Contact us for more information
cls-admet.services@conceptlifesciences.com

CONCEPT LIFE SCIENCES

Concept Life Sciences: **Solubility**

Introduction

Solubility is one of the most important properties in drug discovery. Low-solubility can lead to poor absorption and bioavailability after oral dosing, insufficient solubility for IV dosing, artificially low activity values or erroneous results in different biochemical and functional assays. Low solubility is often associated with high plasma protein binding, slow tissue distribution, and drug-drug interactions. Increased time and resources are required with expensive formulations to develop a poorly soluble drug candidate. Concept use the shake-flask method for either kinetic or thermodynamic solubility depending on your preference.

Thermodynamic solubility: Dry solid compound is used, where particle size and crystal morphology influence the amount solubilized. Results provide important information for PK formulations, pH adjustment and co-solvent being two of the most used methods to solubilize poorly soluble drug compounds during later stages of development.

Deliverable: Aqueous solubility concentration in µM.

Kinetic solubility: The solubility of a given compound can be influenced by the presence of DMSO. Concept kinetic solubility determination uses a DMSO stock solution to mimic early in-vitro biological testing because test solutions are derived from the same DMSO stock solution which is the storage format for most compounds in early development. The solutions in these early screening assays inevitably contain small amounts of DMSO.

Deliverable: Aqueous solubility concentration in µM.

Customer provides

Compound identifier and molecular formula.

Thermodynamic: 2 x 0.5mg solid.
Kinetic: 0.5mg solid or 50µl 10mM DMSO stock.

Format

1.5mL glass vials. Aqueous pH7.4 phosphate buffer 0.1M (or customer specific). Orbital shaking incubator at 25ºC.

Protocol

Buffer: pH7.4 phosphate buffer (0.1M), or multiple pH on request, as well as FaSSIF, FeSSIF, simulated gastric fluids.

1mg (**thermodynamic**) or 10µL of 10mM DMSO stock (**kinetic**) of test compound is added to 1000µL and 990uL of buffer respectively in a glass vial. The vial is sealed, and shaken on an orbital mixer for (1) 24hr to reach equilibration for **thermodynamic**, or (2) 1hr for **kinetic**. After the mixing time, the solution is centrifuged (3000rpm at Room Temp for 20mins). The supernatant is removed to a second vial and centrifuged again. After the second spin, two samples are prepared from the supernatant at different concentrations – a high and a low dilution (since concentrations are unknown) in order to prepare for LC-MSMS quantitation against a standard curve. An internal standard is utilized in the final dilution for analysis.

Controls

Nicardipine.

Quantitation

Sample supernatants are analyzed by LC-MS/MS using a 4-point standard curve prepared from 10mM DMSO stock.

Data analysis and results

For each test compound injection:

$$\text{Response ratio} = \frac{\text{Test peak area}}{\text{Internal standard peak area}}$$

Test compound response ratio is converted to concentration (µM) from the standard curve.

Concept thermodynamic solubility