

### Attain More Certainty by Revealing Your Compound's Cellular Target Profile and Mode of Action!

The KINAXO technology portfolio delivers comprehensive insights into a compound's native target profile and reveals its influence on cellular signal transduction pathways.

Profound knowledge about your compound's target interactions and binding affinities across the proteome enables development programs to reach high confidence levels regarding *in vivo* selectivity and mode of action.

- Identify all protein targets that interact with a small molecule within a cell line or tissue proteome
- Rank cellular compound/target binding affinities across a broad dynamic range
- Profile small molecules against native cellular derived proteins
- Unveil target interactions not revealed by *in vitro* panel profiling
- Discover your compound's influence on cellular signal transduction pathways and reveal its mode of action

### Applications

#### • Compound selection for (pre)-clinical studies

- Enhance lead compound selection by comparing candidate interaction profiles and target affinities across the entire proteome of a cell line or tissue sample.
- Reveal off-target interactions that may contribute to potential compound side effects.

#### • Target deconvolution

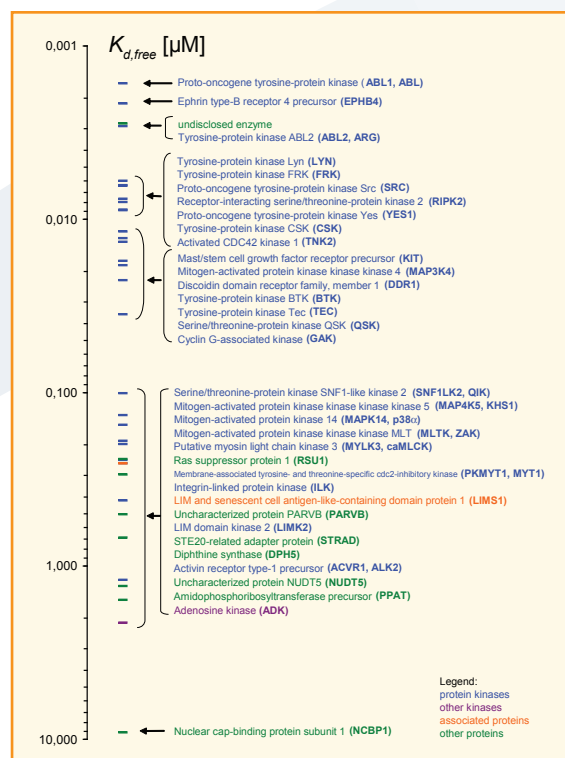
- Uncover the mode of action of compounds identified in phenotypic screening campaigns, helping to set the stage for a more confident development program.

#### • Drug reprofiling and drug rescue

- Identify novel targets and relate them to new indications for approved or failed compounds.

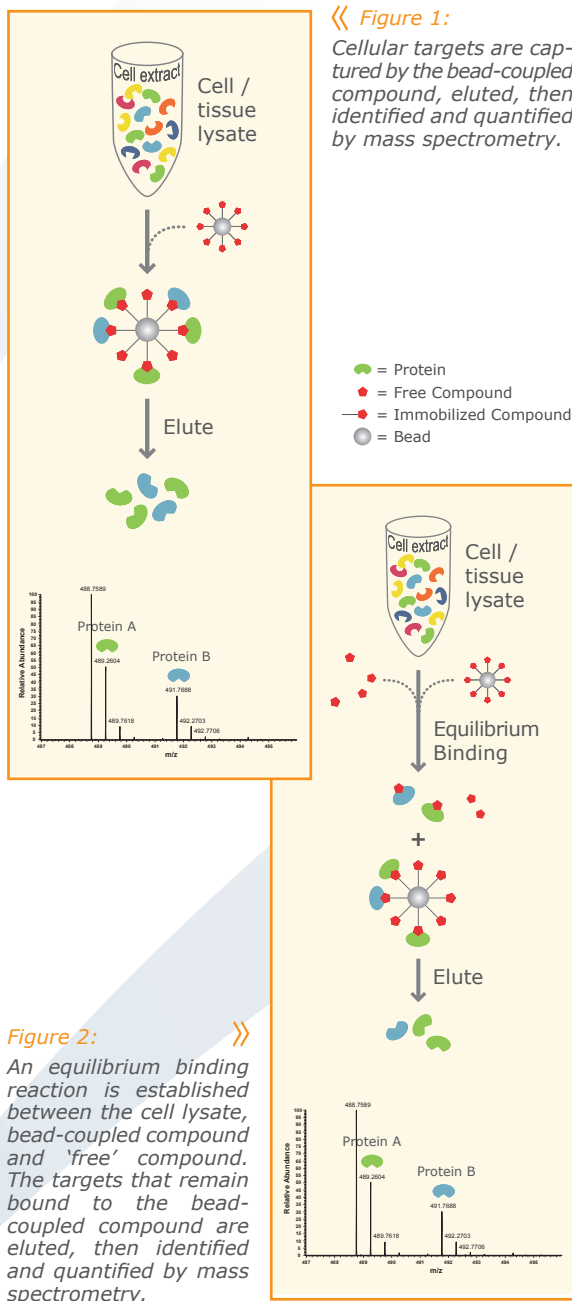
#### • Mode of action analysis

- Use system-wide modeling of signaling events and investigate your drug's influence on cellular phosphorylation patterns to significantly decrease development times in drug discovery



## KINAXO's Cellular Target Profiling® Technology

KINAXO has extensive experience and world-class expertise in the drug profiling applications of mass spectrometry-based chemical proteomics, the core platform that underpins the KINAXO Cellular Target Profiling® service. SILAC (Stable Isotope Labeling by Amino acids in Cell culture), iTRAQ (Isobaric Tags for Relative and Absolute Quantitation) and TMT (Tandem Mass Tags) are proteome labeling technologies employed for quantitative mass spectrometry analysis of cell line and tissue samples. The combination of these technologies with state-of-the-art proprietary affinity-based separation and data processing methods enable the determination of free compound affinities to all cellular targets.



### Determining of cellular target binding affinities

The  $K_{d, free}$  values define the binding affinity of the free (non bead-coupled) compound to each of the targets identified from the cellular proteome. These values are derived by a two step process.

Firstly, the affinities of each of the protein targets for the bead-coupled compound are determined (Figure 1). Secondly, competition experiments are performed where the free compound displaces targets bound to the bead-coupled compound (Figure 2).

These experiments require multiple test conditions in order to generate reliable data to derive binding and competition curves. Metabolic (SILAC) or chemical (iTRAQ, TMT) labeling of the cell line or tissue sample proteomes, combined with proprietary experimental and analytical methods facilitate relative quantitation. The final  $K_{d, free}$  values are derived by applying algorithms to the above two data sets.

### Ensuring the highest quality data

For every customer project a dedicated series of control experiments are performed in order to obtain the highest quality and appropriately annotated data. The controls ensure excluding any background binding signal, avoiding bead saturation and defining the binding equilibrium status for each target/compound interaction.

### KINAXO Service — Refined and Responsive

KINAXO works closely with your team in a collaborative manner to define the experimental design that will most effectively support your drug profiling application.

#### Phase 1 (in collaboration with the client)

- Selecting cell lines and / or tissues to be analyzed
- Define project requirements



#### Phase 2 (performed at KINAXO)

- Carrying out KINAXO's chemical proteomics or phosphoproteomics analysis
- Identifying the compound's target profile / evaluating its cellular mode of action using high-end quantitative mass spectrometry



#### Phase 3 (study report)

- Protein targets ranked by their affinities/ regulated phosphorylation sites
- Biological function of the targets reported where known

### Application and Technical Notes

Please refer to the KINAXO website for the latest Technical / Application Notes and references describing the practice and utility of KINAXO's technology platform: [www.kinaxo.com](http://www.kinaxo.com)

#### About KINAXO

KINAXO Biotechnologies GmbH is a service based company specializing in chemical proteomic methods to support the successful development of small molecule drugs. To continuously expand our technology platform we collaborate with the leading Max Planck laboratories of Axel Ullrich, Henrik Daub and Matthias Mann.

KINAXO has proven itself to the pharmaceutical industry as a distinctive leader in drug-based chemical proteomics, having successfully applied its services to the profiling of small molecule inhibitors of various targets including kinases, cyclooxygenases (COXs) and heat shock proteins (HSPs).

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