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**RESEARCH ARTICLE**

# Predictive models for drugs exhibiting *negative food effects* based on their biopharmaceutical characteristics

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**Abstract**

*Context*: A drug is defined to exhibit food effects if its pharmacokinetic parameter, area under the curve ($AUC_{0-\infty}$) is different when co-administered with food in comparison with its administration on a fasted stomach. Food effects of drugs administered in immediate release dosage forms were classified as *positive*, *negative*, and *no food effects*.

*Objective*: In this study, predictive models for negative food effects of drugs that are stable in the gastrointestinal tract and do not complex with $Ca^{2+}$ are reported.

*Methods*: An empirical model was developed using five drugs exhibiting negative food effects and seven drugs exhibiting no food effects by multiple regression analysis, based on biopharmaceutical properties generated from *in vitro* experiments. An oral absorption model was adopted for simulating negative food effects of model compounds using *in situ* rat intestinal permeability.

*Results*: Analysis of selected model drugs indicated that percent food effects correlated to their dissociation constant, $K$ ($K_a$ or $K_b$) and Caco-2 permeabilities. The obtained predictive equation was: Food effect (%) = $(2.60 \times 10^5 \cdot P_{app}) - (2.91 \times 10^5 \cdot K) - 8.50$. Applying the oral absorption model, the predicted food effects matched the trends of published negative food effects when the two experimental pH conditions of fed and fasted state intestinal environment were used.

*Conclusion*: A predictive model for negative food effects based on the correlation of food effects with dissociation constant and Caco-2 permeability was established and simulations of food effects using rat intestinal permeability supported the drugs' published negative food effects. Thus, an empirical and a mechanistic model as potential tools for predicting negative food effects are reported.

**Keywords:** Predictive model, biopharmaceutical properties, negative food effects, Caco-2 permeability, dissociation constant

## Introduction

The oral route is a convenient, less expensive, and preferred route of drug delivery[1,2]. Co-administration of drugs with food can result in changes in a drug's pharmacokinetics[3,4]. Depending upon a drug's onset, duration of action, and therapeutic window, a change in its pharmacokinetics could affect its treatment outcome[4]. A drug is known to show *food effects* if there are differences in its pharmacokinetics when co-administered with food in comparison with its fasted state administration[5]. Food effects can be reflected in the changes of one or many of a drug's pharmacokinetic parameters (area under the curve, $AUC_{0-\infty}$, peak plasma concentration, $C_{max}$, and time at maximum plasma concentration, $T_{max}$)[3]. Food effects were defined as *positive*, when co-administration of food causes a 25% or more increase in $AUC_{0-\infty}$; *negative*, when food causes 20% or more decrease in $AUC_{0-\infty}$; and lastly, *no effect* when co-administration of food causes no statistically significant change (80–125%) in the $AUC_{0-\infty}$ of a drug, when compared with the fasted state drug administration[5,6]. The extent of food effect was mathematically represented as follows, Equation (1):

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$$\text{Food effect}(\%) = \frac{\left[\text{AUC}_{(0-\infty)\text{fed}} - \text{AUC}_{(0-\infty)\text{fast}}\right]}{\text{AUC}_{(0-\infty)\text{fast}}} \times 100 \quad (1)$$

Depending upon the therapeutic significance of a food effect, a drug label contains recommendations about its use to the patient. Accelerated or delayed food effects (changes in $T_{max}$) or changes in $C_{max}$ for immediate release dosage forms with no change in $AUC_{0-\infty}$ may not be of clinical significance, because the extent of absorption of the drug is the same in fed and fasted state drug administrations[7]. Negative food effect is a clinical disadvantage as the drug exposure to the patient is less than in fasted state administration. Imposition of restriction of drug administration prior to food intake may result in non-compliance and failure of drug therapy. Due to the above stated reasons, it is important to understand and study food effects early in drug development[8].

Predictive relationships between biopharmaceutical properties and fasted state drug absorption have been developed, but a predictive model applying the biopharmaceutical properties of drugs to the drugs' postprandial absorption has been difficult[9]. Empirical observations have been made in literature correlating the biopharmaceutical classification system (BCS) and the food effects of drugs[4,8]. The correlation of food effects with physicochemical properties, such as solubility, partition coefficient, p$K_a$, and so on, of drug molecules and applying logistic regression analysis and BCS were described in recent studies[10,11]. Currently, limited literature is available specifically for the prediction of negative food effects of an individual molecule or a class of molecules[3,4,8].

Previous investigations from our laboratory have shown that negative food effects were exhibited by hydrophilic, highly ionizable, acidic or basic drugs that exhibit low permeability, and whose solubility or permeability is decreased in the fed state condition, when compared with the fasted state condition[6]. Thus, the negative food effects of a drug could be related to its biopharmaceutical properties that govern its absorption. The objectives of this study were to correlate negative food effects of drugs to the drugs' biopharmaceutical properties and to develop an *in vitro/in situ* model for predicting the negative food effects of drugs. The role of specific chemical interactions such as complexation to metal ions/$Ca^{2+}$ and luminal degradation, and bile and lipid digestion products improving the solubilization of poorly aqueous soluble drugs in the fed state are well-documented; and drugs that exhibit such food effects are beyond the scope of this investigation[12–14].

## Materials and methods

### Building the empirical model

The p$K_a$, Caco-2 permeability, and Log *P* of 38 drugs exhibiting negative food effects and no food effects were studied[6]. Drugs that exhibited positive food effects were not included. Correlation of drugs exhibiting negative food effects with Log *P* and Log Caco-2 permeability indicated that the drugs exhibited low Caco-2 permeability ($<3\times10^{-6}$ cm/sec) and low Log *P* values (<1)[6]. Drugs exhibiting >90% fraction dose absorption did not exhibit negative food effects, whereas drugs that had incomplete GI fraction dose absorption exhibited negative food effects[6]. In addition, the drugs exhibiting negative food effects possessed relatively lower p$K_a$ (in case of acidic drugs) and higher p$K_a$ (in case of basic drugs) when compared with other low permeability drugs that did not exhibit negative food effects[6]. Neutral or zwitterionic drugs did not exhibit negative food effects[6].

#### *Criteria for the selection of model drugs*

The model drugs were selected using the following criteria:

1. Drugs exhibiting negative food effects and no food effects were selected from available literature. In the clinical studies, the *fasted state* (preprandial) drug administration typically involved the administration of a drug with water or liquids on an overnight fasted stomach and a *fed state* (postprandial) drug administration involved the administration of a drug after a standard breakfast or a high-fat meal on an overnight fasted stomach[5].
2. Drugs in the molecular size range of 200–450 Da were chosen to keep the unknown variability of size among drugs to a minimum.
3. Only weakly acidic or weakly basic drugs were chosen because zwitterionic and non-ionizable drugs did not exhibit negative food effects[6].
4. Only drugs that did not have any known physiological effects on the gastrointestinal secretions and motility were chosen.

Five drugs exhibiting negative food effects (atenolol, nadolol, tacrine, furosemide, and pravastatin) and seven drugs exhibiting no food effects (ketoprofen, indomethacin, diltiazem, hydrochlorothiazide, ibuprofen, zidovudine, and diclofenac) were used for the empirical model.

#### *Dissociation constants*

The acidic and basic dissociation constants ($K_a$ and $K_b$) of the model compounds were obtained from literature.

#### *Multiple regression analysis*

Statistical analysis was performed using NCSS-PASS® (Dawson Edition 2000, Utah) and MS Excel®.

#### *Caco-2 permeability*

Caco-2 permeability data as shown in Table 2 was either obtained from the literature or experimentally determined at pH 6.7. The methodology used for the

determination of Caco-2 permeability was previously published[6].

### *External data*

One drug that exhibits negative food effects and two drugs that exhibited no food effects that were not previously included in building of the model were selected from the literature to challenge the developed model. Valsartan was reported to exhibit negative food effects (–40%)[15,16]. It is a weakly acidic drug with a p$K_a$ of 3.9 that was classified as a BCS-III drug[17] and exhibited very low permeability across Caco-2 monolayers, that could not be quantitatively determined[16,18]. The Caco-2 permeability of valsartan was assumed to be about $0.5 \times 10^{-6}$ cm/sec (a conservative estimate) for predicting its food effects using the empirical model.

The second test using two drugs exhibiting no food effects was performed to check whether the model "wrongly" predicts the no food effects of a drug (a false negative test) as a negative food effect. One was a weakly acidic drug, theophylline and the other was a weakly basic drug, timolol. Theophylline exhibited a food effect of +8.24% (non-significant)[19]. Its p$K_a$ is 8.7 ($K_a = 1.995 \times 10^{-9}$) and its Caco-2 permeability is $24 \times 10^{-6}$ cm/sec at pH 6.5[20]. Timolol exhibited a food effect of +18.98% (non-significant)[21]. Its p$K_a$ is 9.2 (p$K_b$ = 4.8, $K_b = 1.585 \times 10^{-5}$) and its Caco-2 permeability is $1 \times 10^{-6}$ cm/sec at pH 6.5[20].

Table 1. Upper intestinal conditions used in simulations for fed and fasted states.

| Parameter | Preprandial upper intestinal condition | Postprandial upper intestinal condition |
|---|---|---|
| pH | 6.8[22] | 5[22] |
| NaTC | 3 mM[24] | 15 mM[24] |
| Volume | 240 mL[5] | 1 L[5,44] |

## Biopharmaceutical properties of model compounds

### *Buffer media*

To determine the solubility and permeability properties of model compounds in different conditions, relevant buffers were chosen for studies. These buffers mimicked the pH conditions of the upper intestinal tract[22]. The pH of the upper intestine in the fasted state is about 6.5, whereas the overall postprandial pH after a standard meal (equivalent in calorie content to the FDA recommended high-fat meal) during 4 h in the duodenum was 5.4 (5.0–5.7)[5,23]. Fed state simulated intestinal fluid (FeSSIF), which contains bile at physiological concentrations, was also adopted for physicochemical characterization[24]. The buffers used were phosphate–citrate buffer (pH 5.0 ± 0.1), Sorensen's phosphate buffer (SPB pH 6.7 ± 0.1)[25], and FeSSIF (pH 5.0) simulating the fasted and fed state conditions[24,26]. Phosphate–citrate buffer was used instead of FeSSIF for Caco-2 permeability studies because the integrity of cell monolayer was compromised with FeSSIF.

### *Data*

The information about dose, molecular weight, human fraction dose absorbed, and percent food effects were collected from literature[27,28].

Table 2. Model compounds used for multiple regression analysis.

| Drug | Property | MW | p$K_a$ | p$K_b$ | Dissociation constant ($K_a$ or $K_b$) | Caco-2 permeability ($\times 10^{-6}$ cm/sec) | BCS class | Published food effects (%) | Predicted food effects (%) |
|---|---|---|---|---|---|---|---|---|---|
| Atenolol | Weakly basic | 266.336 | 9.6 | 4.4 | $3.98 \times 10^{-5}$ | 0.18 ± 0.04 | III[45] | –20[46] | –20.18 |
| Nadolol | Weakly basic | 309.401 | 9.6 | 4.4 | $3.98 \times 10^{-5}$ | 0.21 ± 0.03 | III[45] | –20.75[47] | –20.17 |
| Pravastatin | Weakly acidic | 424.528 | 4.2 | 9.8 | $6.31 \times 10^{-5}$ | 0.123[a,48] | III[17] | –37.5[49] | –26.72 |
| Furosemide | Weakly acidic | 330.745 | 3.9 | 10.1 | 0.00013 | 0.73 ± 0.41 | IV[45] | –44[50] | –44.18 |
| Tacrine | Weakly basic | 252.74 | 9.85 | 4.15 | $7.05 \times 10^{-5}$ | 8.48 ± 0.54 | III | –21[51] | –26.83 |
| Ketoprofen | Weakly acidic | 254.281 | 4.3 | 9.7 | $5.01 \times 10^{-5}$ | 80[a,20] | II[45] | –8.6[52] | –3.47 |
| Indomethacin | Weakly acidic | 357.787 | 4.5 | 9.5 | $3.16 \times 10^{-5}$ | 105[a,20] | II[45] | +10.52[53] | +7.85 |
| Diltiazem | Weakly basic | 414.519 | 7.7 | 6.3 | $5.01 \times 10^{-7}$ | 29.8 ± 0.22[a,54] | I[17] | +0.2[55] | –1.88 |
| Ibuprofen | Weakly acidic | 206.281 | 5.2 | 8.8 | $6.31 \times 10^{-6}$ | 8.81 ± 0.49 | II[45] | –8.7[56] | –8.66 |
| Zidovudine | Weakly acidic | 267.242 | 9.6 | 4.4 | $2.51 \times 10^{-10}$ | 8.5 ± 0.2[a,57] | I[45] | –5.53[58] | –6.97 |
| Hydrochlorothiazide | Weakly acidic | 297.72 | 7.9 | 6.1 | $1.26 \times 10^{-8}$ | 2.16 ± 0.68 | III[45] | –3.73[59] | –8.53 |
| Diclofenac | Weakly acidic | 296.148 | 4 | 10 | 0.0001 | 70[a,20] | II[45] | –14.8[60] | –19.92 |

[a]Obtained from literature.

#### *Solubility*

The solubility of atenolol, furosemide, nadolol, and tacrine were obtained from published literature, which were determined in fed and fasted state pH conditions[6].

#### *Intrinsic dissolution rate*

The diffusivities of atenolol and furosemide were determined in fed and fasted state intestinal pH conditions by the intrinsic dissolution rate experiments as previously published[6]. In brief, the drug compact was exposed to pH 6.5 and 5.0 dissolution media in a modified Wood apparatus (VanKel Intrinsic Dissolution Apparatus, Varian Inc., Cary, NC) and rotated at 50, 100, 200, and 350 rpm at 37°C[29,30]. Analysis of the compounds was done using an Opti-Diss UV fiber optic system (Distek Inc. North Brunswick, NJ).

#### *Hayduk–Laudie method (diffusivity in water)*

The diffusivities of nadolol and tacrine were calculated using the Hayduk–Laudie method, because their intrinsic dissolution could not be determined due to their rapid dissolution[31,32].

#### *In situ permeability studies—single pass intestinal perfusion in rat*

The *in situ* permeability of model compounds across rat jejunum that were determined in fed and fasted state pH conditions was obtained from previously published literature[6]. *In situ* permeation of model drugs was performed in male or female Wistar rats (150–175 g). The permeability of each model drug was evaluated in fasted and fed state pH conditions in three rats, using a total of 24 rats for all drugs. Each rat was perfused with drug solution in fasted state pH condition, followed by fed state pH condition. The jejunal segment was flushed with phosphate-buffered saline (PBS) between the pH perfusion studies. All animal studies were conducted at Kakatiya University, Warangal, India and the studies were approved by The Institutional Animal Ethical Committee. The methodology of determination of rat jejunal permeability was previously published[6].

#### *Oral drug absorption model*

A previously published mechanistic mass-balance model was adopted for simulating food effects of model compounds using rat intestinal model for permeability and the physicochemical properties of the molecules corresponding to the fed and fasted state intestinal conditions[33]. The input parameters were diffusivity, solubility, and rat intestinal permeability, and the output parameter was the fraction dose absorbed (fed vs. fasted state). The drug particle size radius was assumed to be 0.025 cm, density of the luminal medium as 1.2 g/mL, and intestinal transit as 3 h[34]. Large intestinal absorption of drugs was not considered in the model. Simulations of food effects were done using the physiological conditions as shown in Table 1 using Micromath Scientist® (v 2.01).

### Analysis of model drug compounds

The model drug compounds were analyzed by HPLC as previously reported[6].

## Results

### Empirical model

There was no correlation between the extent of negative food effects and any of the following drug properties: partition/distribution coefficient, solubility/micellar solubilization, contact angle, surface activity, intrinsic dissolution rate, reported bioavailability, plasma clearance, and dose ($P \geq 0.05$). However, clear trends were observed when the model compounds selected for multiple linear regression analysis were correlated with the drugs' dissociation constants (acidic or basic) ($P < 0.05$) and Caco-2 permeability ($P < 0.05$) as shown in Table 3. As the Caco-2 permeability of compounds increased, the percent food effects became negligible (no food effects) (Figure 1) indicating that high permeability drugs do not exhibit negative food effects. And, as shown in Figure 2, with an increase in the dissociation constant, the drugs showed a tendency to exhibit negative food effects indicating that drugs exhibiting negative food effects were highly ionizable.

A multiple linear regression analysis was performed using least square estimation with percent food effects as the dependent variable and acidic/basic dissociation constants along with Caco-2 permeability as independent variables. The analysis on the model drugs indicated that the percent food effect was dependent on acidic/basic dissociation constant ($K$) ($P < 0.05$) and on Caco-2 permeability ($P_{app}$) ($P < 0.05$), thus, statistically significant relationships ($P < 0.05$) were obtained. The output of the NCSS/PASS software as *predicted food effects* is shown in Table 2 and the statistical test results of the empirical model are shown in Table 3. The equation obtained from the regression fit is:

$$\text{Food effect}\,(\%) = (2.60 \times 105 \bullet P_{app}) - (2.91 \times 105 \bullet K) - 8.50.$$

The $R^2$, $P$-value and power of the regression model were 0.9114, 0.00002, and >0.999, respectively (Figure 3) indicating that the model fit the data with high correlation and power. When both the parameters, namely,

Table 3. Test results of the empirical model.

| Independent variable | Regression coefficient | Standard error | Probability level | Decision | Power (5%) |
|---|---|---|---|---|---|
| Intercept | –8.5 | 2.4 | <0.00634 | Reject null hypothesis | >0.8815 |
| $K$ ($K_a$ or $K_b$) | $-2.91 \times 10^5$ | $0.376 \times 10^5$ | <0.00003 | | >0.9999 |
| $P_{app}$ | $2.6 \times 10^5$ | $0.413 \times 10^5$ | <0.00015 | | >0.9998 |

dissociation constant and Caco-2 permeability were applied together in the empirical model, the correlation coefficient was >0.91. This was a lot of improvement over the correlation coefficients obtained when the parameters were applied independently of each other, as shown in Figures 1 and 2.

The coefficient of determination adjusted for degrees of freedom (Adj. $R^2$) was 0.892 and the obtained $F$-ratio was 46.3. $Q^2$, the leave-one-out (LOO) cross-validated correlation coefficient reflects the prediction ability of the model, and is used to validate the model without selecting another sample or splitting the data[35]. The predictability of the model cross-validated by the LOO method yielded a $Q^2$ of 0.857 indicating a high prediction ability of the model. Also, the histograms of the residuals, as shown in Figure 4, were found to be normally distributed suggesting that the assumption of normality of the error term is correct[36]. The three normality

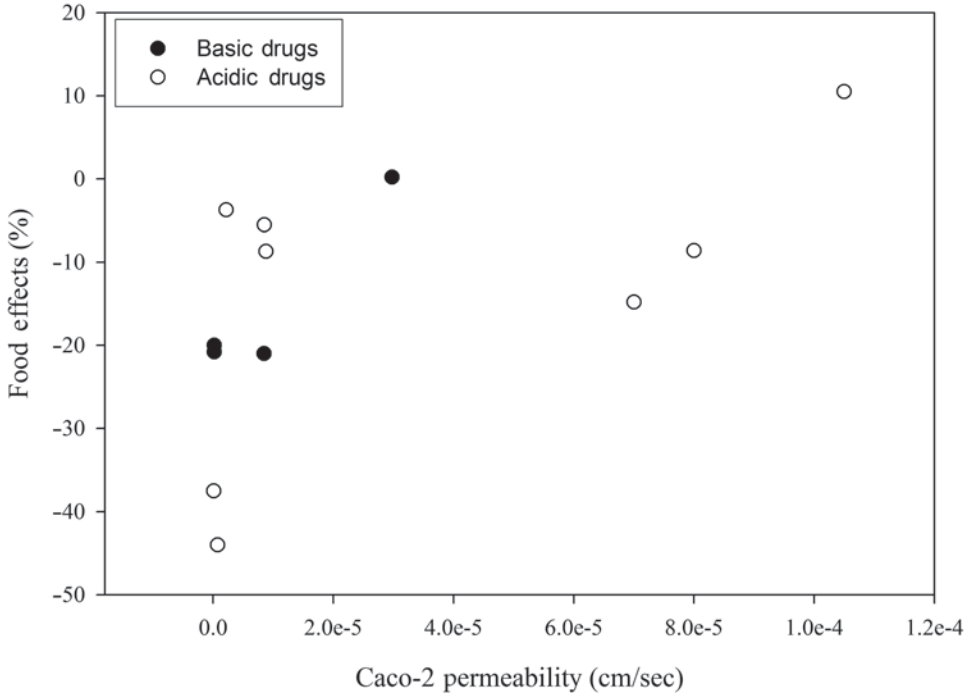


Figure 1. Relationship between Caco-2 permeability and percent food effects (logarithmic plot $R_2 = 0.491$).

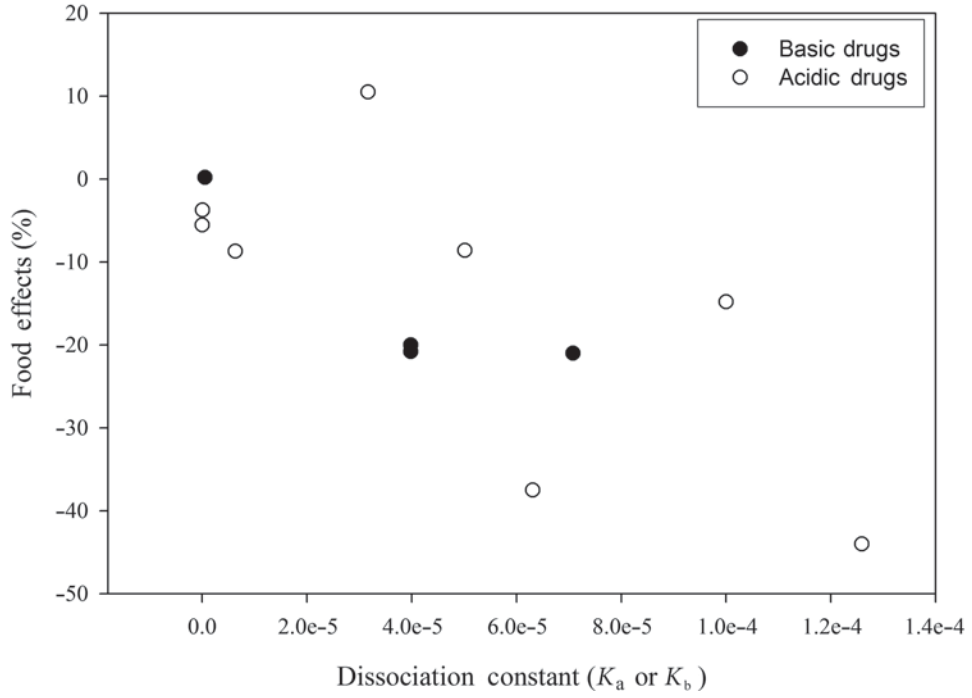


Figure 2. Relationship between dissociation constant and percent food effects (linear plot $R_2 = 0.52$).

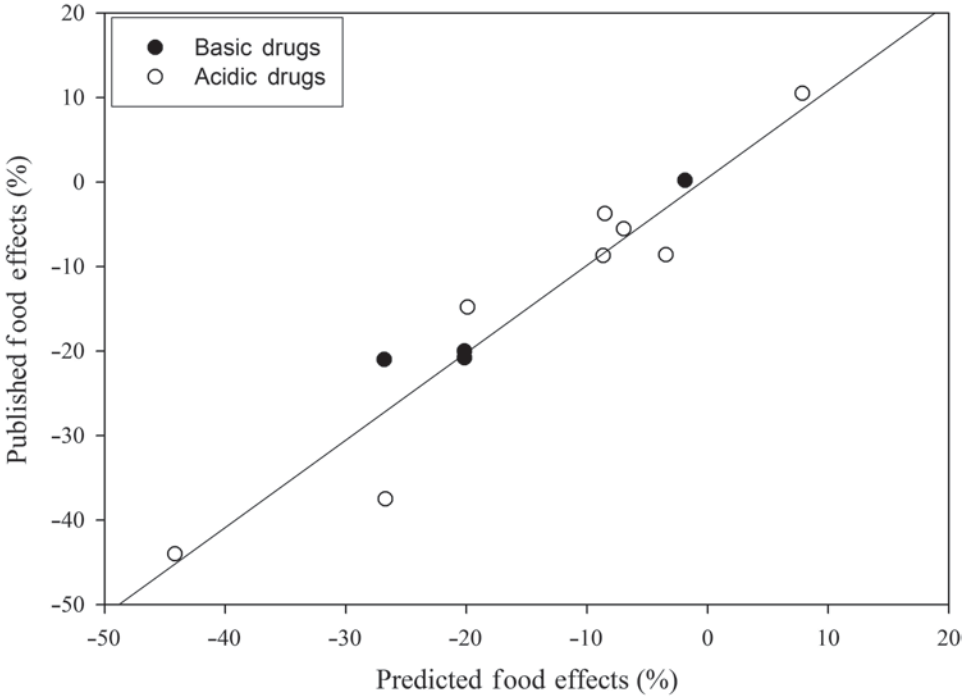


Figure 3. Correlation between predicted food effects and published food effects.

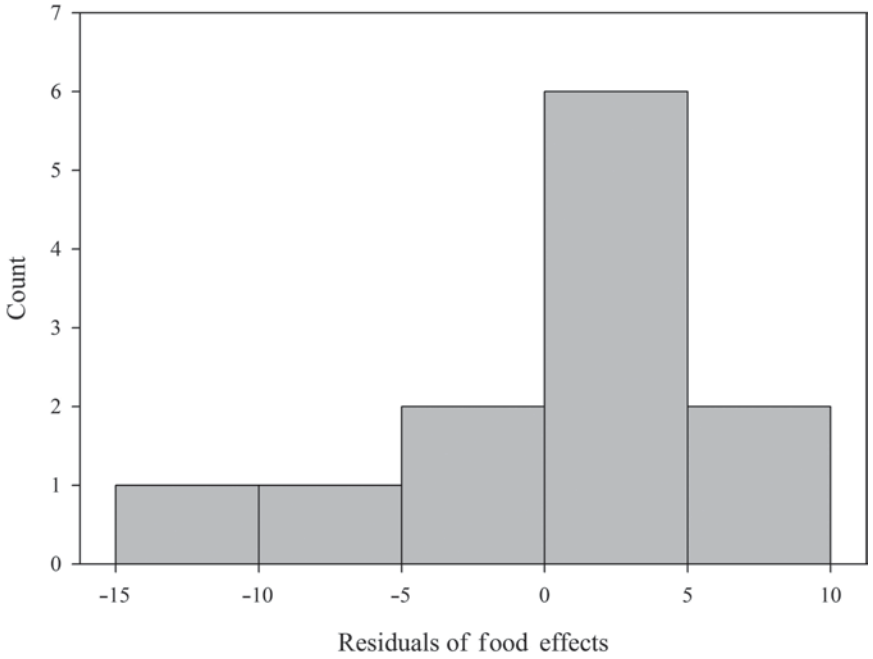


Figure 4. Histogram of residuals of food effects.

tests Skewness, Kurtosis, and Omnibus also indicated that the residuals were normally distributed. In addition, the quantile–quantile ($Q$–$Q$) plot of the residuals showed that all the points lie close to a straight line. This is characteristic of normal distribution of residuals. Residual analysis performed on the data revealed a random, symmetric scattering of points about zero, as shown in Figure 5. Test for serial correlation gave a Durbin–Watson value of 2.06 that was close to 2, which indicated that the residuals are not correlated. Finally, the test for multicollinearity, Eigen values for centered correlations, indicated that multicollinearity may not be a problem for this data set. Thus, all statistical tests strongly indicated the validity of the model.

## External data set

As shown in Table 4, the $K_a$ (or $K_b$) and the assumed Caco-2 permeability of valsartan were fit to the equation to test the predictability of negative food effects by the model. When valsartan data was fit to the empirical

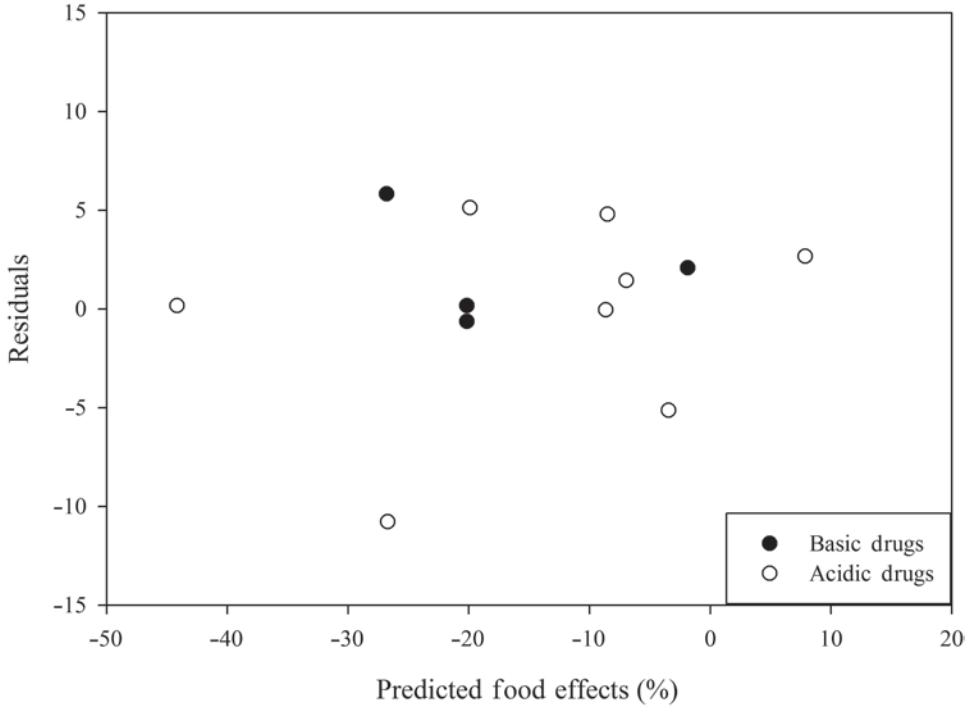


Figure 5. Residual analysis of predicted food effects.

Table 4. Published and predicted food effects of external data set.

| Drug | Published food effects (%) | Predicted food effects (%) |
|---|---|---|
| Valsartan | −40[15,16] | −44.09 |
| Theophylline | +8.24 (non-significant)[19] | −8.17 |
| Timolol | +18.98 (non-significant)[21] | −12.85 |

Table 5. Published and predicted food effects of model compounds.

| Drug | Published food effects (%) | Predicted food effects (%) |
|---|---|---|
| Atenolol | −20[46] | −51.44 |
| Furosemide | −44[50] | −39.44 |
| Nadolol | −20.75[47] | −47.46 |
| Tacrine | −21[51] | −26.86 |

model, the model correctly predicted that valsartan may exhibit negative food effects.

The $K_a$ (or $K_b$) and the published Caco-2 permeability of theophylline and timolol were also fit to the equation to test the predictability of the model. Specifically, the model was checked for giving a false *negative food effect* as an outcome for these *no food effect* drugs. When the published Caco-2 permeability and dissociation constants data for theophylline and timolol were fit to the empirical model, the model correctly predicted that theophylline and timolol may not exhibit negative food effects (Table 4).

### Oral drug absorption model

Applying the determined values of diffusivity, solubility (fed and fasted states), and rat intestinal permeability in the conditions as shown in Table 1, the output parameter (fraction dose absorbed) was simulated for fed and fasted states to calculate the predicted food effect. As shown in Table 5, the published food effects and simulated (predicted) negative food effects showed consistent trends for the model drugs.

## Discussion

Previously published empirical correlations involved statistical testing and correlation of food effects with physicochemical properties, such as solubility, partition coefficient, p$K_a$, and so on, of a large pool of drug molecules[10,11]. The published models did not take the mechanisms involved in food effects into account, and the models cannot distinguish the different food effects exhibited by the molecules. In this study, predictive models specifically for the determination of negative food effects of drugs that are affected in their solubility and permeability properties were investigated. The study included the physiological changes in the gastrointestinal milieu as the mechanism involved in negative food effects exhibited by some drugs.

Since food and administered drugs are predominantly absorbed in the upper small intestine (duodenum and jejunum), changes associated with food intake in the upper intestinal lumen are important causes for negative food effects. Previous investigations from our laboratory indicated that the physicochemical and physiological changes in the presence of food affect the absorption of low permeability drugs more than high permeability drugs[6]. Food may cause negative effects for only poorly absorbed drugs[6]. Such drugs have <90% absorption from

the intestine in fasted state, which may not only be due to limited solubility, but also be due to limited membrane permeability[4,37]. These drugs could be absorbed preferentially in the upper small intestine (region-dependent absorption), and have lower permeability in the lower intestine (e.g. bidisomide and didanosine)[4].

The pH of the upper intestine in the fasted state is about 6.5, whereas the overall postprandial pH after a standard meal in the duodenum is 5.4[23]. The luminal concentration of bile salts is 4–6 mM in the fasted state and 10–20 mM in the fed state[9]. For low permeability drugs, for example, BCS-III and BCS-IV drugs that are chemically stable in the GI tract, if the combined effects of lowered pH and increased bile and lipid digestion products in the fed state cause a significant enhancement of solubility or permeability, then such drugs would exhibit positive food effects or no food effects[6]. If the same combined effects of lowered pH and increased bile and lipid digestion products in the fed state cause a combined effect of decrease in either the solubility or permeability of drugs, then such drugs may exhibit negative food effects[6]. Studies indicated that such negative food effects are exhibited by hydrophilic, highly ionizable, low permeability, acidic (e.g. furosemide) or basic drugs (e.g. atenolol) and whose solubility or permeability is decreased, respectively, in the fed state administration when compared with the fasted state administration[6].

As discussed above, the empirical model indicated that the extent (percent) of negative food effects is dependent on permeability and dissociation constant of molecules. This correlation exemplified that negative food effects are exhibited by low permeability drugs that are likely to be affected by changes in fed versus fasted state intestinal pH conditions, as reflected in the drugs' dissociation constants. Molecules with low $pK_a$ or high $pK_b$ values are highly ionizable; and as a result, in the fed versus fasted state intestinal pH window of 5 to 6.5 the drugs' solubility and permeability properties are affected drastically because of changes in ionization. Molecules with higher $pK_a$ or lower $pK_b$ are relatively weakly ionizable—their extent of ionization does not change significantly in the fed versus fasted state intestinal pH window, as a result, the drugs' permeability or solubility properties are not significantly affected in the fed and fasted state intestinal pH conditions. Therefore, they may not exhibit pH-dependent negative food effects. Non-ionizable molecules in the pH range of the gastrointestinal tract may not exhibit pH-dependent negative food effects.

Even though the Caco-2 permeabilities of the drugs were collected from various sources (our laboratories and literature), consistent relationships between percent food effects and Caco-2 permeability were obtained (Figure 1) when included in the model. This indicated that the predictive model was robust to the inter-laboratory variability of Caco-2 permeability measurements. When the empirical model was challenged with two test molecules that do not exhibit negative food effects, and one molecule that exhibits negative food effects the model correctly predicted the three drugs' food effects. This is a first report that correlates negative food effects and biopharmaceutical properties of structurally diverse molecules.

One of the limitations of this model could be the number of drugs selected for correlation. This was because of the limited number of published drugs exhibiting negative food effects that could not be attributed to either gastrointestinal degradation or complexation. Some new molecules such as bidisomide, avitriptan, and valsartan have been shown to exhibit negative food effects. Bidisomide, a novel antiarrhythmic drug, exhibits negative food effect (more than –27.17%) from high-, medium-, and low-fat meals[38]. One parameter to which this was attributed was its low permeability[38–40]. Bidisomide is a weakly basic molecule with $pK_a$ of 9.3[40] and it was classified as a BCS-III compound, indicating that it is a low permeability molecule[17]. Avitriptan, a new 5-HT1-like agonist, exhibited negative food effects (–35.54%) when co-administered with high-fat, high-carbohydrate, and high-protein meals[41]. It is a weakly basic, incompletely absorbed, and poorly bioavailable drug with a $pK_a$ of 8.0[42]. Valsartan was also reported to exhibit negative food effects (–40%)[15,16]. It is a weakly acidic drug with a $pK_a$ of 3.9[16] that was classified as a BCS-III drug, and exhibited very low permeability across Caco-2 monolayers[17,18]. These three drugs exhibit characteristics that their negative food effects can be correlated by the empirical model, but they could not be incorporated in building of the model because of the lack of their Caco-2 permeability data. Since, the empirical model was built using eight acidic molecules and four basic molecules, this model may be more accurate in predicting the negative food effects of acidic drugs than basic drugs that can potentially exhibit negative food effects. However, this model has shown good predictability for drugs showing no food effects against negative food effects. Another limitation of the model could be that the model predicts the food effects of drugs exhibiting pH-dependent negative food effects, and factors such as formulation-dependent effects on drugs' pharmacokinetics may not be predicted by the model.

Applying the oral absorption model and using the two experimental pH conditions of fed and fasted state intestinal environment (pH 5 and 6.5), the negative food effects were shown to be simulated using an *in vivo* experimental model. The drugs exhibiting negative food effects exhibited statistically significant differences in permeability at pH 5 versus pH 6.7 in rat single pass intestinal perfusion (SPIP) studies (90% CI). When the simulations were run using these values, the predicted food effects matched the trends of published food effects (Table 5) for negative food effect drugs. The deviations of predicted food effects from the published food effects could be due to the vast physiological differences between the intestines of rat and man[43]. Also, the experimental conditions for the *in situ* model were very simplistic when compared with a food effect study in man[5,6].

In summary, for a molecule with known biopharmaceutical properties, the empirical model could be used

to predict the potential of the molecule to exhibit negative food effects. And, simulations of food effects from rat intestinal permeability studies could aid in understanding the probable mechanisms of negative food effects of the compound.

## Conclusions

A predictive model for negative food effects based on the correlation between percent food effects with dissociation constant and Caco-2 permeability was established. Simulations of negative food effects using rat intestinal permeability from SPIP studies at pH 5 and 6.5 supported the drugs' published negative food effects. Thus, an empirical and a mechanistic model as potential tools for predicting negative food effects are reported.

## Declaration of interest

The authors report no declarations of interest.

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