately before the measurements. The particle size of the indomethacin suspension for D10, D50, and D90 was 12.1 μm, 58.8 μm, and 83.8 μm, respectively, and for the felodipine suspension, 1.5 μm, 11.9 μm, and 39.4 μm, respectively. These particle sizes are expected to result in incomplete absorption in the absence of ethanol.[8,9]

In Vitro *Dissolution*

The shake flask method was used to determine (1) S_{app} of indomethacin and felodipine in rat FaSSIF (rFaSSIF) in the absence or presence of 20% ethanol and (2) dissolution of indomethacin and felodipine solutions and suspension in the rat stomach in the absence or presence of 20% ethanol. rFaSSIF was a modification of FaSSIF-V1 containing higher levels of lecithin (5.25 mM) and taurocholate (21 mM). These higher concentrations resemble phospholipid and bile salt concentrations in intestinal fluids of rats which are relatively high as compared to humans. This is due to that rats lack a gall bladder and continuously secrete bile into the duodenum, while humans secrete bile from the gall bladder in much lower volumes dependent on food intake.[10-12] FaSSGF was prepared as described before.[4] The pH was adjusted to 2.5 to resemble the level in the stomach of a fasted rat.[13]

To determine apparent solubility, an excess of crystalline compound was added to 1 mL rFaSSIF or 1 mL rFaSSIF containing 20% of ethanol. After 24 h on a shaker (300 rpm) at 37°C, test tubes were centrifuged (2300 × *g* at 37°C for 10 min). The supernatant was diluted in mobile phase before HPLC-UV analysis.

In order to determine felodipine and indomethacin dissolution in the stomach of a rat, formulations were added to FaSSGF in a ratio relevant for the *in vivo* situation in fasted rats where gut water represents 1.8% of the total body weight.[14] Therefore, 16.7 μL of the formulations was added per mL of FaSSGF, except for the indomethacin suspension, for which 33.3 μL was added per mL FaSSGF. Vials were incubated on a shaker (300 rpm) at 37°C for 30 min, the gastric-emptying time in fasted rats,[13] before samples were centrifuged (2300 × *g* at 37°C for 10 min) to separate the dissolved and the solid phase. Both phases were diluted in mobile phase before HPLC-UV analysis.

HPLC-UV Analysis

HPLC analysis was conducted using an HPLC (Agilent Technologies 1290 Infinity) with a Zorbax Eclipse XDB-C18 column (4.6 × 100 mm) at 40°C. The injection volume was 20 μL. For the analysis of indomethacin, a mobile phase consisting of 0.1% formic acid in acetonitrile:0.1% formic acid in water 70:30 (v/v) and an isocratic flow rate of 1 mL/min were used. UV absorbance of indomethacin was monitored at a wavelength of 320 nm. The mobile phase used to analyze felodipine consisted of acetonitrile:sodium acetate buffer (pH 5) 80:20 (v/v) and was used at an isocratic flow rate of 1 mL/min. UV absorbance of felodipine was monitored at a wavelength of 360 nm. The retention times were 1.94 min for indomethacin and 1.84 min for felodipine.

In Silico *Absorption Simulations*

The potential impact on plasma exposure of an increase in solubility caused by administration of ethanol was investigated by physiologically based pharmacokinetic modeling using the software PK-Sim (v.7.3.0) included in the Open System Pharmacology Suite (https://github.com/Open-Systems-Pharmacology/Suite/releases/tag/v7.3.0). All simulations were performed using default values of the "rat" as specified in the software, if not stated otherwise. Drug-specific values applied for physicochemical parameters,[4] fraction unbound in plasma,[15,16] and solubility values are summarized in

Table 1
Physicochemical Parameters[4] and Unbound Fractions in Plasma[15,16] Used for *In Silico* Absorption Simulations

Drug-Specific Parameters	Indomethacin	Felodipine
logP	3.5	4.8
$logD_{2.5}$	3.5	4.8
$logD_{6.5}$	1.5	4.8
MW (Da)	357.8	384.3
p*Ka*	3.91	NA
fu, plasma	0.10	0.01

NA, not applicable.
Lipophilicity is expressed as logP, log D at pH 2.5($logD_{2.5}$), and log D at pH 6.5 ($LogD_{6.5}$); MW molecular weight; p*Ka* dissociation constant; $f_{u,plasma}$, fraction unbound in plasma.

Table 2
Effect of 20% of Ethanol on Apparent Solubility in Gastrointestinal Fluids of Rats

Drug-Specific Parameters	Apparent Solubility (μg/mL)	
	Indomethacin	Felodipine
FaSSGF[4]	1.7 ± 0.2[6]	2.3 ± 0.4[6]
$FaSSGF_{20\%\ ethanol}$[4]	22.5 ± 0.5[6]	33.1 ± 3.4[6]
rFaSSIF	933 ± 51	191 ± 21
$rFaSSIF_{20\%\ ethanol}$	1775 ± 172	264 ± 5

Tables 1 and 2. The first step of the modeling strategy was to identify drug disposition. Plasma-concentration time profiles after intravenous administration and the administration of oral water solution, assuming no precipitation after administration, was used to identify distribution (tissue distribution calculation method and logP), gut wall permeability, and unspecific metabolic elimination, in liver and also in the gut wall for felodipine. Intestinal solubility was then identified in each small intestinal segment using observed plasma concentration-time profiles after oral administration of water suspension and applying identified disposition parameters and gastric and colonic solubility according to buffer solubility at specified luminal pH. This identification was performed under the assumption that solubility was the major contributor of regional differences in absorption rate. Impact of maximum predicted effect of concomitant administration of ethanol was then simulated by multiplying gastric and duodenal solubility by the increase in solubility observed *in vitro*, that is, FaSSGF with or without ethanol and rFaSSIF with or without ethanol. The simulated ethanol effect was assessed as percental increase in maximum concentration (C_{max}) and area under the curve (AUC) from the simulated plasma concentration profiles.

In Vivo *Protocol*

The animal experiments were performed at Lundbeck (Copenhagen, Denmark) and Janssen (Beerse, Belgium). The protocol used for the *in vivo* studies was approved by the institutional animal ethics committees and were in accordance with Danish and Belgian law regulating experiments on animals, with EC directive 2010/63/EU and the NIH guidelines on animal welfare. Male Sprague-Dawley rats weighing 221-345 g on the day of the experiments were purchased from Charles River (Erkrath, Germany) and acclimatized with access to standard food and water *ad libitum* for at least 1 week before entering the experiment. Food was removed between 16-20 h before and 4 h after dosing. Water was available *ad libitum* at all times. The rats were randomly assigned to groups receiving the different treatments, with 5-6 animals per group.

For intravenous administration, 1 mg/kg compound was injected into the tail vain at 5 mL/kg. Rats were administered with an oral solution or suspension of felodipine or indomethacin by oral gavage. Solutions were dosed at 0.3 mL/kg to obtain a dose of 1 mg/kg. Suspensions of felodipine and indomethacin were dosed with 0.3 or 0.6 mL/kg, respectively, to equal a dose of 10 or 20 mg/kg. The doses of the solutions were based on the solubility of the compounds in the vehicles. High doses were selected for the suspension studies because they needed to result in a solubility-limited absorption in order to observe a potential ethanol effect. Compound administration was followed by an administration of 5 mL/kg of (1) deionized water (0% ethanol) or (2) 20% ethanol in deionized water. The dose of ethanol administered in the current rat study (0.2 mL/kg) is closely related to ethanol intake upon consumption of a standard glass of beer (0.19 mL/kg), wine (0.16 mL/kg), or whisky (0.23 mL/kg) by an adult weighing 70 kg.[17]

Blood samples (200 μL) were obtained by individual tail vein puncture and collected into plasma collection tubes containing dipotassium EDTA. Samples were taken between 5 min and 24 h postdose. The tubes were immediately centrifuged for 10 min at 3200 × *g* to obtain plasma which was stored at −80°C until further analysis. The animals were euthanized after the experiments.

Plasma Sample Preparation and Ultra Performance Liquid Chromatography–Tandem Mass Spectrometry Analysis

Plasma samples (50 μL) were extracted with 150 μL acetonitrile spiked with 50 nM warfarin as internal standard and centrifuged at 2465 × *g* at 4°C for 20 min. Plasma concentrations were determined by analyzing the supernatants using a Water Xevo TQ MS with electrospray ionization coupled to an Acquity UPLC system (Waters, Milford, MA). A Waters BEH C18 2.1 × 50 mm (1.7-μm) column was used for chromatographic separation. The mobile phase consisted of 5% acetonitrile and 0.1% formic acid in water (solvent A) and 0.1% formic acid in acetonitrile (solvent B). Gradient elution at a constant flow rate of 0.5 mL/min was performed as follows: 95% A decreased linearly to 10% from 0.5 to 1.2 min, followed by a constant flow of 10% A for 0.4 min and a linear increase back to 95% A at 1.7 min until the end of the run (2 min). The injection volume was 10 μL. The column oven and auto sampler tray temperature were set at 60°C and 10°C, respectively. The mass spectrometer was operated in the positive electrospray mode for indomethacin and felodipine and in negative mode for warfarin. The retention times of indomethacin, felodipine, and internal standard warfarin were 1:46, 1:56, and 1:42 min, respectively. Precursor-product ion pairs followed were: m/z 358→ 139 (collision energy 20 V) for indomethacin, m/z 385→278 (collision energy 25 V) for felodipine, and m/z 309→163 (collision energy 22 V) for warfarin. Data acquisition and peak integration were performed with MassLynx software (Waters).

Data Analysis

Data are presented as mean values with standard deviation. C_{max} and time to reach maximum plasma concentration (t_{max}) were determined directly from the plasma concentration-time profiles. The $AUC_{0\text{-}24h}$ values were calculated using the linear trapezoidal method. Plasma concentrations below the limit of quantitation were set equal to zero. Accurate determination of the terminal elimination rate constant and $AUC_{0\text{-}\infty}$ was not possible because insufficient data points were collected during the elimination phase, and after 24 h, plasma concentrations of felodipine were typically below the quantification limit.

Statistical analysis was performed with GraphPad Prism 7 (GraphPad Software). Student t-tests (for means) and Mann-Whitney tests (for medians) were used to evaluate differences between values obtained in the control condition (0% ethanol) and the 20% (v/v) ethanol condition. *p* values lower than 0.05 were considered statistically significant.

Results

Ethanol Effects on In Vitro *Dissolution in Simulated GI Fluids of Fasted Rats*

The apparent solubility of indomethacin and felodipine in FaSSGF has been determined previously[4] and was relatively low compared to values obtained in rFaSSIF (Table 2). The presence of ethanol significantly increased solubility of both model compounds in both fluids. Indomethacin solubility increased 13- and 1.9-fold when FaSSGF and rFaSSIF were supplemented with 20% of ethanol. The presence of the same proportion of ethanol increased felodipine solubility 14-fold in FaSSGF and 1.4-fold in rFaSSIF.

In vitro dissolution of the indomethacin and felodipine solutions and suspensions in the stomach during 30 min, the gastric residence

Table 3
In Vitro Dissolution of Indomethacin and Felodipine Solutions and Suspensions Expected in a Fasted Rat Stomach

Formulation	Ethanol	Compound Dissolved (%)	
		Indomethacin	Felodipine
Solution	0%	10.6 ± 1.8	26.8 ± 4.6
	20%	96.9 ± 3.8	95.1 ± 1.5
Suspension	0%	0.35 ± 0.17	0.35 ± 0.21
	20%	1.16 ± 0.10	4.11 ± 0.01

time in fasted rats,[13] is depicted in Table 3. Almost all indomethacin that was added as a solution remained in solution in the presence of 20% of ethanol whereas most of the compound precipitated in the absence of ethanol and only 10.6% ± 1.8% was dissolved after a 30 min incubation. Only small fractions of indomethacin suspension were expected to dissolve in a rat stomach but the presence of ethanol increased this fraction 4.2-fold (1.16% vs. 0.35%). Similar results were observed for dissolution studies with felodipine. Most of the compound added in solution was expected to precipitate in the absence of ethanol with only 26.8% of felodipine in solution after 30 min. The presence of 20% of ethanol increased this value to 95.1%. A small fraction of the felodipine suspension dissolved in the absence of ethanol (0.35%). However, the presence of 20% of ethanol increased this value to 4.11%, a 14.4-fold increase.

Ethanol Effects on Simulated Absorption

Total plasma clearance and volume of distribution were identified to 0.48 mL/min/kg and 0.2 L/kg for indomethacin and 60 mL/min/kg and 36 L/kg for felodipine. Fraction absorbed after oral administration of a solution was estimated to 1 for both drugs while the estimated oral bioavailability was 100% and 25% for indomethacin and felodipine, respectively. For indomethacin, apparent intestinal solubility was identified to 700 μg/mL in the duodenum and lower ileum and <1 μg/mL in all other small intestinal segments. Apparent intestinal solubility was identified to 180 μg/mL in duodenum, <10 μg/mL in jejunum, and 10-20 μg/mL in ileum for felodipine. The maximum estimated increase in C_{max} and AUC by ethanol coadministration, applying the fold increase in gastric and duodenal solubility given by *in vitro* measurements, was simulated to 30% and 21% for indomethacin and 36% and 26% for felodipine. Simulated plasma concentration-time profiles after oral administration of suspension without and with ethanol are displayed in Figures 1 and 2.

Ethanol Effects on In Vivo *Exposure in Fasted Rats*

The plasma concentration-time profiles determined following administration of indomethacin are shown in Figure 3, and the corresponding pharmacokinetic parameters are summarized in

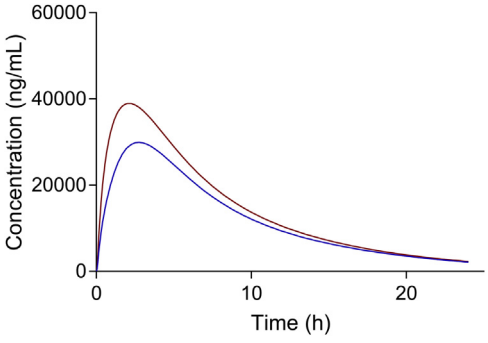

Figure 1. Plasma concentration profiles for indomethacin after administration of oral suspensions with water (blue line) or 20% ethanol (red line) simulated in PK-Sim.

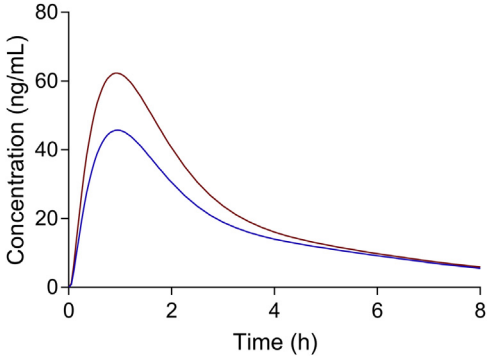

Figure 2. Plasma concentration profiles for felodipine after administration of oral suspensions with water (blue line) or 20% ethanol (red line) simulated in PK-Sim.

Table 4. Statistical analysis was performed to compare the same formulation, that is, solutions or suspensions, followed by immediate administration of water or ethanol. For rats receiving indomethacin in solution, statistical tests showed a significantly higher value for C_{max} in the absence of ethanol (4963 ± 1138 ng/mL) than for those that had 20% of ethanol administered (3558 ± 966; $p = 0.03$). No significant differences were observed between the $AUC_{0\text{-}24h}$ and t_{max} of this solution administered with water and ethanol. Pharmacokinetic parameters were similar when suspensions of indomethacin were administered with water or 20% of ethanol. Although not significant, t_{max} values for indomethacin solutions and suspensions tended to increase when ethanol was administered. The $AUC_{0\text{-}24h}$ was corrected for the administered dose ($AUC_{0\text{-}24h}$/D) to allow comparison between these solutions and suspensions. No statistically significant differences were observed between the

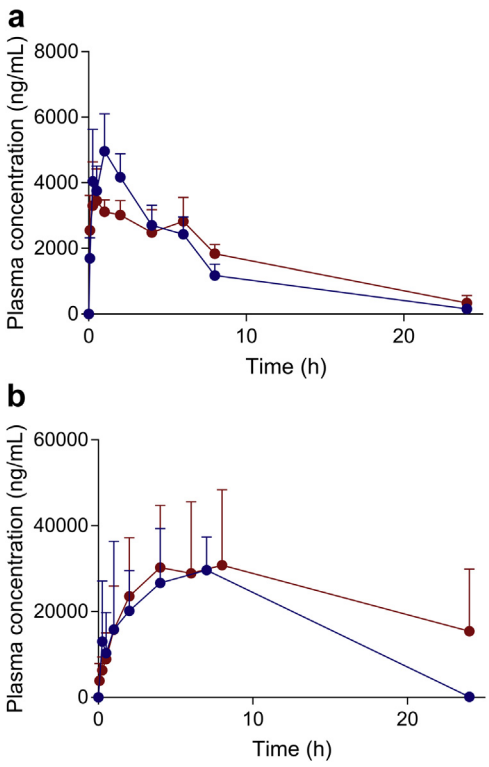

Figure 3. Mean plasma concentration-time profiles (±SD) for indomethacin after administration of (a) oral solutions or (b) oral suspensions. The formulations were administered with water (blue line) or 20% ethanol (red line).

Table 4
Summary of Pharmacokinetic Parameters for Indomethacin After Administration of Solutions and Suspensions With Water (0% Ethanol) or Ethanol (20%)

Formulation	Ethanol Administered	Sample Size	C_{max} (ng/mL)[a]	t_{max} (h)[b]	$AUC_{0\text{-}24h}$ ((ng/mL)h)[a]	$AUC_{0\text{-}24h}$/D (h/mL)[a]
Solution[c]	0%	6	4963 ± 1138	1.00 (0.25-1.00)	34,552 ± 6721	0.11 ± 0.02
	20%	6	3558 ± 966[e]	0.50 (0.25-6.00)	39,026 ± 5355	0.12 ± 0.01
Suspension[d]	0%	5	29,667 ± 7723	4.00 (0.50-7.00)	413,753 ± 75,084	0.09 ± 0.01
	20%	6	30,830 ± 17,577	4.00 (4.00-8.00)	571,582 ± 349,456	0.11 ± 0.05

[a] Mean ± SD.
[b] Median (range).
[c] 1 mg/kg.
[d] 10 mg/kg.
[e] $p < 0.05$ compared to 0% ethanol.

2 formulations (i.e., solutions and suspensions) containing indomethacin, irrespective of the presence of ethanol.

Figure 4 shows the plasma concentration-time profiles after administration of felodipine with water or ethanol. Pharmacokinetic parameters determined based on these profiles are presented in Table 5. No differences were detected between parameters determined following administration of felodipine solutions and suspensions with or without ethanol. The $AUC_{0\text{-}24h}$/D was significantly lower upon administration of felodipine suspensions than after administration of felodipine solutions.

Discussion

This study evaluated the effect of ethanol as a cosolvent on the intestinal absorption of indomethacin and felodipine in rats. The indomethacin results corroborated the expectations: no increase in indomethacin absorption was observed with subsequent intake of ethanol *in vivo*. Although significant precipitation of the indomethacin solution and incomplete dissolution of the indomethacin suspension were predicted to occur in the stomach of the rats (Table 3), the acidic compound is expected to dissolve completely in the small intestine (due to ionization), followed by rapid absorption. The preclinical dose number (PDo) supports this proposed mechanism. The PDo was calculated by dividing the dose (mg/kg) by the compound solubility in rFaSSIF (mg/mL) to obtain the volume of rFaSSIF required to fully dissolve the dose.[18] In the absence of ethanol, PDo values were 1 and 21 mL/kg for the indomethacin solution and suspension, respectively, lower than the expected gut volume after administration of the formulation with water (23 mL/kg).[14] In addition, $AUC_{0\text{-}24h}$/D for solutions and suspensions of indomethacin is comparable, indicating that dissolution and solubility do not limit absorption (Table 4).

In contrast, $AUC_{0\text{-}24h}$/D of felodipine was higher following administration of solutions than suspensions, indicating a dissolution- or solubility-limited absorption of this dose of the compound in the rat intestine (Table 5). This is supported by the PDo values for felodipine suspensions in the absence (52 mL/kg) or presence (38 mL/kg) of ethanol. Although the PDo decreased considerably in the presence of ethanol, *in vivo* dissolution and solubility of felodipine seemed unaffected by ethanol, as simultaneous intake of felodipine suspensions with ethanol did not alter the exposure of this drug in plasma.

Physiologically based pharmacokinetic simulations were conducted to estimate the maximum effect on plasma exposure based on solubility measurements (Table 2); other potential effects of ethanol were not considered in the simulations. The simulations predicted a small ethanol effect, ~20%-30% in C_{max} and AUC, for both felodipine and indomethacin. This effect was not observed *in vivo*, which either can be due to an absence of *in vivo* effects or that this level of difference is too small to be discriminated given the variability in the *in vivo* data. In all, simulations support the observations that the effects of ethanol on gastric and intestinal solubility is negligible for the *in vivo* exposure of felodipine and indomethacin in rats. A possible explanation for the lack of an ethanol effect on felodipine absorption *in vivo* could be that dilution, absorption, and transfer of ethanol upon arrival to the stomach resulted in ethanol concentrations too low to affect local solubility in the GI tract. The presence of 5% (v/v) of ethanol had negligible effects on felodipine solubility.[3] Assuming that the total amount of fluid in the GI tract of fasted rats is approximately 1.8% of total body weight,[14] the GI tract of the rats used in the present study contained 4.0-6.3 mL of fluid. Administration of a drug solution (0.07-0.10 mL) or suspension (0.07-0.21 mL) and an ethanol solution (1.11-1.72 mL) would therefore result in a substantial dilution of the ethanol. Gastric absorption of ethanol is, however, limited (~5%/30 min) due to the relatively small surface of the gastric epithelium.[19-21] A disappearance of 75% of ethanol from the stomach within 30 min of ingestion, despite the low absorption rate, indicates a rapid transfer of ethanol to the duodenum.[20] Once in the duodenum, ethanol (which is a small, neutral molecule) is rapidly absorbed through passive diffusion.[21,22]

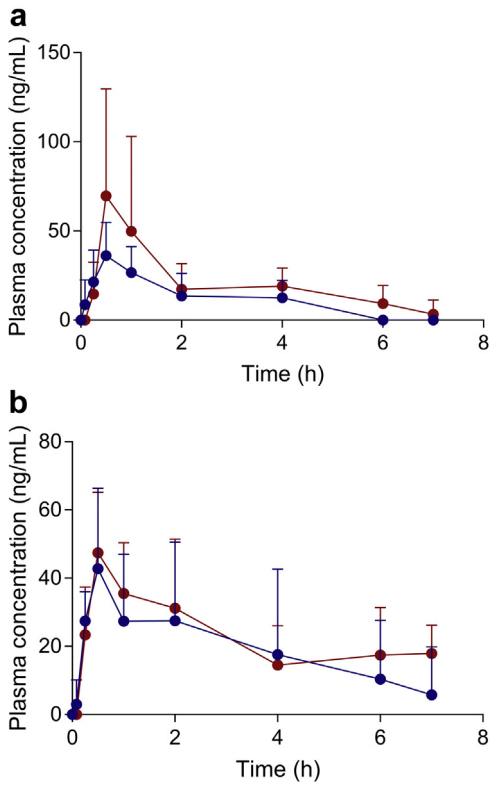

Figure 4. Mean plasma concentration-time profiles (±SD) for felodipine after administration of (a) oral solutions or (b) oral suspensions. The formulations were administered with water (blue line) and 20% ethanol (red line).

Table 5
Summary of Pharmacokinetic Parameters for Felodipine After Administration of Solutions and Suspensions With Water (0% Ethanol) or Ethanol (20%)

Formulation	Ethanol Administered	Sample Size	C_{max} (ng/mL)[a]	t_{max} (h)[b]	$AUC_{0\text{-}24h}$ ((ng/mL)h)[a]	$AUC_{0\text{-}24h}$/D (h/mL)[a]
Solution[c]	0%	6	36.18 ± 18.57	0.50 (0.25-1.00)	81.41 ± 32.93	$2.63 \times 10^{-4} \pm 1.04 \times 10^{-4}$
	20%	6	69.69 ± 59.98	0.25 (0.25-0.50)	101.19 ± 67.79	$3.17 \times 10^{-4} \pm 2.15 \times 10^{-4}$
Suspension[d]	0%	6	51.09 ± 21.83	0.50 (0.25-0.50)	143.16 ± 132.39	$4.62 \times 10^{-5} \pm 4.15 \times 10^{-5}$
	20%	6	47.41 ± 17.76	0.50 (0.25-2.00)	160.15 ± 86.60	$5.27 \times 10^{-5} \pm 2.87 \times 10^{-5}$

[a] Mean ± SD.
[b] Median (range).
[c] 1 mg/kg.
[d] 20 mg/kg, $AUC_{0\text{-}24h}$/D suspension is significantly lower than $AUC_{0\text{-}24h}$/D solution.

In addition to affecting GI solubility, ethanol has been associated with a number of physiological processes in the GI tract that are of importance for drug absorption and the resulting plasma concentration profile. Ethanol at 4%-40% has been shown to delay gastric emptying, partly due to pylorospasms.[20,22,23] This effect can account for the tendency for higher t_{max} values after administering formulations of indomethacin with ethanol. Delayed gastric emptying results in a slower, more continuous transfer of drug solutions and suspensions from the stomach to the duodenum, which could explain the significantly lower C_{max} value determined for the indomethacin solution administered with 20% ethanol.

Physiological effects in the GI tract in the presence of ethanol can also affect permeability. In the stomach, ethanol can enhance the blood flow in the gastric mucosa, which increases the sink conditions in the serosal compartment and thus the permeability.[24] In the intestine, increased absorption in the presence of ethanol (18% v/v) could occur as a result of higher mucosal and microvascular permeability.[25] It is doubtful that these effects manifested in the present study for 2 reasons: (1) the majority of drug absorption occurs in the intestine,[20] which limits the effect of an increased gastric blood flow on plasma exposure; and (2) due to dilution, absorption, and transfer of ethanol, it is unlikely that concentrations of 18% (v/v) were reached in the GI tract.

To date, no data are available regarding effects of ethanol on dissolution and solubility of poorly soluble drugs in the GI tract of humans. *In silico* simulations, based on apparent solubility data in simulated GI fluids containing 20% of ethanol, predicted an increase in *in vivo* exposure of nonionized lipophilic compounds.[4] However, a recent study by Rubbens et al.[17] reported ethanol concentrations in gastric and duodenal fluids that were much lower than the 20% used in the *in silico* simulations.[4,17] The consumption of beer, wine, or whisky, with ethanol contents of 5.2%, 11.0%, and 40.0%, respectively, resulted in ethanol concentrations of maximum 4.1%-11.4% and 2.0%-5.9% in the stomach and duodenum. Moreover, those concentrations rapidly declined, resulting in less than 0.05% ethanol in both stomach and duodenal fluids after 2 h. The authors explained the low ethanol concentration by dilution, absorption, and transfer of ethanol upon arrival in the stomach. Dilution in the human stomach is highly dependent on the volume of the beverage ingested. The fasted state stomach of humans typically contains 50 mL fluid or less.[26] Consumption of 500 mL of beer, 200 mL of wine, or 80 mL of whisky would thus result in 1.10-, 1.25-, or 1.63-fold dilutions of ethanol in the human stomach.[17] In humans, 10%-20% of orally ingested ethanol has been reported to be absorbed in the stomach,[19,27] whereas ethanol transfer from the stomach to the duodenum is rapid. In some cases, ethanol t_{max} determined in the duodenum of healthy volunteers was even lower than in the stomach, indicating an instantaneous transfer of ethanol to the intestine.[17]

This study aimed at evaluating the absorption effect of GI ethanol concentrations on poorly water-soluble compounds in general. However, despite dilution, absorption, and transfer of ethanol in the human GI tract, ethanol effects on the extent of absorption of commercial felodipine formulations are unlikely to occur in humans. Felodipine is marketed as an extended-release formulation making use of amorphous felodipine (Plendil) which already results in complete absorption in the absence of the cosolvent.[4,15,28] In addition, when changing from amorphous to micronized felodipine, maintaining the exact composition of Plendil, absorption was complete. However, in comparison, crude, nonmicronized felodipine formulated in the Plendil composition resulted in significantly lower absorption (personal communication, AstraZeneca). Hence, also in humans, felodipine shows dissolution rate—limited absorption and is in need of enabling formulation strategies (e.g., amorphization) to be completely absorbed.

Despite the absence of a clear effect of ethanol on drug absorption in rats and the low probability of these effects to occur in humans, it is important to emphasize that many drugs can interact with ethanol on a pharmacodynamic and pharmacokinetic level. On a pharmacodynamic level, ethanol can enhance effects of, for example, antipsychotics and antidepressants, as both ethanol and these compounds reduce the activity of the central nervous system.[29] In addition, ethanol can increase the effect of some antihypertensive drugs, because ethanol causes vasodilation.[29] The pharmacokinetic profile can be affected by dose dumping, as mentioned before.[1,19] However, most ethanol-drug interactions at the pharmacokinetic level result from interference with metabolizing enzymes. Ethanol is both a substrate and inducer of cytochrome P450 (CYP) 2E1, and a substrate of alcohol dehydrogenase, and it can therefore interact with drugs that are substrates, inducers, or inhibitors of these enzymes.[29] The therapeutic effect of disulfiram, a compound used to treat chronic alcoholism, is based on such an enzyme interaction.[30] In our study, enzyme interactions were avoided by the careful selection of model compounds that are metabolized instead by CYP2C (indomethacin) and CYP3A (felodipine).[31,32]

Conclusion

The concomitant intake of ethanol (dosed as 20% v/v) with felodipine or indomethacin solutions and suspensions did not increase the *in vivo* plasma exposure of these compounds in fasted rats. Dilution, absorption, and transfer of ethanol from the stomach and intestine could result in ethanol concentrations that did not reach levels required to affect local solubility in the GI tract. In contrast with our hypothesis, increased bioavailability, due to increasing drug dissolution and solubility in GI fluids in rats, is unlikely to occur. The literature suggests the same situation for humans where intragastric and intraduodenal concentrations (1) are expected to be relatively low as compared to ingested ethanol concentrations and (2) rapidly decline. This most likely results in GI ethanol concentrations that are too low to affect local solubility.[17]

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