



Physicochemical characterization



Summary

Physicochemical strategies focus on the determination and interpretation of solubility, lipophilicity and related molecular properties as factors and predictors of pharmacokinetic behavior to speed up drug discovery and drug development processes.

The major reason for this trend is the fact that in the pharmaceutical industry the most successful drug is quite often not the most potent one but rather the one that has the optimum balance of suitable potency, safety, pharmacokinetics for mutation, drug-drug-interactions and manufacturing costs.

Across Barriers offers its clients a comprehensive range of physicochemical services to characterize drug substances. All work performed complies fully with international guidelines and regulations (GLP, ICH, FDA).

Introduction

By using combinational chemistry and high throughput technologies bioactive compounds are discovered by thousands. But these techniques tend to shift leads towards

- more lipophilic and therefore potentially less soluble compounds and
- compounds with a higher number of hydrogen bond donors and acceptors and larger molecular volume ¹.

To shorten the time-consuming development and high rate of attrition of active compounds drug research is coming to incorporate structure-permeation, structure-distribution, structure-metabolism and structure-toxicity relations into drug-design strategies.

The challenge remains to find the proper combination of various techniques of physicochemical characterization to pick the right compounds quickly while keeping the risk of eliminating “good” candidates as low as possible.

It is known for a long time now that solubility and permeability are the driving forces for a successful drug development.

The intrinsic physicochemical properties pKa, log P and lipophilicity parameters such as KIAM (distribution coefficient using artificial membranes bound to chromatographic stationary phases) and parameters like molecular size and shape determine the behavior of molecules with respect to solubility, permeability and stability.

Relations between drug disposition and related physicochemical parameters are shown in fig. 1.

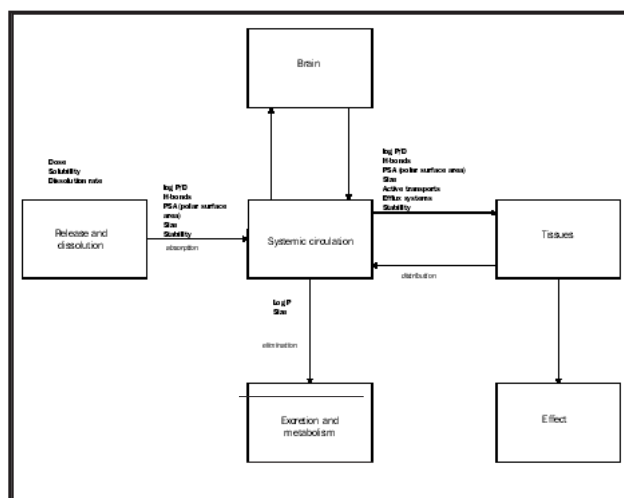


Figure 1. Drug disposition and related physicochemical parameters

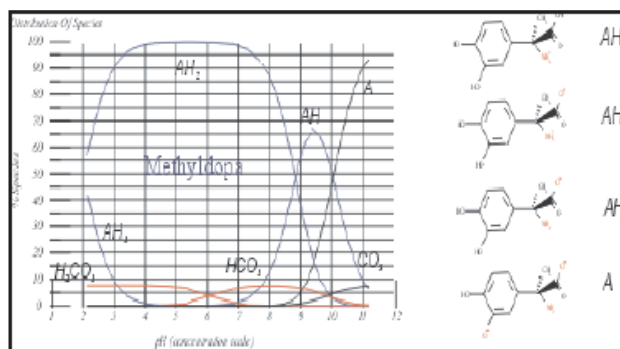
Physicochemical profiling

Determination of logP

Measuring logP quantifies the partitioning of a neutral (unionized) substance between water and 1-octanol. At across Barriers, logP measurements are performed by potentiometric titration which permits measurements to be made across the entire pH range. In addition, this technique enables measurements to be performed on ampholytic compounds or on substances which tend to form microemulsions in the conventional shake flask method.

Determination of the ionization constant (pKa)

Using potentiometric titration (Sirius GlpKa) or spectroscopic detection (Sirius D-PAS diode array detector), we are able to measure ionization coefficients of sample weights down to 0.5 mg. It is also possible to measure multiple overlapping pKa values with ionization constants ranging from 1.5 to 12. Water-insoluble compounds can also be analyzed by adding suitable cosolvents (e.g. methanol). The desired pKa value is then obtained by extrapolation to the zero cosolvent fraction region. Data can also be presented in an appropriate graphical form, for example when displaying the pH-dependent distribution of various ionic species.

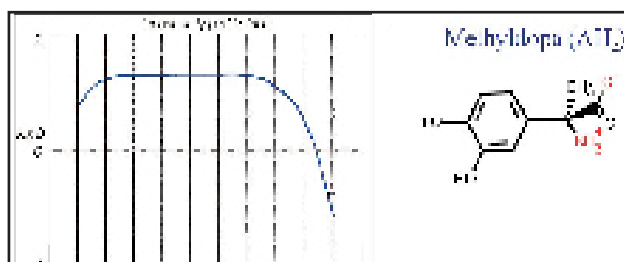


Determination of logD

Determining the lipophilicity profile is a key factor controlling the in vivo behavior of substances. Lipophilicity has a major effect on both pharmacokinetic measurements (permeation through physiological membranes, plasma protein bonding) and pharmacodynamic investigations (receptor affinity and receptor specificity) of a test compound. As with logP measurements, logD is

¹ R. A. Prentis, Y. Lis, S. R. Walker - Br. J. Clin. Pharmacol. 1989, 25, 387

determined potentiometrically and its value reflects the partitioning of an ionized substances between 1-octanol and a buffer. The resulting lipophilicity profiles are a valuable means of interpreting substance behavior.



Solubility determinations / Solubility profiles

At Across Barriers, we use HPLC coupled with UV detection to measure the solubility of compounds in a variety of aqueous buffer systems. Using this technology, we can measure the saturation solubility of the solute at a specified temperature. The resulting pH-solubility profiles provide information about the expected solubility of the test substance during its passage through the gastrointestinal tract. This type of investigation is suitable for well characterized substances of known crystal structure and purity.

A drug substance is classified as highly soluble when the highest dose strength is soluble in 250 ml or less of water over the pH range from 1 to 7.5.

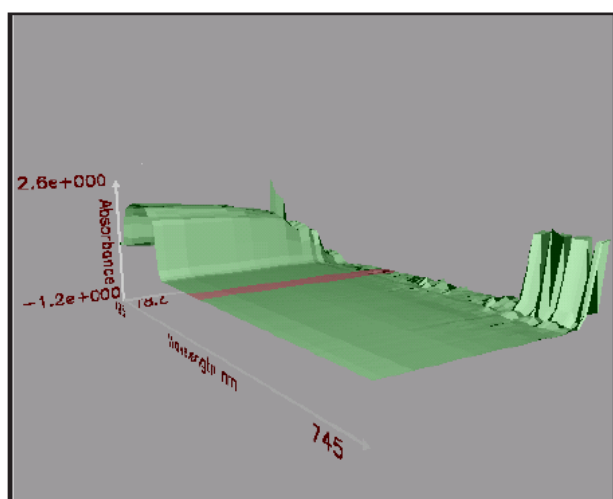
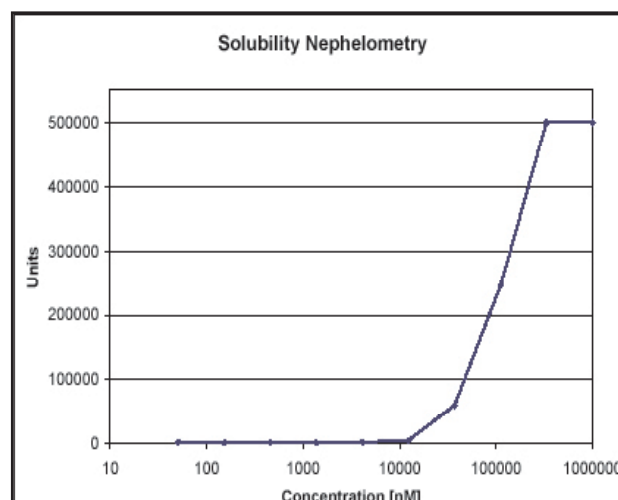


Figure 1: Determination of the BCS solubility class boundary for drug substances using titrimetry and diode array detection. (Example shown: propranolol-HCl)



rapid screening methods. Frequently, the substances are available dissolved in DMSO. A rapid estimate of solubility can be achieved by adding the test solution dropwise to different buffer systems under specified thermodynamic conditions until precipitation begins. The end point is detected by turbidimetry or by a photodiode array sensor. The method, which requires sample weights in the low milligram region, can be used to predict potential solubility problems that could hamper product development. The BCS solubility class can therefore be determined at a very early phase.

Development of dissolution profile testing

Across Barriers is equipped with state-of-the-art equipment with which it can measure dissolution profiles in accordance with USP specifications (USP Apparatus I or II). The development of a reliable dissolution test is based on the solubility and pKa data for the test substance and is carried out in accordance with the FDA's recent BCS recommendations. Quantitative determination of drug dissolution is performed using UV/photodiode array or HPLC/photodiode array detection. No matter whether the substance is classified as an IND/CTX, an NDA or an ANDA, the documentation provided by Across Barriers, which includes a method validation report, is of approval quality.

At earlier stages of drug or cosmetic development, it is often wholly sufficient to gauge solubility using

Determination of KIAM

KIAM is the partition coefficient of a substance between an immobilized artificial membrane and the surrounding solution. The technique used to measure KIAM requires only small amounts of the test substance. It offers a means of screening for substance-specific permeation properties at a very early stage of the development process and is thus a valuable tool when selecting promising drug candidates. The experiments are done using HPLC with UV detection. The stationary phase used in the HPLC column (IAM.PC.DD2 from Regis Technology Inc.) is based on phosphatidylcholines which mimic the surface of a biological membrane. In most cases, there is outstanding correlation between the retention factors and the permeability in physiological membranes in in-vivo and in-vitro experiments.