# SPOTTING THE WEAKEST BINDERS

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## A NEW LEVEL OF PERFORMANCE AND FLEXIBILITY IN DRUG DISCOVERY

By bringing together modern label-free technology, application development know-how and sophisticated software, Creoptix offers a unique optical biosensor tool for binding kinetics. Engineered around our proprietary Grating-Coupled Interferometry (GCI)[1] technology, the Creoptix WAVEsystem delivers high-quality kinetic data across a broader range of samples than traditional SPR equipment.

**Screen, rank and characterize weak binders with off-rates up to 10 $s^{-1}$**

**Study binding kinetics even at large analyte:ligand MW ratios (up to 1:1000)**

**Experiment with crude mixtures, detergents and other additives without clogging**

## GRATING-COUPLED INTERFEROMETRY (GCI)

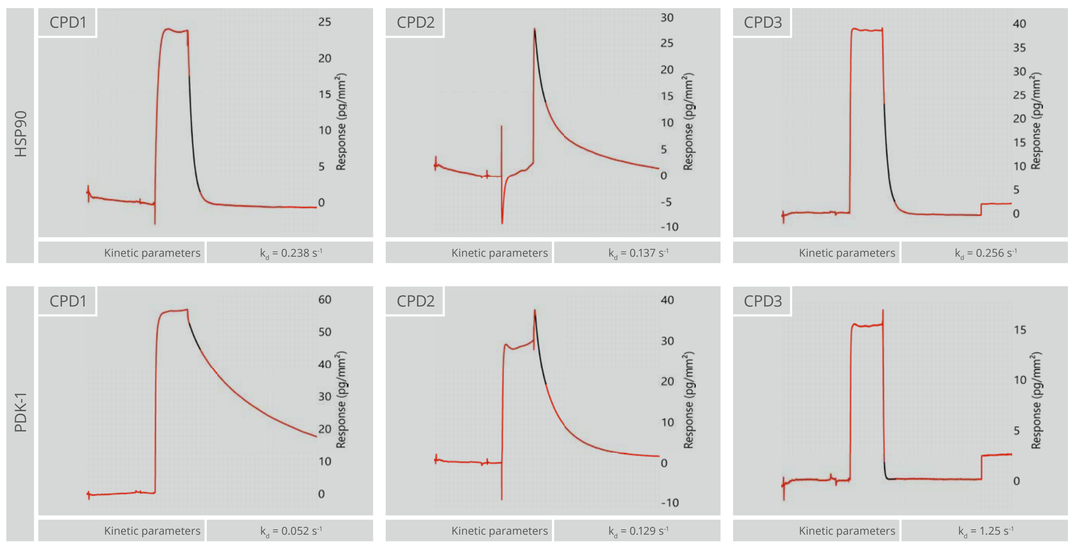
More information about GCI

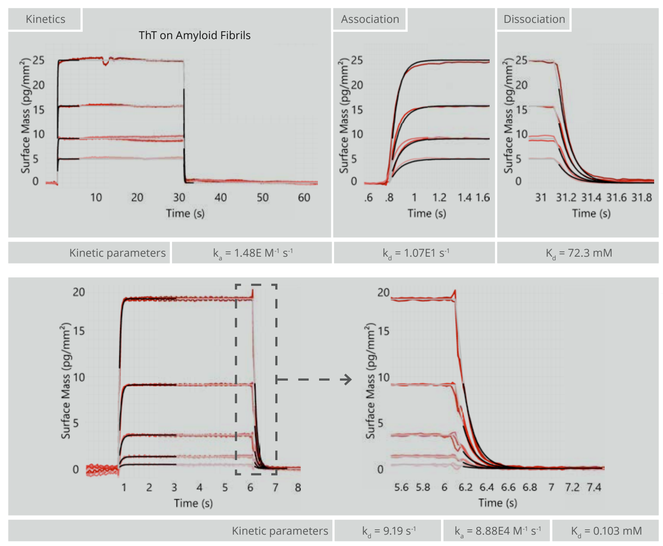
GCI is a surface-based, label-free biosensing technique. When target molecules (e.g. proteins) are attached to the sensor surface, binding of analytes leads to an increase in mass and hence to a change in the refractive index within the evanescent field near the surface. In GCI, refractive index changes on a sensor surface are measured as time-dependent phase-shift signals. The long light-to-sample interaction length of the waveguide provides intrinsically high signal-to-noise levels for improved sensitivity.

The Creoptix® WAVEsystem combines GCI with innovative no-clog microfluidics, allowing the study of interactions even between very small ligands and large complexes. The system uses a robust microfluidic sensor, the WAVEchip®, where (membrane) proteins,[2] antibodies, VLPs, peptides or other molecules can be immobilized using various chemistries.


## WEAK BINDERS, STRONG DATA

Weak binders such as those found in fragment-based screening are typically ranked by affinity rather than kinetics due to their very fast off-rates, which can not be resolved by traditional SPR instrumentation. Here we show that the Creoptix® WAVEsystem provides an outstanding resolution whereby very fast kinetics can be reliably determined at off-rates up to 10 $s^{-1}$.



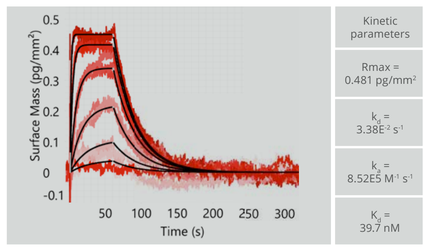

Self-assembled amyloid fibrils were immobilized via amine coupling on a 4PCZ WAVEchip® (zwitterionic surface). The small molecule thioflavin (ThT, 319 Da) was injected in four (4) concentrations (50 mM - 6.25 mM) for 30s at 400 ml/min. Raw data were double referenced and globally fit with a 1:1 binding model showing accurate determination of an off-rate around 10 $s^{-1}$.



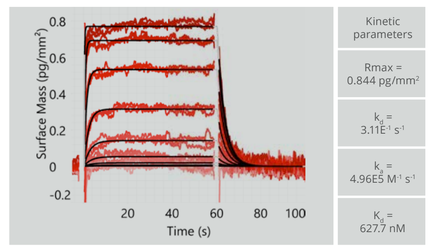

Sensorgrams of a 6-mer oligonucleotide (1.7 kDa) binding onto its complementary ssDNA (11 kDa biotinylated 34-mer) captured on streptavidin on a PCP-S WAVEchip. The interaction was measured at 25°C. Zoom into the dissociation shows excellent data fitting and an accurate determination of an off-rate around 10 $s^{-1}$.

## SMALL MOLECULES CAN'T HIDE ANYMORE

Sensitivity is key and often limiting for accurate and reliable analysis of molecular interactions. The high-sensitivity of the Creoptix® WAVEsystem allows researchers to confidently analyze binding interactions at very low signal levels and high analyte-to-ligand molecular weight (MW) ratios.




Sensorgrams of acetazolamide (222 Da) binding to Carbonic Anhydrase II (29 kDa) immobilised at low density onto a PCH WAVEchip.




Sensorgrams of a small drug molecule (295 Da) binding to a target protein (110 kDa) immobilised at low density onto a PCH WAVEchip. Note the analyte-to-ligand MW ratio is > 300.

GCI IS FEATURED IN




## REFERENCES

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3 Brough et al., "Application of Off-Rate Screening in the Identification of Novel Pan-Isoform Inhibitors of Pyruvate Dehydrogenase Kinase", J. Med. Chem., 60 (6), pp 2271–2286 (2017)
4 Brough et al., "4,5-Diarylisoxazole Hsp90 Chaperone Inhibitors: Potential Therapeutic Agents for the Treatment of Cancer", J. Med. Chem., 51 (2), pp 196–218 (2008)

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