SPOTTING THE WEAKEST BINDERS

creoptix sensors

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A NEW LEVEL OF PERFORMANCE AND FLEXIBILITY IN DRUG DISCOVERY

Employing our Grating-Coupled Interferometry (GCI)[1] technology to deliver superior sensitivity over traditional Surface Plasmon Resonance (SPR) technologies, researchers can reliably determine off-rates of up to $10 s^{-1}$, starting with just a crude reaction mixture. Combined with microfluidics that sustain a wide variety of solvents - including acetonitrile and high concentrations of DMSO -, the occurrence of false positives is minimized.

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)

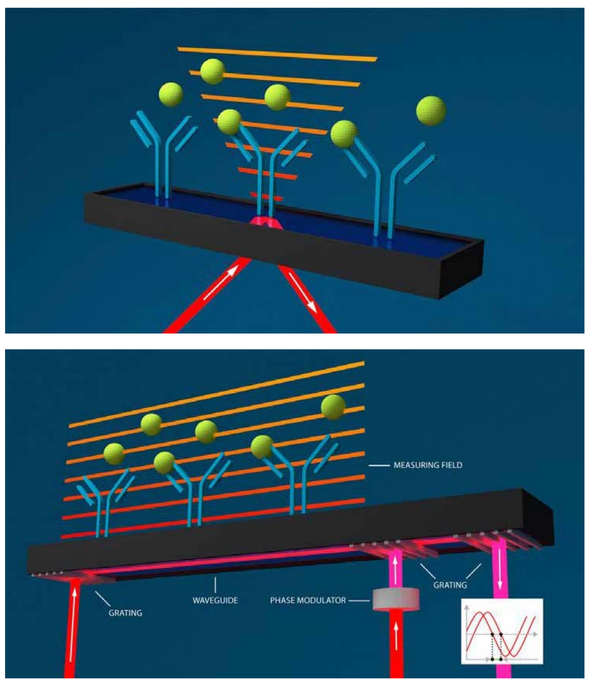
Experiment with crude mixtures, detergents and other additives without clogging

GRATING-COUPLED INTERFEROMETRY (GCI)

Grating-coupled interferometry (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.

With high sensitivity and the ability to resolve extremely rapid dissociating kinetics, and innate compatibility with the high molecular weight ratios, the GCI technology improves fragment-based screening and kinetic analysis of small molecules to accelerate drug development.

More information about GCI



Surface Plasmon Resonance (SPR) is a label-free, optical biosensing technology that measures changes in refractive index close to a sensor surface. Traditional Surface Plasmon Resonance sensors consist of an electrically conducting metal film (gold) upon which the ligand of interest is immobilised.

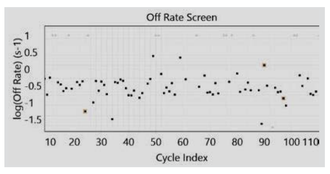
In SPR, molecular interactions are detected as changes in refractive index within an evanescent field (orange) of the surface plasmon shown as energy dips at specific incidence angle.

Interpretation of Waveguide Interferometry data relies on accurate alignment of the reference beam and the measurement beam. However, during classical Waveguide Interferometry, perfect alignment is extremely challenging and sensitive to environmental effects such as temperature shifts or mechanical distortions or vibrations.

Grating-Coupled Interferometry. In grating-coupled interferometry (GCI) the reference beam is also coupled into the waveguide. Consequently, interference happens within the waveguide and a high-resolution, time-dependent and robust phase shift signal is created .

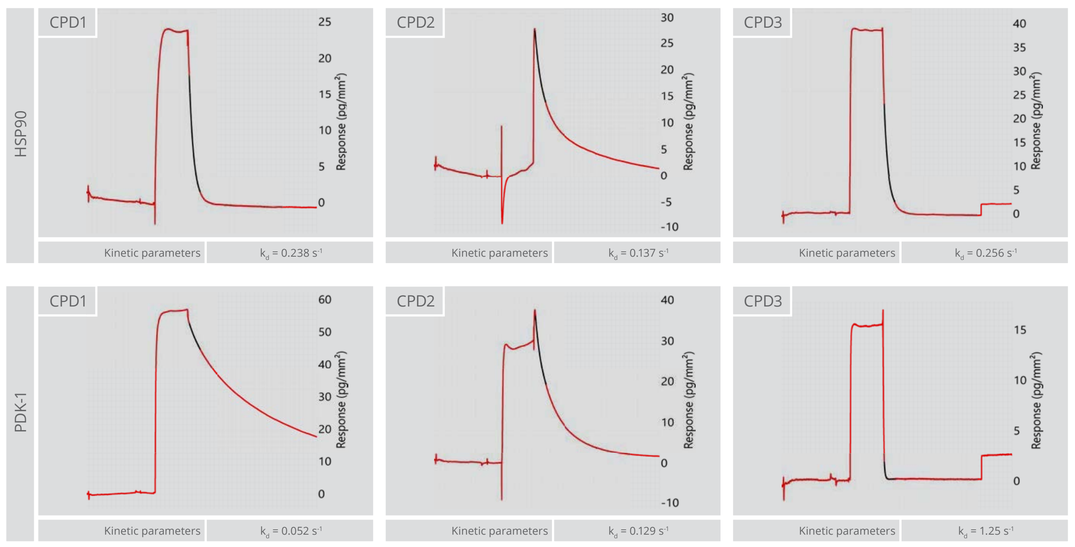
HITS WON'T BE MISSED

The GCI technology provides a versatile platform for fragment screening and analysis. Thanks to its ultra-fast transition times, excellent resolution for compounds with k_d's well above 1 s^{-1} can be achieved.



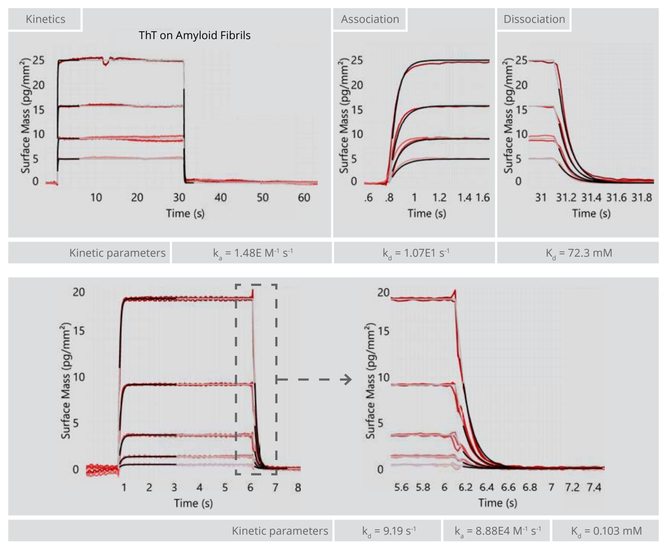
Selected HSP90 or PDK-1 hits (highlighted) are in good agreement with the reported literature[3-4]. Aberrant samples can be simply and automatically excluded based on the apparent k_d and the Chi² (grey triangles).

Sensorgrams of selected compounds of a 83 crude non-purified compounds[3-4] library-subset, screened against His-tagged Pyruvate Dehydrogenase Kinase (PDK-1) and captured on a PCH WAVEchip. Samples were injected at ~ 20 µM. His-tagged HSP90, captured on a different channel, served as negative control.



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 GCI technology provides an outstanding resolution whereby very fast kinetics can be reliably determined at off-rates up to 10 s^{-1}.

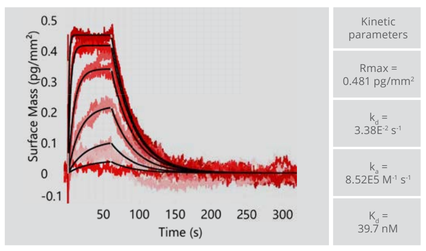


Self-assembled amyloid fibrils were immobilized via amine coupling on a 4PCZ WAVEchip® (zwitterionic surface). The small molecule thioflavin (ThT, 319 Da) was injected in four (4) concentrations (50 mM - 6.25 mM) for 30s at 400 ml/min. Raw data were double referenced and globally fit with a 1:1 binding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

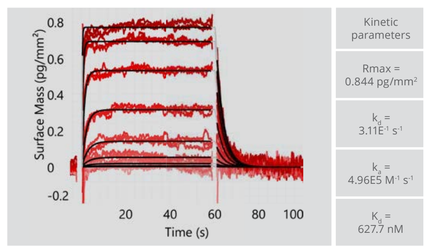
Sensorgrams of a 6-mer oligonucleotide (1.7 kDa) binding onto its complementary ssDNA (11 kDa biotinylated 34-mer) captured on streptavidin on a PCP-S WAVEchip. The interaction was measured at 25°C. Zoom into the dissociation shows excellent data fitting and an accurate determination of an off-rate around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

PNAS — Mechanistic basis for the activation of plant membrane receptor kinases by SERK-family coreceptors

nature plants — LETTERS — The SERK3 *elongated* allele defines a role for BIR ectodomains in brassinosteroid signalling

eLIFE — RESEARCH ARTICLE — Dynamics of human protein kinase Aurora A linked to drug selectivity

REFERENCES

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2 Hohmann et al., "The SERK3 elongated allele defines a role for BIR ectodomains in brassinosteroid signalling.", Nature Plants, 4:345–351 (2018)
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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