Zurich University of Applied Sciences
Life Sciences and Facility Management
Institute of Chemistry and Biological Chemistry

Development of a high sensitivity label-free waveguide interferometry instrument: A project of Creoptix GmbH with the Center for Biochemistry ZHAW as main research partner

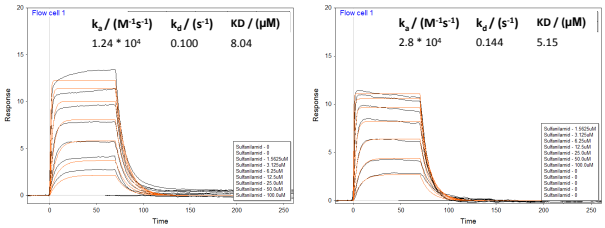
Kaspar Cottier[1], Matyas Vegh[1], Helmut Knapp[2], Siegfried Graf[2], Daniel Gygax[3], Peter Spies[3], Roland Josuran[4], Angelika Koller[4], Reto Gianotti[4], Romina Dörig[4] und Christiane Zaborosch[4]

[1]Creoptix GmbH, Einsiedlerstrasse 25, CH-8820 Wädenswil, [2]CSEM Alpnach, Untere Gründlistrasse 1, CH-6055 Alpnach, [3]School of Life Sciences FHNW, Institute for Chemistry and Bioanalytics, Gründenstrasse 40, CH-4132 Muttenz, [4]Zurich University of Applied Sciences ZHAW, Institute of Chemistry and Biological Chemistry, Einsiedlerstrasse 31, CH-8820 Wädenswil

Abstract
Creoptix GmbH has developed a novel and innovative technology for label-free detection of molecules based on grating-coupled interferometry (GCI). GCI is a proprietary technology characterized by a very high sensitivity at low technical complexity. One main application will be the measurement of binding affinities in research and development projects such as drug discovery. Goal of an interdisciplinary CTI project with the partners from ZHAW, FHNW and CSEM together with Creoptix is the development of a first GCI instrument including disposables and the optimization of its functionality, followed by field tests to pave the way to market introduction.

Introduction
The main advantage of label-free methods over conventional methods based on absorption, fluorescence or radioactivity is that the interaction between the molecules is not disturbed by any label. Furthermore, labeling and washing steps are not needed and thus reducing reagent use, costs and time. Compared to other label-free sensors such as the ones based on surface plasmon resonance (SPR), grating-coupled interferometers show higher sensitivity due to a longer interaction with the sample (some millimeters compared to micrometers with SPR). In addition, their evanescent fields have a shorter penetration depth of 80-100 nm compared to 200-300 nm with SPR, and thus interferometric waveguide sensors are less sensitive to matrix effects caused by changes in bulk refractive index.

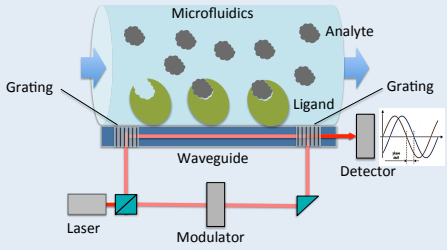
Results
For comparison, measurements were performed on a SPR-based Biacore T200 (GE Healthcare), currently the most sensitive label-free instrument on the market. The determination of the binding of low molecular weight drug candidates for target proteins requires highest sensitivity. The GCI prototype QUBE CX-3 showed equivalent kinetic data as Biacore T200 for the binding of sulfanilamide (172 Da) to carbonic anhydrase, however with an about 4 fold higher signal-to-noise ratio (see figure below).



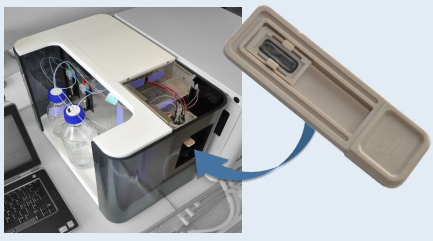
Binding of sulfanilamide to carbonic anhydrase. Comparative measurements with the GCI prototype QUBE CX-3 (left) and the SPR-based Biacore T200 (right)

The GCI prototype was also capable to measure the very high affinity (K_D = 90 pM) of the interaction of human interleukin-1β and a monoclonal antibody which is in clinical development. Importantly, for this interaction, the very low dissociation rate constant (k_d = 5.7x10^{-5} s^{-1}) could reliably be measured on the GCI prototype demonstrating a high stability of the instrument.

Grating-Coupled Interferometry



Working principle of Creoptix' grating-coupled interferometer: Central element is a chip with a tantalum pentoxide optical waveguide. The surface of the chip is functionalized to allow immobilization of ligands or receptors to the chip. Binding of analyte occurs when the analyte is passed over the chip in a microfluidic system. The readout is achieved by detecting a time-dependent interference signal which is generated by the superposition of a phase-modulated reference wave and an unmodulated signal wave within the waveguide. The binding of the analyte to the immobilized receptor in the evanescent field results in a phase change of the signal wave, thereby shifting the interference pattern.



The QUBE CX-3 with the disposable chip cartridge with integrated microfluidics: The innovative integration of the microfluidics into a disposable cartridge will lower device down-time and together with the modern software will improve the ease of use with respect to existing instrumentation.

Conclusion
In this project a grating-coupled interferometer with highest sensitivity was successfully developed in collaboration of a young innovative company with universities of applied sciences and the CSEM. The feasibility studies for the use in Life Sciences will help to pave the way to the market launch planned for end of 2013.

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