Multi– Channel and Imaging Surface Plasmon Resonance Analyser

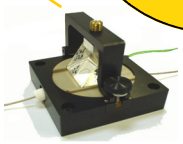
Cyclic flow and one- way flow

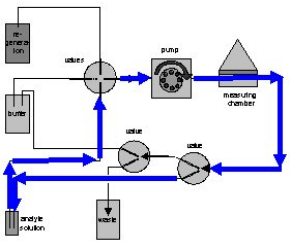
Profiling of interactions in up to 1000 spots simultaneously

→Binding specificity (Yes/ No)

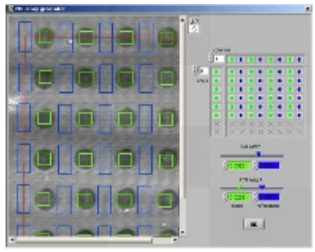
Two error corrections via monitoring of mass density and surface roughness, and via background

→Kinetics and affinity constants





Applications:
- Antibody-antigen reaction
- Protein-drug interaction
- Peptide-protein reaction
- Protein-saccharide interaction
- Binding of lipids and membranes
- LMW (low molecular weight compounds) binding on SPR-slides with receptor film
- Quality control of microarrays in air without SPR-cell



Technical features:
- Ellipsometric Platform EP³ (635 nm wavelength, 25mW laser, goniometer accuracy ± 0.01°, precision ± 0.001°angle-of -incindence)
- programming of profiling routines SPR cell (volume 100 µl, material: PEEK®, for SPR slides, Optislides, glass slides, for use with liquid or gas)
- Kinetic AddOn software for multi-spot profiling, supports calculation of kinetic constants
- SPR slides for use with 635 nm or 532 nm wavelength
- 2x objective (1.3 mm x 2.0 mm field of view, 4 µm resolution, e.g. for array with 70 spots (with 100 µm spot diameter at 200 µm pitch)
- index matching oil
- background correction
- correction for time-dependent change of roughness of bound layer
- sensitivity: typically 2 pm ≈ 2 pg/mm²

Options:
- liquid handling with 6 channels incl. peristaltic pump for cyclic flow (0.05 – 2 ml/min, 0.6 ml volume incl. tubes)
- glasses and SPR slides for individual coating
- Optislides, metal-free glass-slides with high sensitivity, for profiling without SPR
- automatic sample handling stage, for automatic scanning of many fields of view on large microarrays
- temperature control with Peltier element or water
- syringe pump for one-way flow

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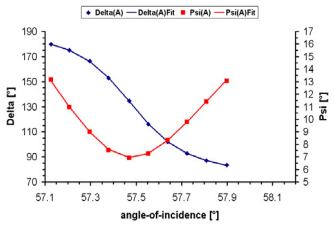
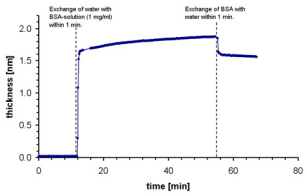
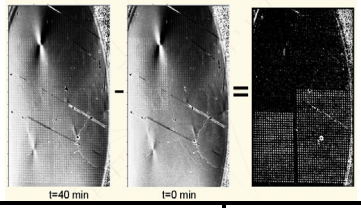
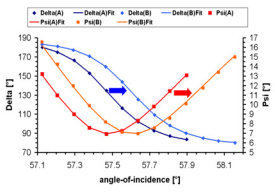
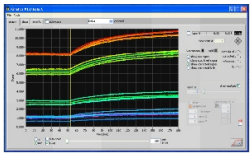
Working steps with the EP³ SPR:

Profiling of interactions	Specificity measurement	Examples
Preparation (clean SPR cell, SPR prism and SPR slide first with NaOH and second with HCl, put the SPR slide in the SPR cell and connect it with the SPR prism by a drop of immersion oil, center the microarray in the field of view and adjust the focus)		
EP³ records an **angle spectrum before kinetics** (optional, only needed for correction of time-dependent change of roughness of bound layer)		Clean gold surface of SPR slide in buffer Fit results: refractive index n (gold) = 0.276 ± 0.001, extinction k(gold) = 3.690 ± 0.001, thickness d(gold) = 43.656 ± 0.001 nm, n(liquid) = 1.3496 ± 0.0001 
	EP³ records Delta/Psi maps and converts them into maps of thickness/ mass density and of effective surface roughness (recommended for specificity measurement)	Delta map of two arrays of mouse IgG and rabbit IgG, 2000 spots (8 µm diameter, 40 µm pitch). Scratches and impurities are only observable in ellipsometric images but not in fluorescence images! 
EP³ runs a program of injection and washing steps, and profiles all spots simultaneously Example: Binding of BSA (1 mg/ml) on gold surface of SPR slide, 1 nm ≈ 1 ng/mm² 1σ standard deviation of baseline ± 0.002 nm		
	EP³ records Delta/Psi maps and converts them into maps of thickness/ mass density and of effective surface roughness, a map of specificity is given by the difference of Delta maps run before and after the kinetics	Arrays of mouse IgG and rabbit IgG after incubation with anti-rabbit IgG (50 nM) 
EP³ records an **angle spectrum after kinetics** (optional, only needed for correction of time-dependent change of roughness of bound layer)		Spectrum after binding of BSA (1 mg/ml) on gold surface of SPR slide fit results: refractive index n (liquid) = 1.346 ± 0.003, thickness d(BSA) = 3.7 ± 2.1 nm, extinction k = 0 
View kinetics profiles and calculate kinetics constants Kon, Koff, Kd with the Kinetics AddOn software		

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