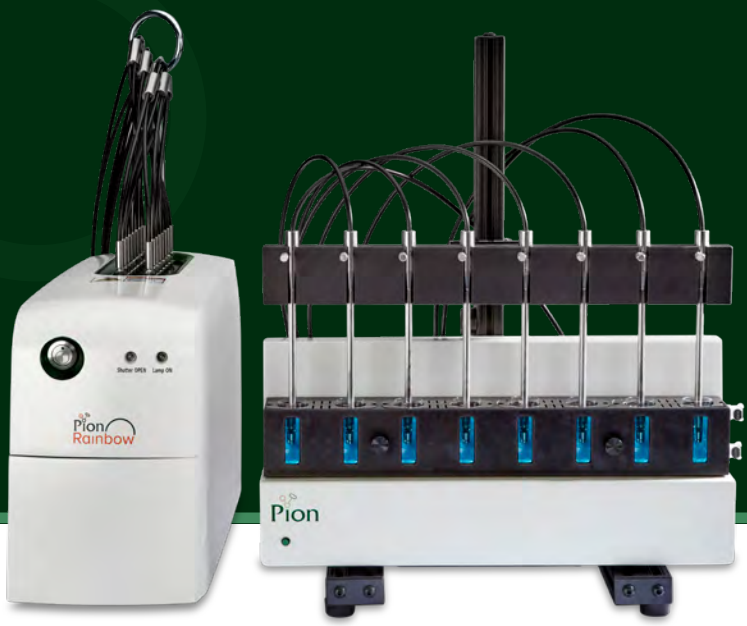
APPLICATION NOTE

Evaluating Amorphous Solid Dispersions Using Dissolution-Permeation Assays

Poor aqueous solubility remains one of the most persistent challenges in oral drug development, particularly for Biopharmaceutics Classification System (BCS) Class II molecules where absorption is limited by solubility.[1]



Pion's Rainbow R6™ and MicroFLUX™

Improving the ability of Class II molecules to dissolve and be absorbed in the body is essential for their successful development as effective medications. Formulation strategies such as amorphous solid dispersions (ASDs) are an important tool for increasing the solubility of poorly soluble active pharmaceutical ingredients (APIs) and improving their oral bioavailability.

Hot melt extrusion (HME) is a robust and scalable manufacturing technology for producing ASDs. In this process, the API is molecularly dispersed in a polymer matrix under controlled heating and shear conditions, resulting in an amorphous form of the drug embedded within the carrier. Compared to the crystalline API, ASDs typically dissolve more rapidly and can generate drug concentrations that exceed the equilibrium solubility of the crystalline form. Achieving this state of supersaturation enhances absorption in the gastrointestinal (GI) tract.

The polymers used in ASDs facilitate rapid dissolution of the amorphous API and act as precipitation inhibitors. By delaying crystallization, these polymers help maintain supersaturation for longer periods, thereby extending the window for absorption and improving bioavailability. Once precipitation occurs, however, the solubility advantage of the amorphous form is lost.

As various formulations of the same API may generate similar peak concentrations but exhibit different precipitation behaviors and ultimately, *in vivo* behavior, it is essential to use kinetic solubility studies to track concentration as a function of time and identify the onset of precipitation. These measurements provide insight into how formulation composition, dose, and biorelevant media influence the extent and duration of supersaturation.

This application note describes two experimental approaches for studying supersaturation and precipitation from ASD formulations using both dissolution-only and combined dissolution–permeation assays.

In the first study, ASDs of the poorly soluble APIs piroxicam and loratadine were prepared by HME, and their supersaturation behavior was characterized in biorelevant media using the MicroDISS Profiler™ which consists of a small volume dissolution bath and the Rainbow R6 fiber optic spectrometer. Real-time concentration monitoring enabled direct measurement of kinetic solubility and precipitation onset under physiologically relevant conditions.

Supersaturation Behavior of ASDs of Loratadine and Piroxicam

ASDs of the poorly soluble model compounds loratadine and piroxicam were prepared by HME using Plasdone™ S-630 copovidone as the polymer carrier. Extrudates were prepared at API loadings of 30% and 40% across a range of processing temperatures (120–180°C). The resulting powders were characterized by X-ray powder diffraction (XRPD) and polarized light microscopy to assess solid-state form. All formulations were confirmed to be amorphous, independent of drug loading or extrusion temperature, demonstrating that stable ASDs could be reliably produced for both APIs using this approach.

Dissolution behavior and supersaturation profiles of the ASDs were evaluated using the MicroDISS Profiler™. Loratadine ASDs were evaluated in fasted-state simulated intestinal fluid (FaSSIF), while piroxicam ASDs were tested in fed-state simulated intestinal fluid (FeSSIF), reflecting the physiologically relevant environments for these compounds.

Figure 1 shows the dissolution of crystalline loratadine in FaSSIF. Equilibrium was reached after 1 hour of dissolution indicating solubility more than 20 times higher in FaSSIF than in a corresponding aqueous buffer at pH 6.5 (data not shown).

The loratadine ASDs exhibited different dissolution behaviors compared to crystalline loratadine powder. The majority of extrudates reached supersaturation followed by a slight decrease in concentration in the studied timeframe (Figure 2). Kinetic solubility of all formulations resulted in up to a 2-fold higher solubility than that of crystalline loratadine, regardless of extrudate preparation temperature.

Samples prepared using 150/130°C processing temperatures were underperforming, while the 140°C samples had the highest supersaturation.

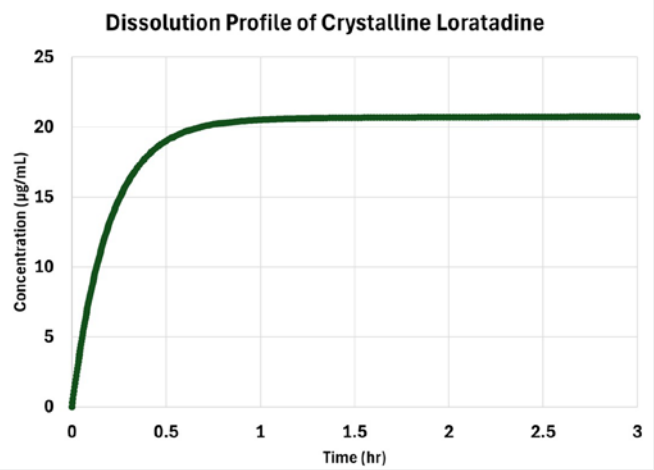


Figure 1. Dissolution profile (µg/mL versus hours) of 3mg of crystalline loratadine in 10mL FaSSIF at 25°C and 200 RPM stirring speed.

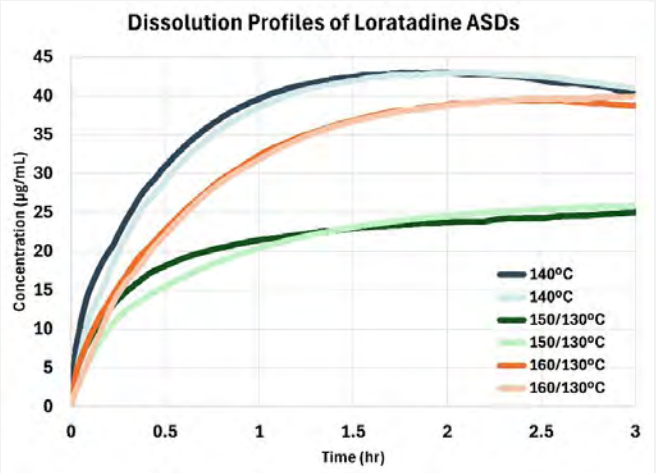


Figure 2. Dissolution profiles (µg/mL versus hours) of loratadine ASDs with 40% API load prepared under different temperature conditions. Data collected in FaSSIF at 25°C and 200 RPM stirring speed.

Piroxicam exhibited more complex supersaturation behavior in the study. In $FeSSIF_{blank}$ media, crystalline piroxicam approached its solubility limit (~20 µg/mL) within 20 minutes whereas the ASD dissolved rapidly to concentrations of ~70 µg/mL within 6 minutes (Figure 3).

Following the addition of a second dose of ASD into the dissolution vessels, the amorphous piroxicam reached a fully dissolved state at approximately 150 µg/mL and remained supersaturated. Only a third additional dose forced precipitation after the dissolved concentration

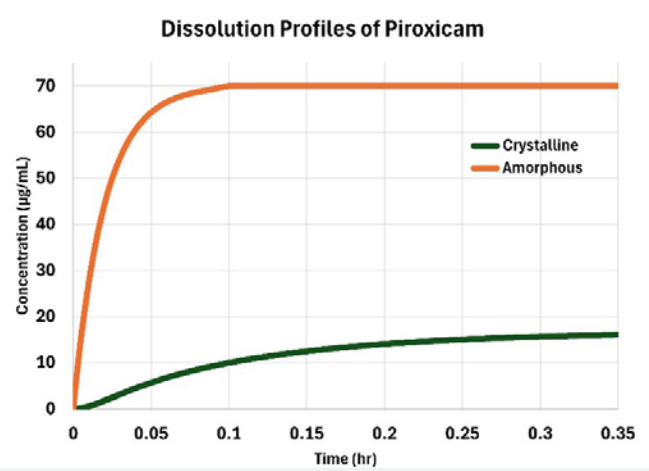


Figure 3. Dissolution profiles (µg/mL versus hours) of piroxicam and its holt melt extrudate with Copovidone in FeSSIF$_{blank}$.

exceeded 200 µg/mL, a solubility 10 times higher than that of crystalline piroxicam (Figure 4).

Figure 5 shows a comparison of the dissolution/precipitation profiles of crystalline piroxicam and its hot melt extrudates in FeSSIF$_{blank}$ during the first five hours of the experiment (>25 hours). The two samples that received a single dose of amorphous piroxicam remained supersaturated at 70 µg/mL for the duration of the experiment while the samples that received three doses of ASD reprecipitated (as shown in Figure 4), falling out of the supersaturation phase and reaching equilibrium at the

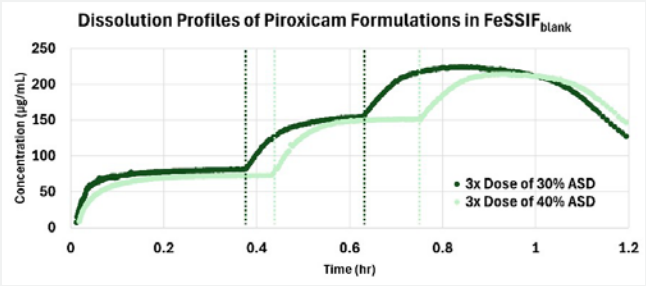


Figure 4. Dissolution/precipitation profiles (µg/mL versus hours) of piroxicam hot melt extrudates showing stepwise addition of a second and third dose of ASD into the aqueous buffer (FeSSIF$_{blank}$).

solubility level of crystalline piroxicam.

These experiments clearly demonstrate the existence of a critical supersaturation window; below this threshold, supersaturation can be maintained for prolonged durations, while exceeding it results in rapid loss of the solubility advantage that the ASD provides.

When the experiments were repeated in the complete biorelevant FeSSIF, crystalline piroxicam exhibited approximately two-fold higher solubility compared to buffer alone, while the kinetic solubility of amorphous piroxicam reached up to 450 µg/mL (Figure 6). However, this spike in the kinetic solubility was quickly followed by reprecipitation of the ASDs within the first 30–45 minutes, with equilibrium again being reached at the same solubility as crystalline piroxicam.

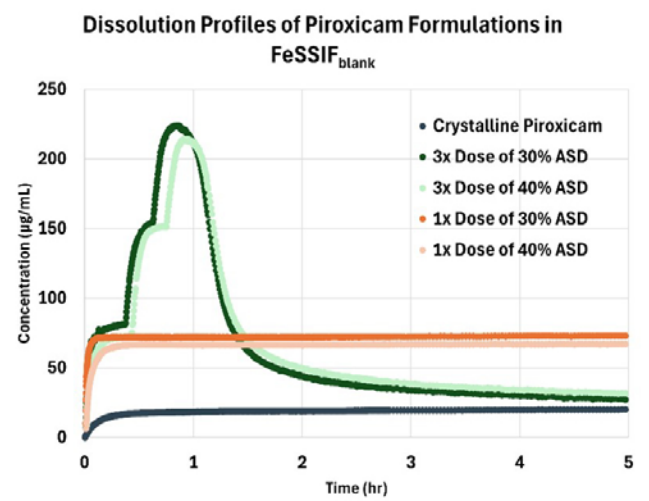


Figure 5. Dissolution/precipitation profiles (µg/mL versus hours) of crystalline piroxicam and its hot melt extrudates in aqueous buffer (FeSSIF$_{blank}$).

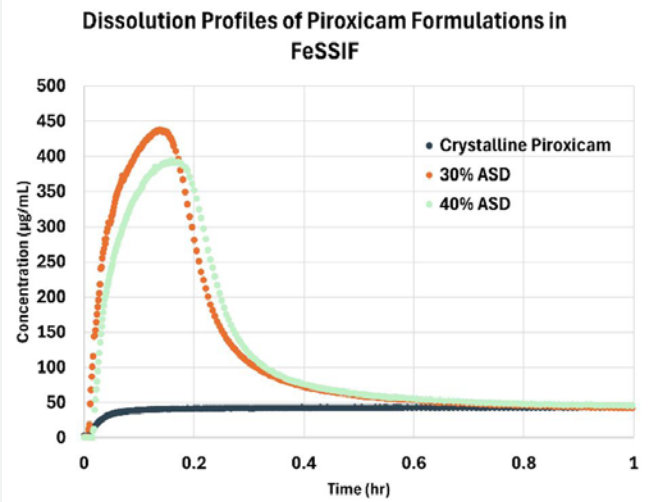


Figure 6. Dissolution/precipitation profiles (µg/mL versus hours) of crystalline piroxicam and its hot melt extrudates in FeSSIF.

These results demonstrate that hot melt extruded ASDs of loratadine and piroxicam can reliably generate supersaturation under biorelevant conditions, but the magnitude and persistence of that supersaturation depend strongly on formulation variables, dose, and media composition. The data illustrates the existence of a formulation-specific critical supersaturation window, below which elevated concentrations can be sustained and above which rapid precipitation erodes the solubility advantage. Real-time kinetic solubility monitoring with the MicroDISS Profiler™ provides essential insight into these dynamic dissolution–precipitation processes, enabling more informed optimization of ASD formulations before advancing to permeability or *in vivo* studies.

Why Dissolution Alone Is Not Enough: The Need to Measure Permeability

The dissolution studies described above clearly demonstrate the ability of ASDs to generate and, in some cases, maintain supersaturation in biorelevant media. Real-time concentration monitoring enables direct measurement of kinetic solubility, identification of critical supersaturation thresholds, and characterization of precipitation behavior, all key parameters for understanding formulation performance. However, while dissolution data are necessary, they are not always sufficient to predict *in vivo* drug absorption.

Supersaturation is valuable only to the extent that it translates into increased drug permeation across the intestinal epithelium. In the gastrointestinal tract, absorption is governed by the dynamic interplay between dissolution and absorption. A formulation that generates high peak concentrations may still provide limited *in vivo* benefit if rapid precipitation occurs before the drug can permeate the intestinal membrane, or if chosen excipients lower the permeation. Conversely, a formulation that maintains a lower level of supersaturation for a longer duration or with favorable excipient matrix may achieve greater overall absorption by sustaining higher drug permeation over time.

This limitation becomes particularly important for supersaturating systems, where concentration in the lumen can change rapidly due to dissolution and precipitation. Under these non-steady-state conditions, absorption rate is not constant but instead varies as a function of donor concentration and excipient matrix. As a result, dissolution experiments alone, regardless of how well supersaturation is characterized, cannot fully capture the absorptive performance of an ASD formulation.

To address this gap, dissolution must be coupled with permeability measurements that directly quantify transmembrane permeation. Simultaneous monitoring of donor concentration and drug appearance in an acceptor compartment enables direct assessment of how supersaturation behavior impacts permeability. In this context, parameters such as permeation profiles and area under the acceptor concentration–time curve provide more meaningful indicators of formulation performance than dissolution data alone.

Dissolution–Permeation Analysis of Meloxicam ASDs

The objective of this study was to evaluate how differences in supersaturation behavior among ASDs of meloxicam translate into transmembrane flux in the dissolution-permeation device.

ASDs of meloxicam were prepared using HME with several commonly used pharmaceutical polymers, including Soluplus®, Kollidon® VA 64, and a Kollidon® VA 64/Kolliphor® TPGS blend:

· 15% Meloxicam/85% Soluplus
· 15% Meloxicam/85% Kollidon VA64
· 15% Meloxicam/72.5% Kollidon VA64/12.5% Kolliphor TPGS

Extrusion was performed using a co-rotating twin-screw extruder under controlled processing conditions to ensure consistent formulation quality.

Solid-state characterization (XRD and DSC, not shown) confirmed that the Soluplus formulation was fully amorphous, while the Kollidon VA 64 formulation contained some residual crystallinity. These differences provided a useful basis for comparing how polymer selection influences supersaturation stability and downstream absorption.

Dissolution and permeability experiments were conducted using the MicroFLUX™ system, an add-on to the MicroDISS Profiler™ that integrates side-by-side donor and acceptor compartments separated by a lipid-coated, artificial membrane. This configuration allows real-time monitoring of drug concentration in both compartments, enabling simultaneous assessment of dissolution, precipitation, and transmembrane permeation.

A gastrointestinal-optimized artificial membrane (GIT-1 lipid solution) was used to model passive diffusion. The donor compartment contained simulated gastric fluid,

while the acceptor compartment was filled with acceptor sink buffer (pH 7.4). Fiber-optic UV probes continuously monitored concentration changes in both compartments throughout the experiment.

Dissolution and Supersaturation Behavior of Meloxicam Formulations

Initial dissolution experiments showed that crystalline meloxicam exhibited very low solubility in simulated gastric fluid (SGF) media (~0.7 µg/mL). In contrast, all ASD formulations rapidly generated supersaturated solutions, with the extent and duration of supersaturation depending strongly on polymer composition and formulation load (Figure 7).

The Soluplus ASD achieved high levels of supersaturation and maintained this state for extended periods, exhibiting classic "parachute" behavior with slow precipitation. In comparison, the Kollidon VA 64 and VA 64/TPGS formulations also generated supersaturation but precipitated more rapidly following the initial concentration peak.

These results highlight an important distinction: formulations that achieve similar peak concentrations may differ substantially in how long supersaturation is maintained, which can directly impact *in vivo* absorption.

The dissolution-permeation experiments were performed with higher meloxicam loading that resulted in higher peak concentrations but similar supersaturation-precipitation trends (Figure 8). The Soluplus formulation stayed supersaturated for over 16 hours of the experiment

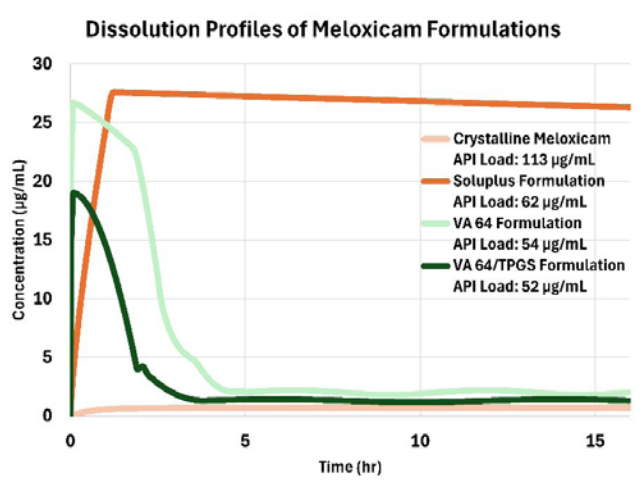
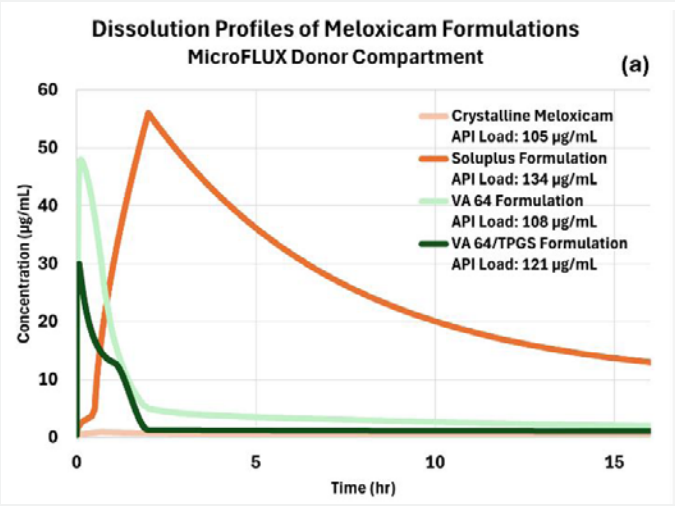


Figure 7. Examples of dissolution/precipitation profiles (µg/mL versus hours) for crystalline meloxicam, Soluplus formulation, VA 64 formulation and VA 64/TPGS formulation. Assays were performed in 20mL of SGF medium.

while both Kollidon VA64 and Kollidon VA64/Kolliphor TPGS formulations precipitated (at different rates) after an initial supersaturation phase.

Except for untreated meloxicam, where permeation was low but almost constant over the course of the experiment, the permeation of ASD formulations varied drastically depending on their supersaturation–precipitation profiles in the donor compartment. The initial permeation was faster for the Kollidon VA64 and Kollidon VA64/Kolliphor TPGS formulations, but the Soluplus



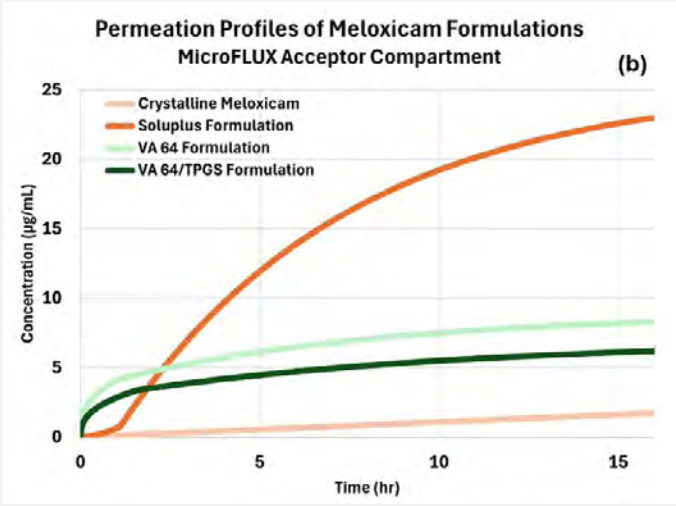


Figure 8. Simultaneous dissolution-permeation profiles of meloxicam (µg/mL versus hours) in donor (a) and acceptor (b) compartments in the MicroFLUX.

formulation maintained a high permeability for a longer period.

To better capture overall absorption potential under these dynamic conditions, the area under the acceptor concentration–time curve (AUC) was evaluated (Figure 9). Acceptor AUC integrates permeation over time and provides a single, absorption-relevant metric that reflects both the magnitude and duration of drug transport. Using this metric, the Soluplus formulation demonstrated superior overall transport despite not having the highest initial permeability, underscoring the importance of sustained supersaturation rather than peak concentration alone.

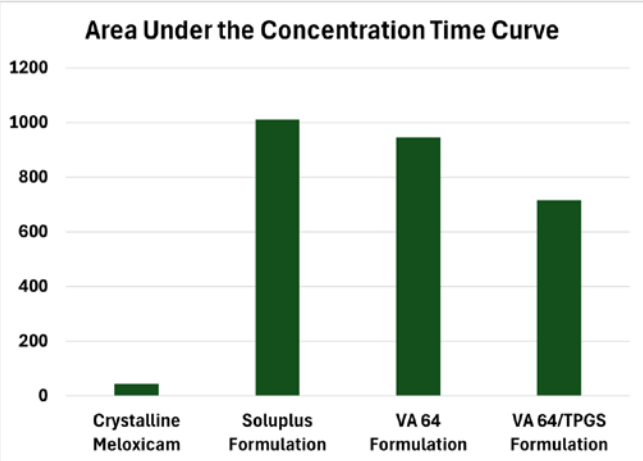


*Figure 9. Area under the concentration versus time curves ((ug/mL)*min) in the acceptor compartments for crystalline meloxicam and its ASD formulations.*

This study underscores that dissolution alone cannot predict *in vivo* performance for supersaturating ASD formulations. Supersaturation drives absorption, but only while it is maintained with favorable excipients. Time-resolved permeation and acceptor AUC offer a clearer picture of absorption potential, while polymer choice is key to balancing stability and transport. By measuring dissolution and permeability together, the MicroFLUX™ platform links supersaturation behavior directly to absorption outcomes, guiding smarter formulation decisions.

Conclusion

ASDs produced by HME are a common strategy for improving the apparent solubility of poorly water-soluble drugs. By generating supersaturated solutions that exceed the equilibrium solubility of the crystalline API, ASDs provide a great option to enhance oral bioavailability. However, overall absorption enhancement depends not only on how much drug dissolves, but on how long that supersaturation can be maintained and whether it translates into effective absorption.

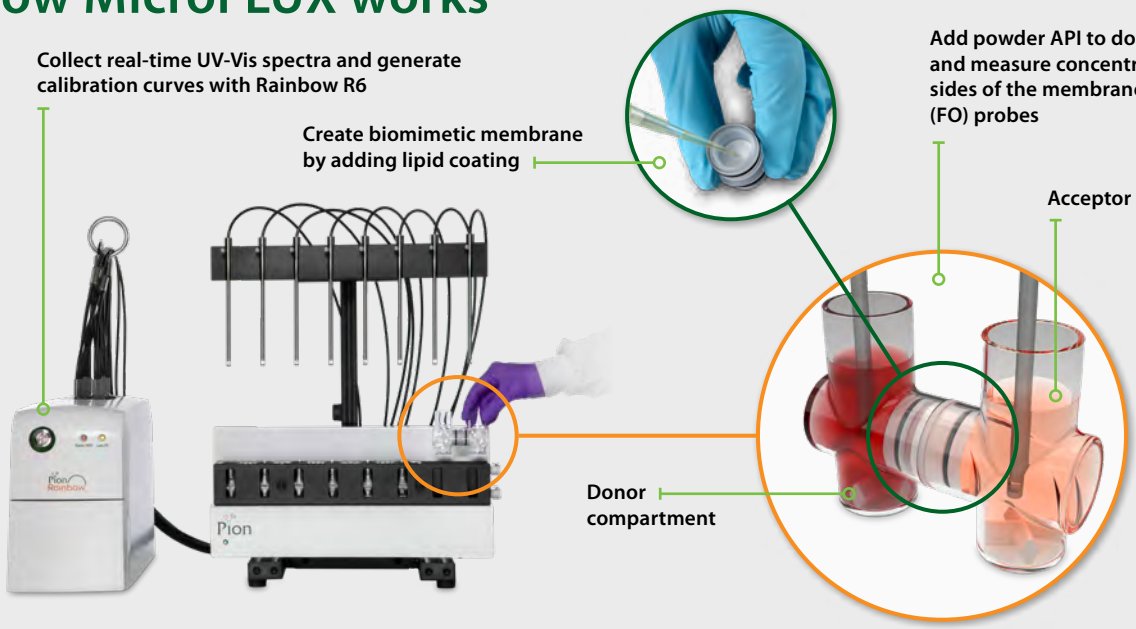
The studies presented in this application note demonstrate that formulation performance cannot be reliably assessed using equilibrium solubility or dissolution endpoints alone. Real-time dissolution experiments with loratadine and piroxicam showed that ASDs can generate markedly different supersaturation and precipitation profiles depending on API properties, dose, polymer selection, and biorelevant media. Critically, these experiments revealed the existence of formulation-specific critical supersaturation thresholds, beyond which rapid precipitation erodes the concentration advantage that ASDs are designed to provide. Measuring kinetic solubility and precipitation onset is therefore essential for understanding and optimizing supersaturating formulations.

The meloxicam dissolution–permeation study further underscores that supersaturation alone is not sufficient to predict *in vivo* performance. When supersaturation is transient, high peak concentrations do not necessarily lead to improved absorption. Instead, sustained supersaturation that supports continuous transmembrane flux is a more meaningful determinant of bioavailability. By simultaneously measuring dissolution, precipitation, and permeability, the MicroFLUX™ approach captures the dynamic interplay between solubility and permeability that governs absorption under non–steady-state conditions.

Taken together, these results highlight a clear progression in formulation assessment: From measuring supersaturation generation, to identifying precipitation kinetics, and ultimately quantifying absorption-relevant permeation. Integrating real-time dissolution and permeability measurements provides a more mechanistic and predictive framework for evaluating ASD performance, reducing reliance on the assumption that dissolution behavior alone reflects *in vivo* outcomes.

As the industry continues to advance increasingly complex and poorly soluble drug candidates, such integrated experimental approaches will be critical for de-risking formulation development, guiding polymer and dose selection, and improving confidence in bioavailability predictions.

How MicroFLUX works



The proprietary lipid-coated artificial membrane and acceptor sink buffer establish a biometric model of passive intestinal transport, incorporating unstirred water layer effects. The membrane technology, adapted from Pion's PAMPA platform, supports real-time permeation monitoring under biorelevant conditions.

Reference

1. Samineni R, Chimakurthy J, Konidala S. Emerging Role of Biopharmaceutical Classification and Biopharmaceutical Drug Disposition System in Dosage form Development: A Systematic Review. *Turk J Pharm Sci.* 2022 Dec 21;19(6):706-713. doi: 10.4274/tjps.galenos.2021.73554.

Small Volume Dissolution

The microDISS Profiler system consists of a Rainbow R6 and the MB8 small volume dissolution bath. It allows testing in media volumes as low as 2 mL to 20 mL. With small amounts of API to run a dissolution study, the MicroDISS Profiler allows 100X less volume needed than a traditional USP apparatus.

MicroDISS Profiler system