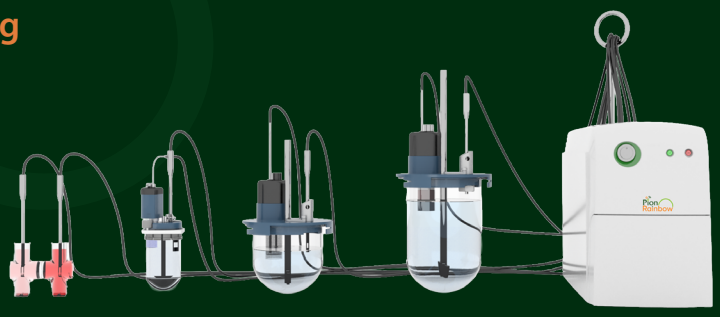
APPLICATION NOTE

Improved prediction of food effect on oral drug absorption

Dissolution-permeation measurements using Pion's MicroFLUX, MiniFLUX, BioFLUX, and MacroFLUX devices provide a mechanistic, clinically predictive approach to understanding food effects, enabling more confident formulation design and decision-making in the development of orally administered drugs.



Pion's MicroFLUX™, MiniFLUX™, BioFLUX™, MacroFLUX™, and Rainbow R6™

Oral administration is the most common route of drug delivery due to the convenience, relative cost-effectiveness, and ease of administration. Whether an oral drug is taken in a fed or fasted state can markedly affect bioavailability and how much of the drug reaches the systemic circulation. The "food effect" describes the extent to which food influences drug absorption and can be positive, negative, or zero.[1] Properties of the active pharmaceutical ingredient (API) and its formulation can also contribute to the food effect.

Dosage instructions regarding food intake in conjunction with orally administered drugs are determined through clinical studies, which assess the impact of food on drug absorption.

A variety of theoretical and empirical prediction schemes for food effects based on the physicochemical properties of drugs have been reported. In addition, various *in vitro* dissolution-permeation experiments have been applied to predict food effects.

This application note describes methods for predicting food effects using *in vitro* dissolution–permeation devices, as reported in peer-reviewed publications. Unlike the use of dissolution data alone, the addition of permeation data can accurately predict positive and negative food effects.

The Bile-Related Food Effect

Gastrointestinal conditions in the fed state differ from those in the fasted state.[2] For example, the concentration of bile micelles is about 5-fold higher in the fed state compared to the fasted state.[3] Bile micelles are tiny, water-soluble aggregates of digested lipids and bile salts that transport fat-soluble nutrients to the intestinal wall for absorption. They affect the dissolution, supersaturation, and precipitation profiles of drugs[4] and impact their effective intestinal permeation.[5]

The higher concentration of bile salts in the fed state aids the solubilization of poorly water-soluble drugs through micelle formation.[6] This solubilization, however, may not always increase oral absorption. Bile micelles may entrap and reduce the free drug concentration available at the epithelial membrane (EPM) surface. Micelle formation may also reduce the diffusion coefficient of the drug in

the unstirred water layer (UWL) adjacent to the epithelial membrane, leading to a decrease in effective permeability.

The food effect depends on the rate-limiting step for absorption. The Fraction absorbed Rate Limiting Step (FaRLS) classification categorizes oral drug absorption into five classes:

- dissolution rate limited (DRL)
- epithelial membrane permeation limited (PL-E)
- unstirred water layer permeation limited (PL-U)
- solubility – epithelial membrane permeation limited (SL-E)
- solubility – unstirred water layer permeation limited (SL-U)

In a bile micelle-containing environment, drug molecules exist as bile micelle-bound and unbound species in rapid equilibrium (Figure 1). The solubility of a drug ($S_{dissolv}$) is the sum of the concentrations of these species ($S_{dissolv,u}$ and $S_{dissolv,b}$) in equilibrium with a solid-state drug. An increase in bile micelles increases $S_{dissolv,b}$ and $S_{dissolv.}$ However, the concentration of unbound (free) drug remains the same ($S_{dissolv,u}$).

Both bile micelle-bound and unbound drug molecules can permeate the UWL adjacent to the EPM, whereas only unbound (free) drug molecules can permeate the EPM (according to free drug theory).

In the case of SL-E drugs, the permeation is not increased even when $S_{dissolv}$ is increased by bile micelles because the free drug concentration remains the same (= $S_{dissolv,u}$). In other words, an increase in $S_{dissolv}$ by bile micelles is cancelled out by a decrease in the "effective" permeation coefficient (P_{eff}), which is defined based on the total drug concentration dissolved in the intestinal fluid. Fraction absorbed (Fa) and flux value (J) are proportional to $S_{dissolv}P_{eff}$.

Various *in vitro* dissolution-permeation experiments have been investigated for food effect prediction. While SL-U drugs were mainly used as model drugs in these studies, little is known about the applicability of these systems for SL-E drugs. One study[7] showed that the food effect on Fa of pranlukast can be appropriately predicted by the dissolution/permeation system; no other studies of SL-E have been reported.

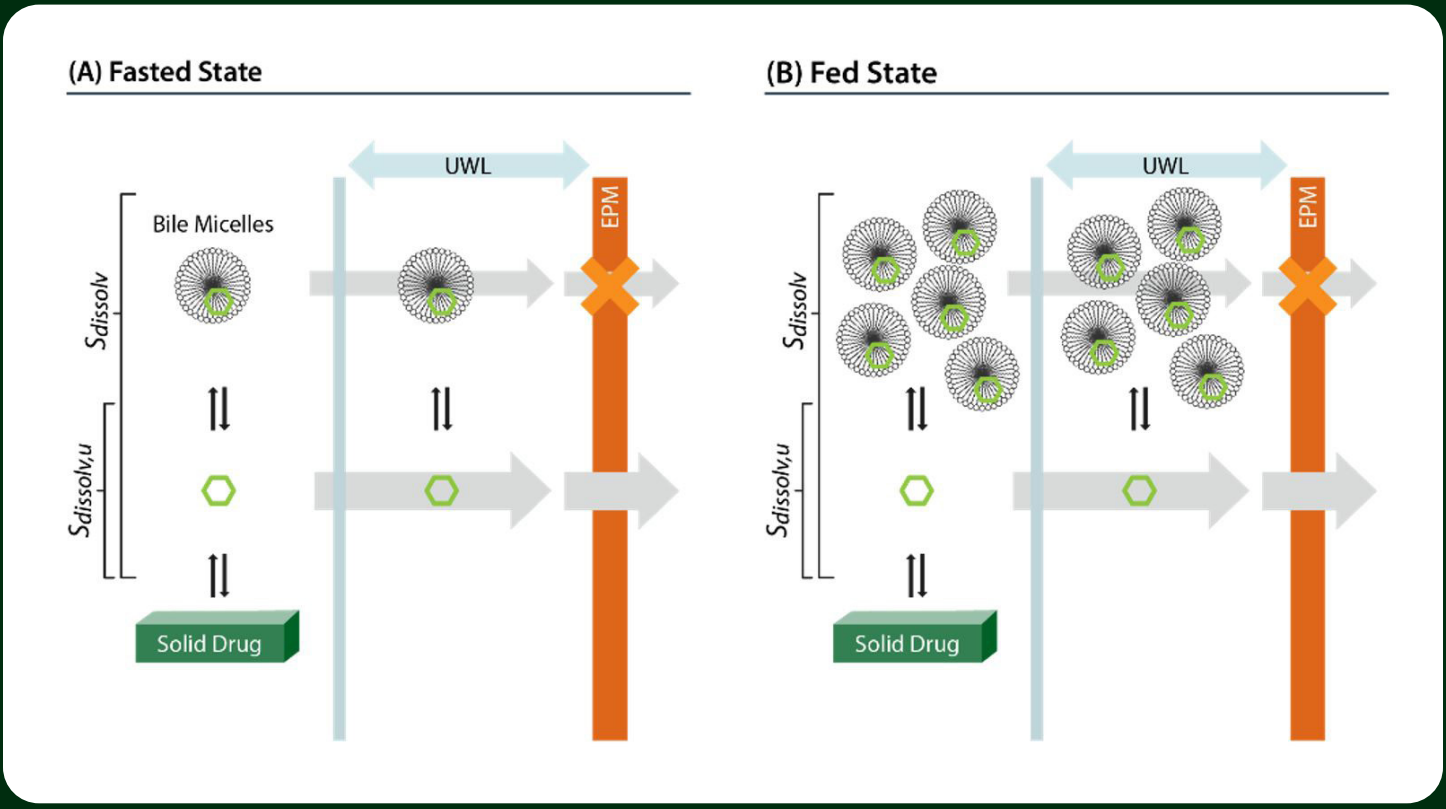


Figure 1. Bile micelle solubilization and permeation in a fasted state (A) and a fed state (B). The width of the grey arrows indicates the permeation coefficient of a drug across the unstirred water layer (UWL) and epithelial membrane (EPM) (dimension: length/ time). For SL-U cases, the overall absorption will increase in Fed State due to the permeation of bile micelles through UWL, while the absorption will remain the same for SL-E cases as the process is rate limited by EPM permeation.

Overview of Experimental Design

A complete description of the materials and methods used in this study, as well as the physicochemical properties of the model drugs, can be found in Higashiguchi, *et al*.[8]

Bosentan, fidaxomicin, pranlukast, and rifaximin were employed as model SL-E drugs while celecoxib and danazol served as model SL-U drugs. Bosentan, fidaxomicin, and rifaximin are beyond-rule-of-five (bRo5) drugs. An increasing number of bRo5 drugs, including over 30% of approved kinase inhibitors, are reaching clinical trials and FDA approval, and about half of small molecules targeting protein–protein interactions fall into the bRo5 category.[9]

The food effect for SL-E drugs was predicted based on FaRLS. Dissolution-permeation experiments performed in a MicroFLUX device were then used to further understand the mechanism of the food effect by bile micelles (Figure 2).

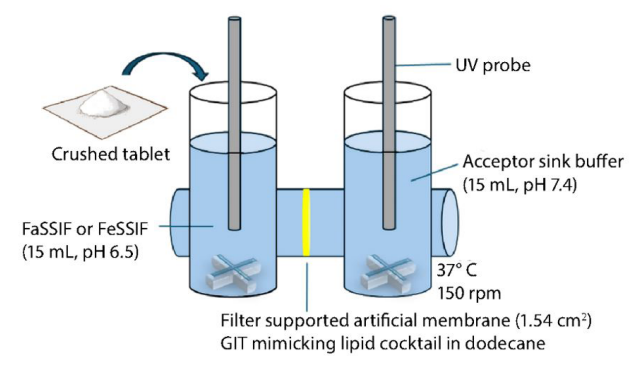


Figure 2. MicroFLUX device used to perform dissolution-permeation experiments

The Pion MicroFLUX is a small-volume, *in vitro* dissolution-permeation apparatus designed to simulate drug absorption by measuring the movement of compounds across a biomimetic lipid-coated artificial membrane, which separates a donor chamber (containing the sample) and an acceptor chamber (representing the blood). It allows for continuous, real-time measurement of drug concentration in both chambers, enabling detailed assessment of drug dissolution, permeation, and the effects of different formulation strategies and excipients.

Fasted and fed state simulated intestinal fluids (FaSSIF and FeSSIF, respectively) were used as the donor solution in the device. Each drug formulation in a biorelevant dose to volume ratio was added after the tablets were gently crushed using a mortar and pestle. The drug concentrations in the donor (C_D) and acceptor (C_A) solutions were monitored using a UV probe for 2 h, except for pranlukast, which was monitored for 4 h. The flux value (J) was calculated from the slope of the drug concentration-time curve in the acceptor solution in the last 30 min.

Results and Discussion

Food effect prediction by solubility ratio, dissolution ratio, and dissolution-permeation results.

Figure 3 compares the fed/fasted ratios of $S_{dissolv}$, $C_{D,final}$, J, and the clinical area under the plasma concentration–time curve (AUC). Sdissolv data (dark green) represents the ratio of the solubility measured in fed and fasted conditions. The $C_{D,final}$ (orange) is the ratio of the donor concentration in the dissolution-permeation assay measured in fed and fasted conditions. J (light green) is the flux through the membrane of the MicroFLUX. The dissolved drug concentration in the donor fluid (C_D) was significantly increased in FeSSIF compared to FaSSIF in all cases. In the case of SL-E drugs, absorption was lower or less affected.

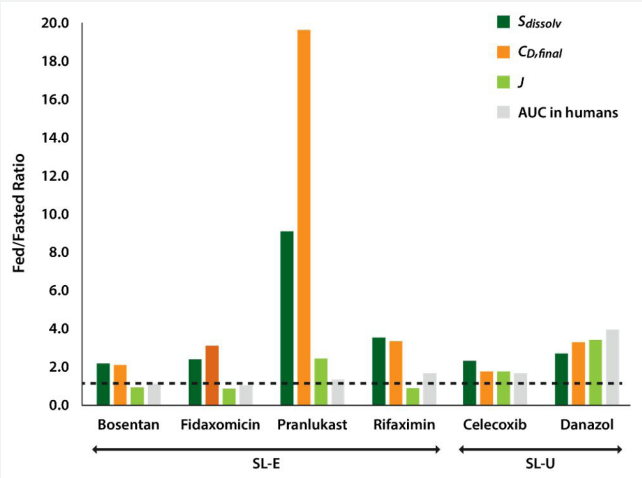


Figure 3. Comparison of the fed/fasted ratios of $S_{dissolv}$, $C_{D,final}$, J, and clinical area under the plasma concentration–time curve (AUC). The dotted line is the predicted Fa ratio for SL-E (ratio = 1.2). All data are ratios of a particular parameter measured under a fed versus fasted condition. $S_{dissolv}$ data (dark green) represents the ratio of the solubility measured in fed and fasted conditions. The $C_{D,final}$ (orange) is the ratio of the donor concentration in the dissolution-permeation assay measured in fed and fasted conditions. J (light green) is the flux through the membrane of the MicroFLUX. The ratio of $C_{D,final}$ can therefore be higher than the $S_{dissolv}$ value.

In the case of rifaximin, significant supersaturation was observed in the dissolution data in the early time points. However, there was little difference in absorption between FaSSIF and FeSSIF, suggesting that the unbound drug concentrations in FaSSIF and FeSSIF were the same in the supersaturated state. In the case of SL-U, the increase in C_D in the fed state condition translated to an increase in absorption, in good agreement with previous findings.

Overall, these findings highlight that reliance solely on solubility data can over-predict the food effect, while predictions based on dissolution-permeation data are more closely aligned with the *in vivo* results (AUC).

Large-Scale Dissolution/Permeation Studies

In addition to the smaller volume MicroFLUX and MiniFLUX devices, which are typically used in earlier development, large volume dissolution-permeation devices such as the BioFLUX and MacroFLUX are well-suited for later-stage development where full tablets or capsules can be used in the assay without having to be crushed.

A combined dissolution-permeation evaluation using the full-scale MacroFLUX device to estimate the direction and extent of food effects observed in the clinical setting for several BCS Class II and Class IV drugs with different underlying food effect mechanisms was published by Novartis in 2025.[10]

BCS Class II drugs, typically falling into the SL-U class, have high intestinal permeability, meaning they can easily pass through cell membranes, but low aqueous solubility, meaning they do not dissolve readily in water. Class IV drugs, typically falling into the SL-E class, have low aqueous solubility and low intestinal permeability, making oral delivery relatively challenging.

In the published study, food effects were accurately predicted for 60% of drugs within 1.25-fold based on dissolution-permeation analysis, compared to 30% when only dissolution analysis was used, as summarized in Table 1. Of the ten compounds evaluated, only five displayed the same direction of food effect compared to clinical studies based on dissolution results alone, while nine compounds aligned with the direction based on dissolution-permeation data analysis. Use of dissolution alone did not identify any of the negative food effects. The study also showed that the assumption that compounds exhibiting a positive food effect due to increased dissolution/solubility from fasted to fed state does not always hold.

Upon further analysis of the Remibrutinib data, it was found that there was no clinically relevant food effect observed for Remibrutinib due to a marginal fed/fasted ratio. Dissolution-permeation results predicted a negative food effect with a fasted/fed ratio of 0.53, while the dissolution results predicted a significant positive effect (4.08). Based on this finding, the study authors conclude that the predictions from dissolution-permeation studies were closer to clinical data compared to dissolution results. Overall, the MacroFLUX device was confirmed to be a valuable tool that can predict clinical food effects, support formulation development, and guide the design of clinical pharmacology studies.

Table 1. Assessment of the direction of the food effect for the ten drugs evaluated in the study.

Compound	*In vivo* food effect		Prediction accuracy	
API	**C_{max} Ratio**	**AUC Ratio**	**$C_{dissolution}$ Ratio**	**Flux Ratio**
Asciminib	Negative	Negative	✗	✓
Dabrafenib	Negative	Negative	✗	✓
Eltrombopag	Negative	Negative	✗	✓
Iptacopan	None	None	✓	✓
Lapatinib	Positive	Positive	✓	✓
Nilotinib	Positive	Positive	✓	✓
Palbociclib	Positive	Positive	✓	✓
Remibrutinib*	Positive	None	✗	✗
Ribociclib	None	Positive	✓	✓
Trametinib	Negative	Negative	✗	✓

Positive: Fed to Fasted ratio more than 1.25, None: Fed to Fasted ratio between 0.08 and 1.25, Negative: Fed to Fasted ratio less than 0.08.
**Dissolution testing of Remibrutinib predicted a strong positive food effect, while dissolution-permeation analysis revealed a negative effect that aligned much more closely with clinical results showing no clinically meaningful food effect.*

Conclusion

The data described above demonstrate that evaluating both dissolution and permeation is essential for accurately predicting food effects, particularly for SL-E and SL-U drugs. FaRLS theory provides the mechanistic basis for understanding why increased solubility in the fed state does not always result in increased absorption because, although bile micelles can enhance total solubility, they often reduce the free drug fraction available to cross the epithelial membrane. This balance explains why dissolution data alone may suggest a positive food effect, while dissolution-permeation measurements more closely reflect clinical outcomes.

The Remibrutinib case clearly illustrates this distinction. Dissolution testing predicted a strong positive food effect, yet dissolution-permeation analysis revealed a marginal negative effect that aligned much more closely with clinical results showing no clinically meaningful food effect. This example underscores the limitations of relying solely on dissolution data and highlights the value of incorporating permeation into predictive models.

By coupling mechanistic theory with experimental systems such as Pion's MicroFLUX, MiniFLUX, BioFLUX and MacroFLUX devices, drug developers can gain a more accurate, clinically relevant understanding of food effects. This integrated approach not only reduces the risk of an incorrect prediction but also supports formulation optimization, improves the design of clinical pharmacology studies, and ultimately helps accelerate the development of orally administered therapies, including challenging bRo5 compounds. As demonstrated here, dissolution-permeation analysis is a powerful complement to dissolution testing, providing a predictive framework that bridges *in vitro* studies with *in vivo* outcomes and strengthens decision-making throughout the drug development process.

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Pion's MiniFLUX system can be used to scale up dissolution-absorption studies, with volumes up to 250 mL