

# SPRm 200 Series



A new way to study cell membrane proteins binding kinetics

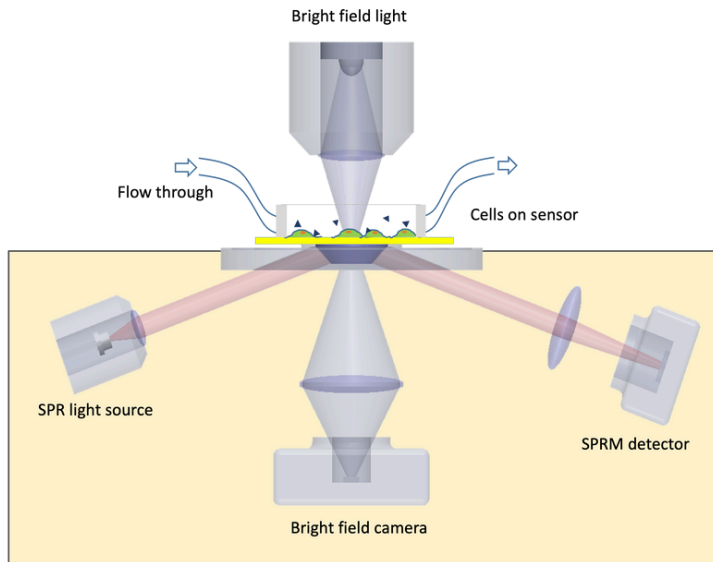


- Label-free binding measurements of in vitro cell membrane proteins
- Quantitative mapping of binding affinity and kinetics in real time
- Simultaneous optical imaging with SPR measurements
- Cell heterogeneity and avidity analysis
- Nanometer-scale binding events monitoring

SPRm 200 system opens a new frontier in the study of molecular interactions by integrating SPR technology with optical imaging. Designed especially for in vitro, label-free measurement of binding activity and cell kinetics, SPRm 200 provides a spatial visualization of cellular structures together with local binding activities. Real-time interactions between the drug and membrane protein can be measured in their native state without extracting proteins from the cell. SPRm 200 also measures nanometer scale binding activities needed for studying bacteria and virus interactions and the development of new methods for nanoparticle drug delivery.

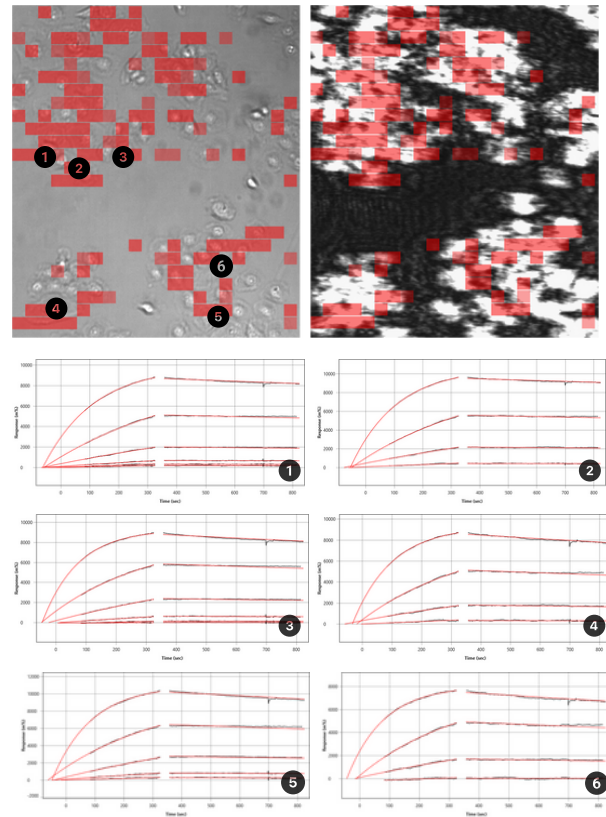
## Integration of SPR with optical microscopy

Surface Plasmon Resonance Microscopy (SPRM) combines optical imaging with SPR technology, providing spatial mapping of the binding activity on living cells. A light condenser illuminates the cell grown on the sensor surface, and the optical microscope camera captures the bright field image of the cells. Simultaneously, the SPR light source projects a beam at its resonance angle onto the sensor and the reflected beam is collected by the SPRM detector.



The detector measures the SPR response at each pixel and maps them into a SPR image. At every pixel, a sensorgram is recorded, thus providing much more localized information. SPRM makes it possible to study heterogeneous surface binding and interactions

of membrane proteins in their native states.

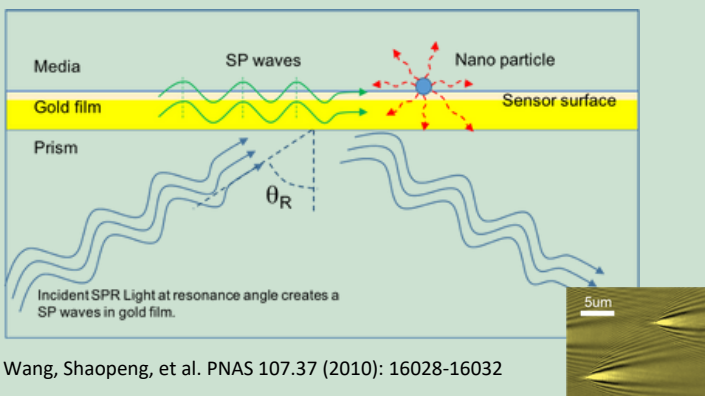


With the extensive data collected, the SPRM software presents the binding affinity and kinetic values in the form of a histogram.

## Nanoparticle detection

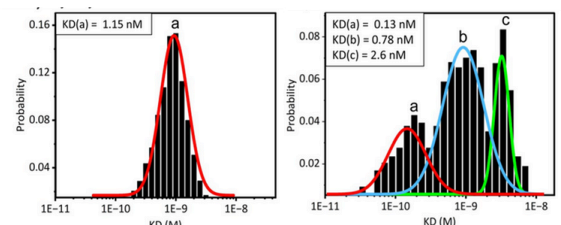
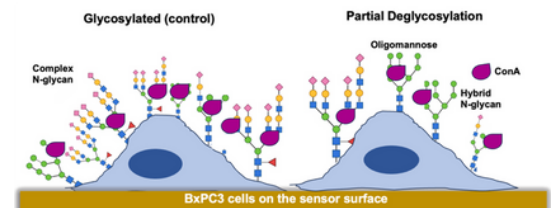
SPR light projected onto the sensor at its resonance angle creates a propagating surface plasmonic (SP) waves along the metal film surface. When a nanoparticle binds to the sensor surface, it acts as a scattering center in the SP waves, creating a wake pattern with a footprint up to 100X than its actual size. This enlarged footprint enables the detection of particles smaller than the optical diffraction limit, allowing nanometer scale binding activities be monitored and studied by measuring and mapping these footprints.

The occurrence and intensity changes of the wake patterns in the SPR image provide rich information about binding events between the sensor surface and the nanoparticles, as well as their interactions with other molecules in the media.



Wang, Shaopeng, et al. PNAS 107.37 (2010): 16028-16032

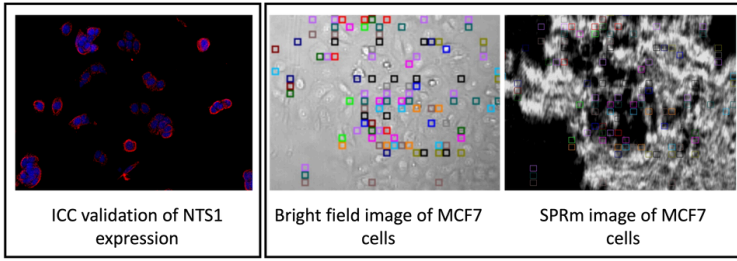
## Real-time monitoring of glycan heterogeneity in cancer cells



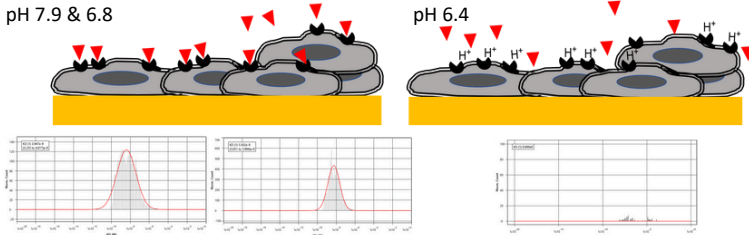
Con A binding affinity and heterogeneity increases upon enzymatic N-linked deglycosylation of BxPC3 cells. A single predominant binding mode for Con A in glycosylated control BxPC3 cells was observed ( $KD = 1.2$  nM). After BxPC3 cells underwent PNGase F treatment to remove N-linked glycans, three distinct binding interaction modes for Con A on BxPC3 cells were observed ( $KD = 0.1$  nM,  $0.8$  nM,  $2.6$  nM).

Aguilar Díaz de León, Jesús, et al. Plos one 19.5 (2024): e0304154

## Small and large molecule targeting GPCRs

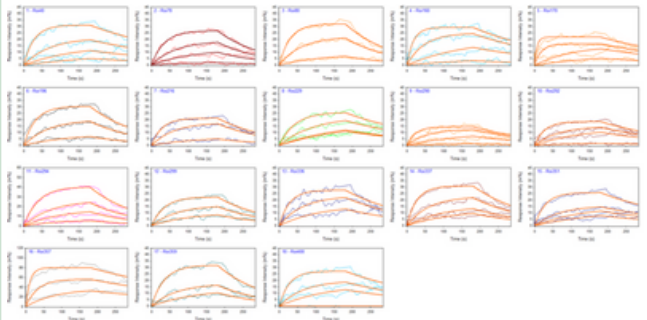
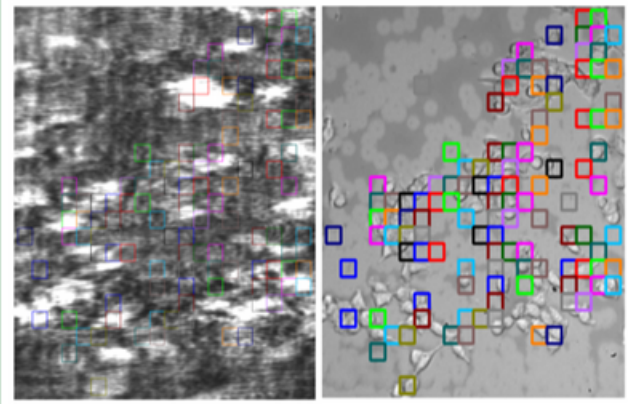


685 Da compound targeting NTS-1 receptor on MCF7 breast cancer cell line.  $K_D = 2 \text{ nM}$ ,  $k_a = 2.2 \times 10^6 \text{ M}^{-1} \text{ s}^{-1}$ ,  $k_d = 4.7 \times 10^{-3} \text{ s}^{-1}$ .



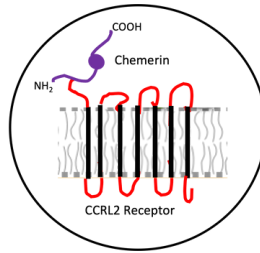
Targeting GPR4 with a ~500 Da compound at different pHs on hGPR-4 expressing HeLa cells.  $K_D = 4 \text{ nM}$  and  $5 \text{ nM}$  at pH 7.9 and 6.8 respectively. No binding is observed at pH 6.4 due to protonation.

Stalewski, Jacek, et al. J Pharmacol Exp Ther 386:35–44 (2023)

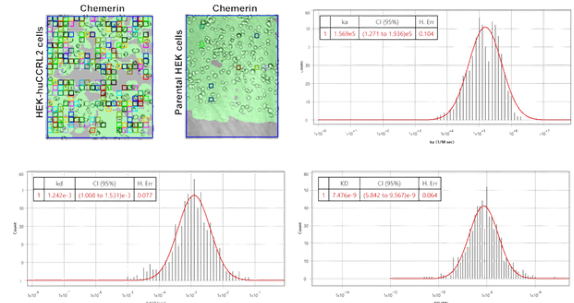


AZ1395 (300 Da) small molecule targeting GPR39 on HEK 293 cell line.  $K_D = 42 \text{ nM}$ ,  $k_a = 6.9 \times 10^3 \text{ M}^{-1} \text{ s}^{-1}$ ,  $k_d = 4.3 \times 10^{-3} \text{ s}^{-1}$ .

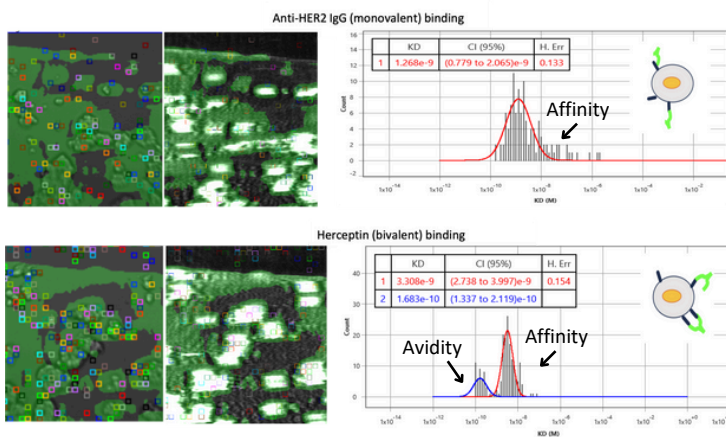
Human chemerin (16k Da) binding to CCRL2 7TM GPCR expressed HEK 293 cells.  $K_D = 5.5 \text{ nM}$ ,  $k_a = 3.1 \times 10^5 \text{ M}^{-1} \text{ s}^{-1}$ ,  $k_d = 1.2 \times 10^{-3} \text{ s}^{-1}$ . Human chemerin bound to parental cells show very little binding events. SPRM results are comparable to radiolabel chemerin affinity data.



Su, Zhenwei, et al. PLoS ONE 18(1) (2023): e0280590



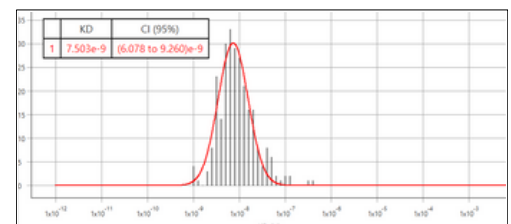
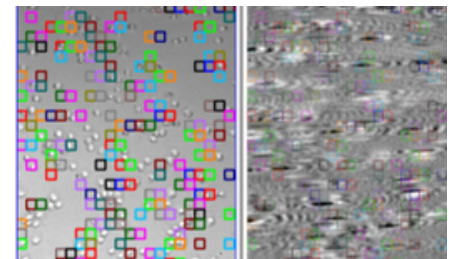
## Avidity measurements for antibodies



Kinetic parameters measured from anti-HER2 and Herceptin binding interactions with HER2 on the MCF7 cell line. The monovalent anti-HER2 antibody shows only one histogram peak at  $K_D = 1.3 \text{ nM}$ . In contrast, the bivalent Herceptin shows two histogram distribution for  $K_D$ . The  $K_D$  peak of larger value is attributed to single-arm affinity interactions and corresponds well to the monovalent  $K_D$  value (3.3 nM). The smaller  $K_D$  peak with stronger interaction is attributed to avidity effects (168 pM).

## Live suspension cells

Live B-cells (suspension cells) were capture on the SPRm 200 sensor surface using commercial cell adhesives. Binding with glycoproteins were measured.  $K_D = 7.5 \text{ nM}$ ,  $k_a = 2.5 \times 10^5 \text{ M}^{-1} \text{ s}^{-1}$ ,  $k_d = 2.0 \times 10^{-3} \text{ s}^{-1}$ .



# SPRm 200 Specifications

Base Station	Light source	690 nm
	Incident angles	40-76 Deg (continuous)
	Baseline noise	< 0.6 RU RMS (0.1 mDeg RMS)
	Baseline drift	3 RU/hr (0.5 mDeg/hr) (when ambient drifts < 1°C/hr)
	Temperature Control Range	15°C to 40°C (10°C below ambient temperature max)
	Field of view	Bright Field: 1200 x 900 um SPR: 600 x 450 um
	Magnification	Bright Field: x10 SPR: x20
	Resolution	Bright Field & SPR: 1 µm
	Stage translation / rotation	3mm x 3mm / 360 deg
	Outer dimension	690 (w) x 330 (h) x 340 (d) mm
	Weight	23 kg
	Power supply	110-230 V 50/60 Hz
Fluid Handling	Sample volume	1 to 1500 µL (application dependent)
	Kinetic constant	$k_a < 1 \times 10^7 \text{ M}^{-1} \text{ s}^{-1}$ $k_d > 1 \times 10^{-5} \text{ s}^{-1}$
	Dissociation constant	$K_D = 10^{-3} \text{ M (1 mM) to } 10^{-12} \text{ M (1 pM)}$
	Molecular weight cutoff	200 Da
Control System	Computer	Windows operating system
	Software	ImageSPR™ software including Data Analysis and Kinetics Analysis
Autosampler (option)	Sample capacity	2 x SBS standards (384 / 96), 2 x 48 Vials (1.5mL), 2 x 12 Vials (10mL)
	Sample cooling	Minimum: 4°C +/- 2°C
	Outer dimension	300 (w) x 575 (h) x 360 (d) mm
	Weight	21 kg
Automatic Buffer Exchange Pump and Degasser (option)	Buffer exchange	Automatic buffer exchange up to six sources
	Buffer degasser	In-line
	Buffer delivery	Continuous
	Outer dimension	305 (w) x 191 (h) x 330 (d) mm
	Weight	6.8 kg

## Sensors and consumables

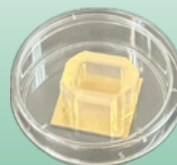
### Gold Sensor Chip

Highly uniform gold film for reproducible SPRM measurements.



### Cell Chamber Sensor Kit

Gold sensor chip with a polymer well for seeding cells.



Website: [www.BiosensingUSA.com](http://www.BiosensingUSA.com)  
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