

Interactions on the Octet Platform

ReNaFoBis 2018

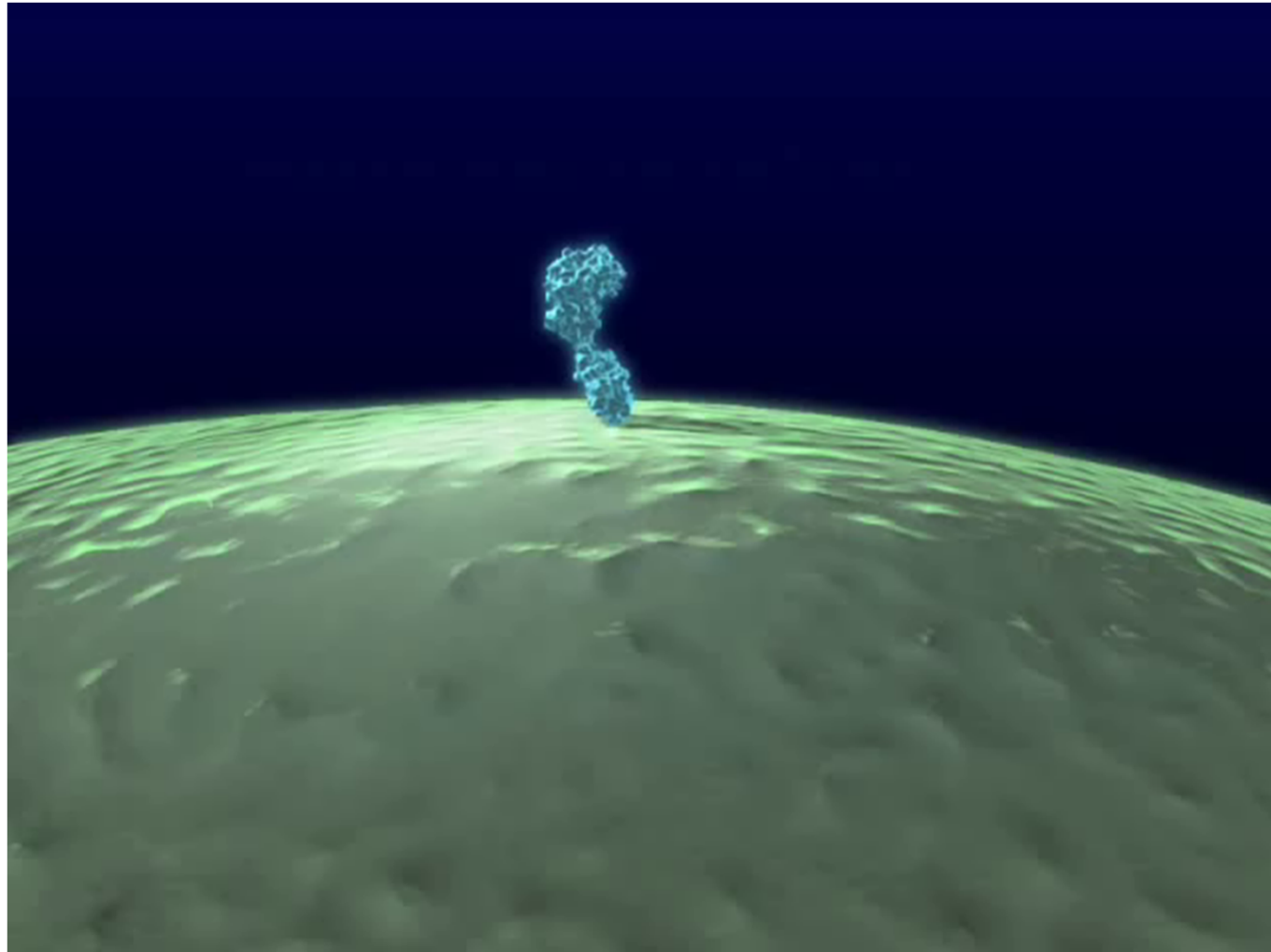
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Its all about Interactions...



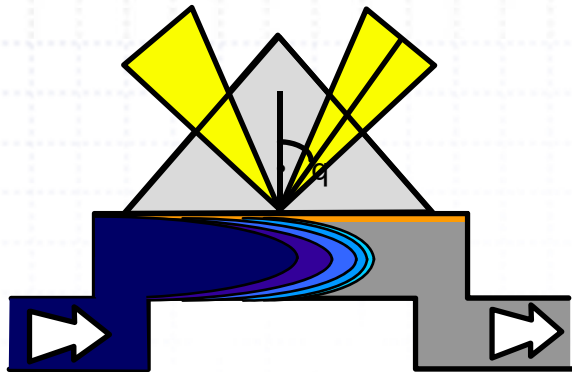
http://www.youtube.com/watch?v=Ms_ehUVvKKk



Real Time, Label-free Platform : OCTET



Label-free , Real-time Techniques

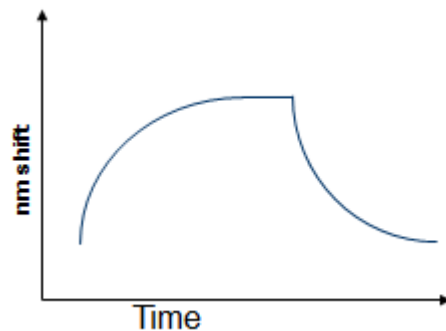


Mainstream

- BLI (Bio Layer Interferometry)
- SPR (Surface Plasmon Resonance)

Others

- QCM (Quartz Crystal Microbalance)
- SAW (Surface Acoustique Wave)



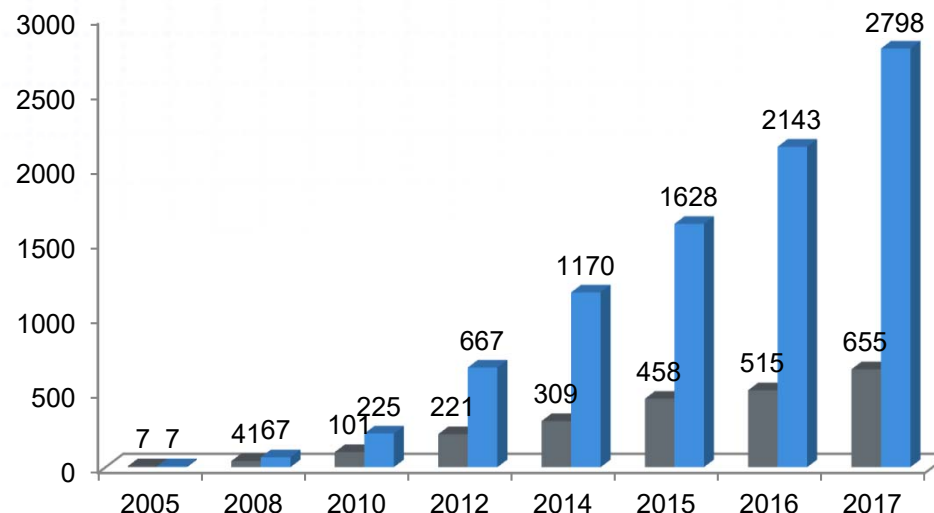
- They differ in detection method, but they all provide « Sensograms »
- They largely differ in speed, throughput, sample compatibility, usability

BLI=fast, accurate, easy / SPR=extreme sensitivity

BLI: Trending towards 3000+articles in 2018



PROTEIN|SCIENCE



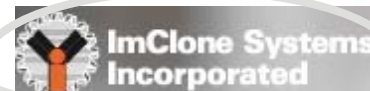
Cancer Research



BIOCHEMISTRY
including biophysical chemistry & molecular biology



Very strong Industry acceptance
Pharma, Biotech, Diagnostics



Molecular interactions in Biopharma

Discovery & Basic research

- Specificity = Which molecule?
 - Targets
 - Signalling mechanisms
- Affinity = How strong is the binding?
- Kinetics = How fast it will act?
- Mapping = Where it will bind?

Bioprocess development / recombinant proteins

- Concentration = Do I have enough?
 - Clones, cell cultures conditions
- Purity = Safe to use?
- Formulation (Stability & Activity)

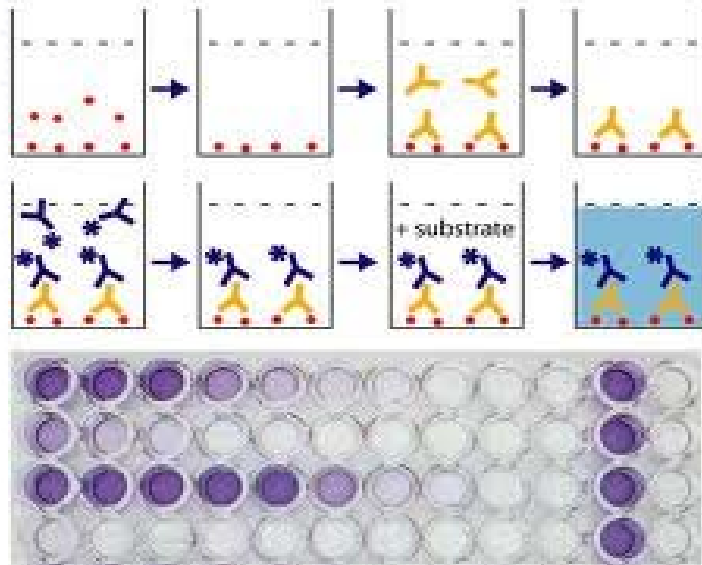
Clinical development

- Action or Immune reaction
- ADA



Traditional routes to answers: with label and at equilibrium

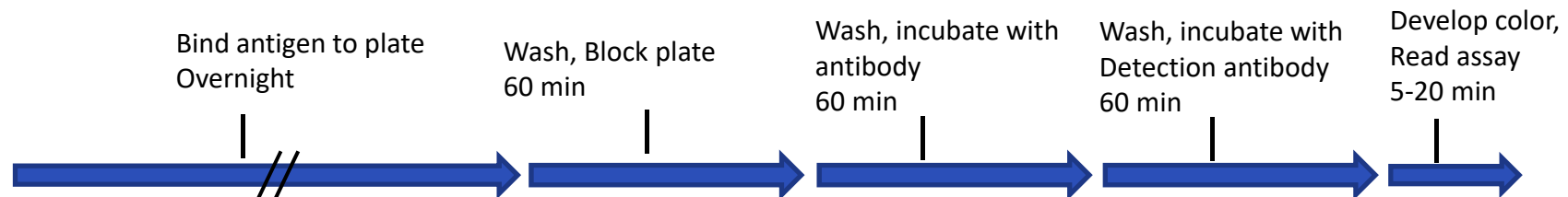
Protein Quantitation with ELISA:



Total assay time: 3-4 hours plus overnight incubation

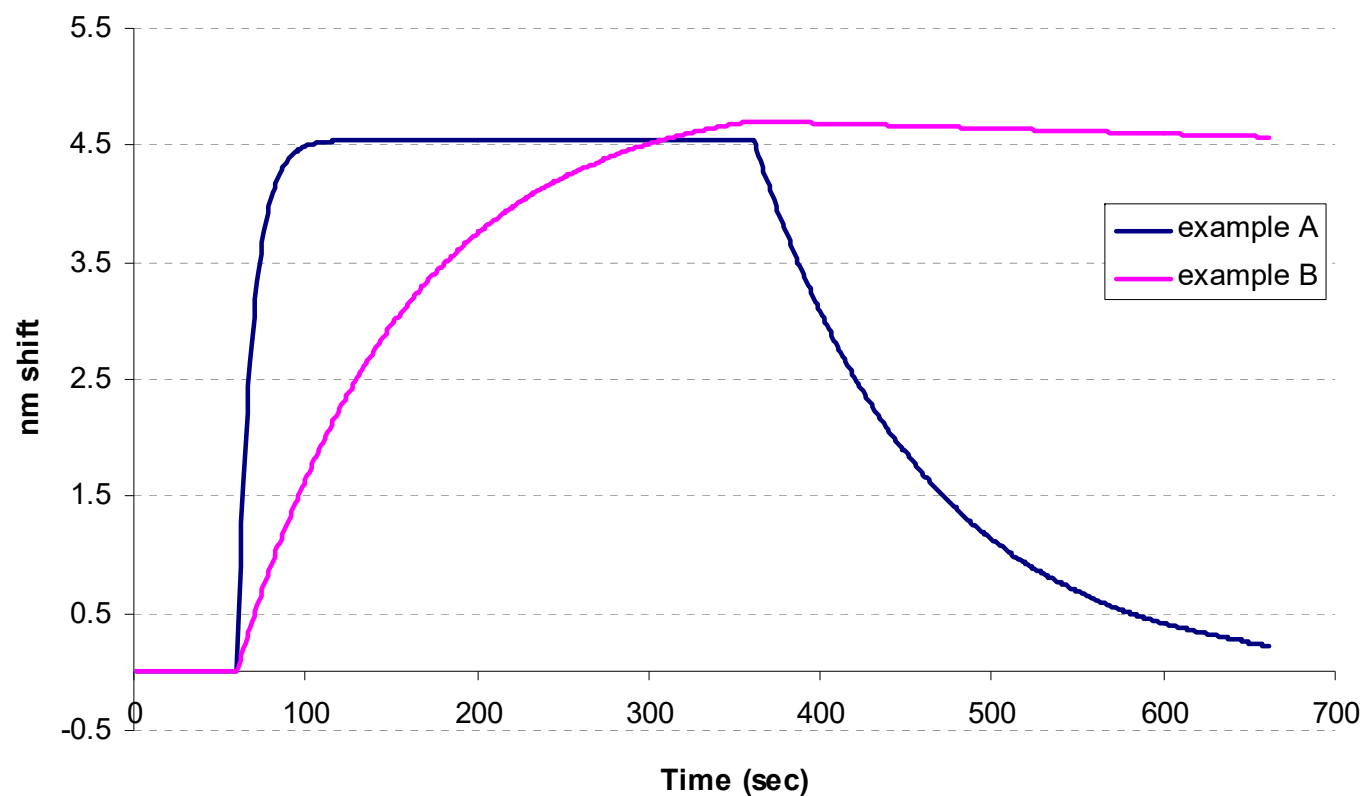
Requires 100-200 μ l sample

Target-specific antibodies

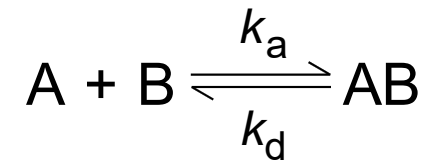
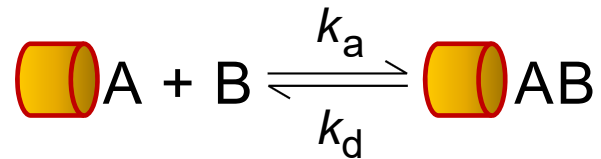
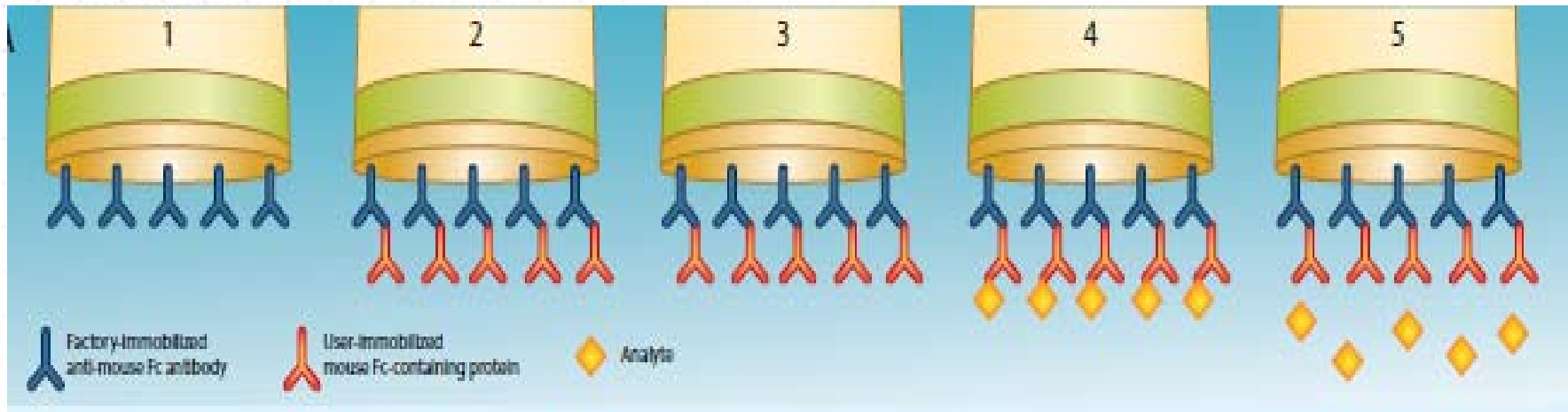


Real time approach : More Informative

	Association	Dissociation	$K_D = \text{Affinity}$
Example A	1.00E+04	1.00E-02	1.00E-06
Example B	1.00E+02	1.00E-04	1.00E-06



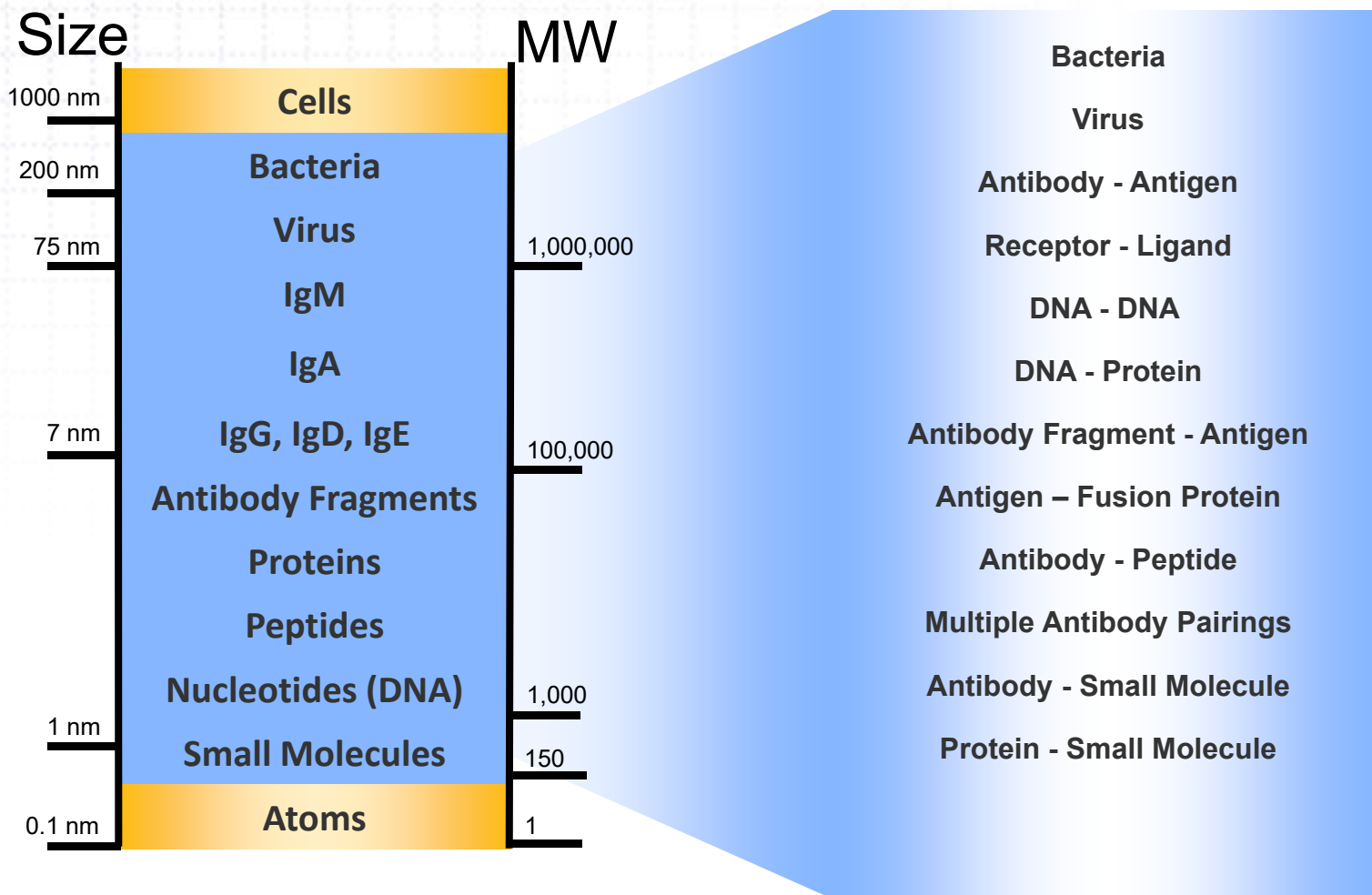
Immobilization & observation



- Protein Concentration
- Association rate
- Dissociation rate
- Binding Affinity

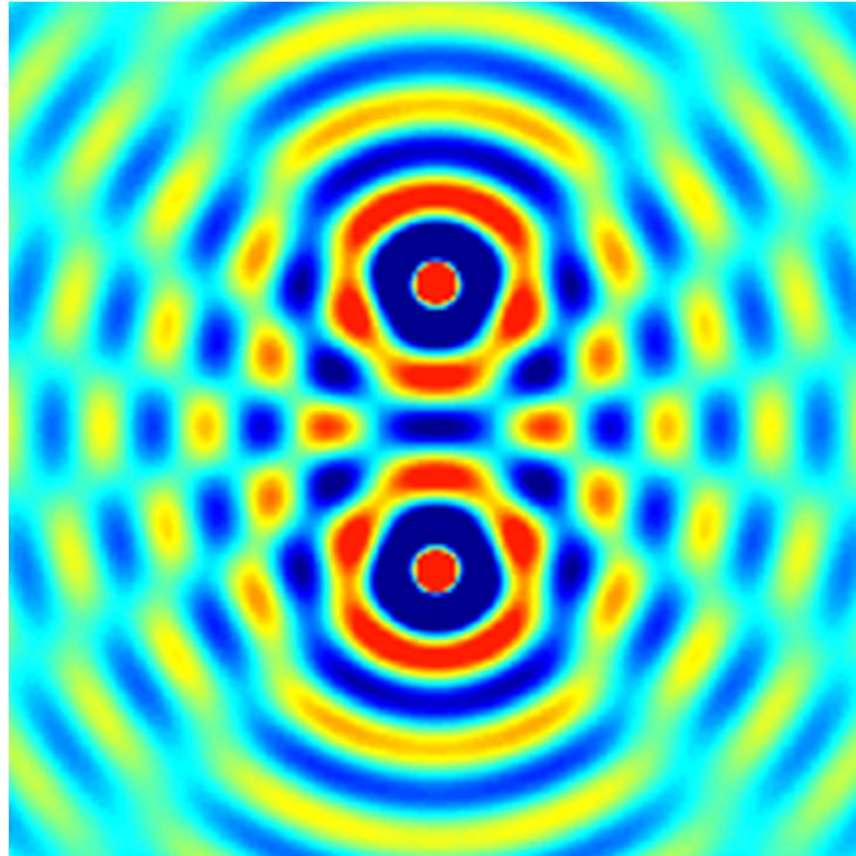
Myszka et. al. *Direct Comparison of Binding Equilibrium, Thermodynamic and Rate Constants Determined by Surface- and Solution-based Biophysical Methods*
Protein Science 2002, 11 (5), 1017-1025.

Large application range

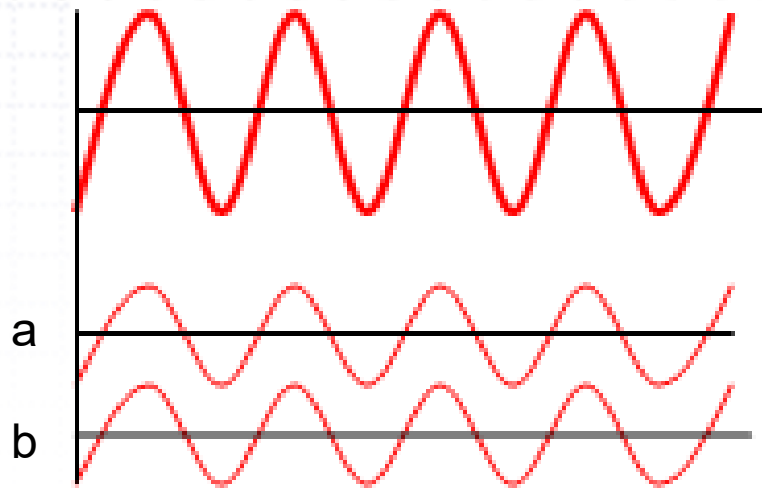


Current Octet assays within blue area

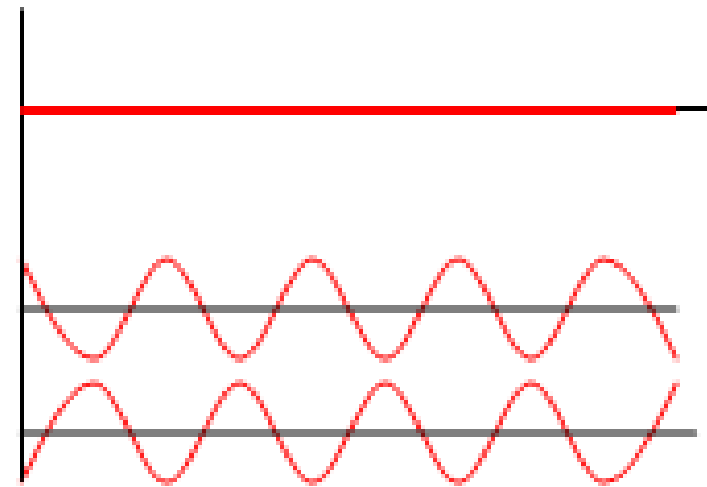
What is Interferometry?



Measuring of interactions between waves

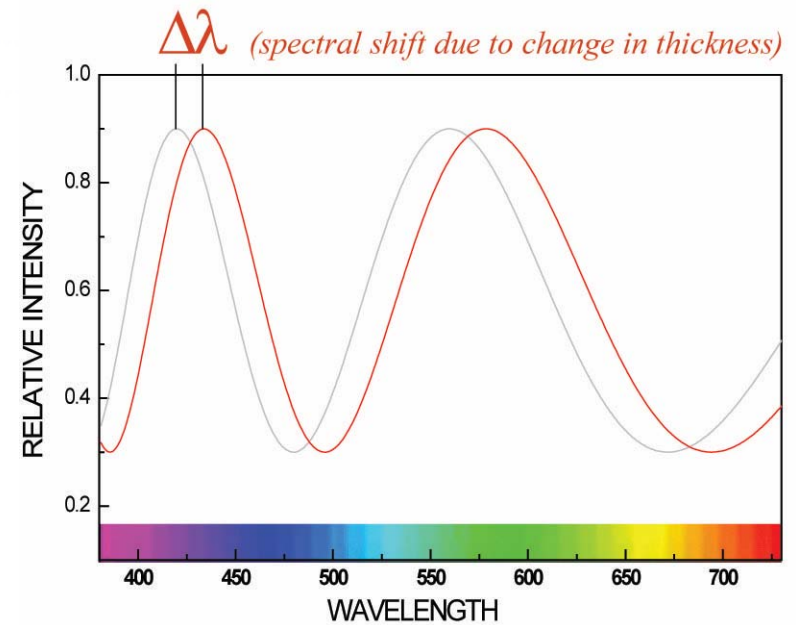
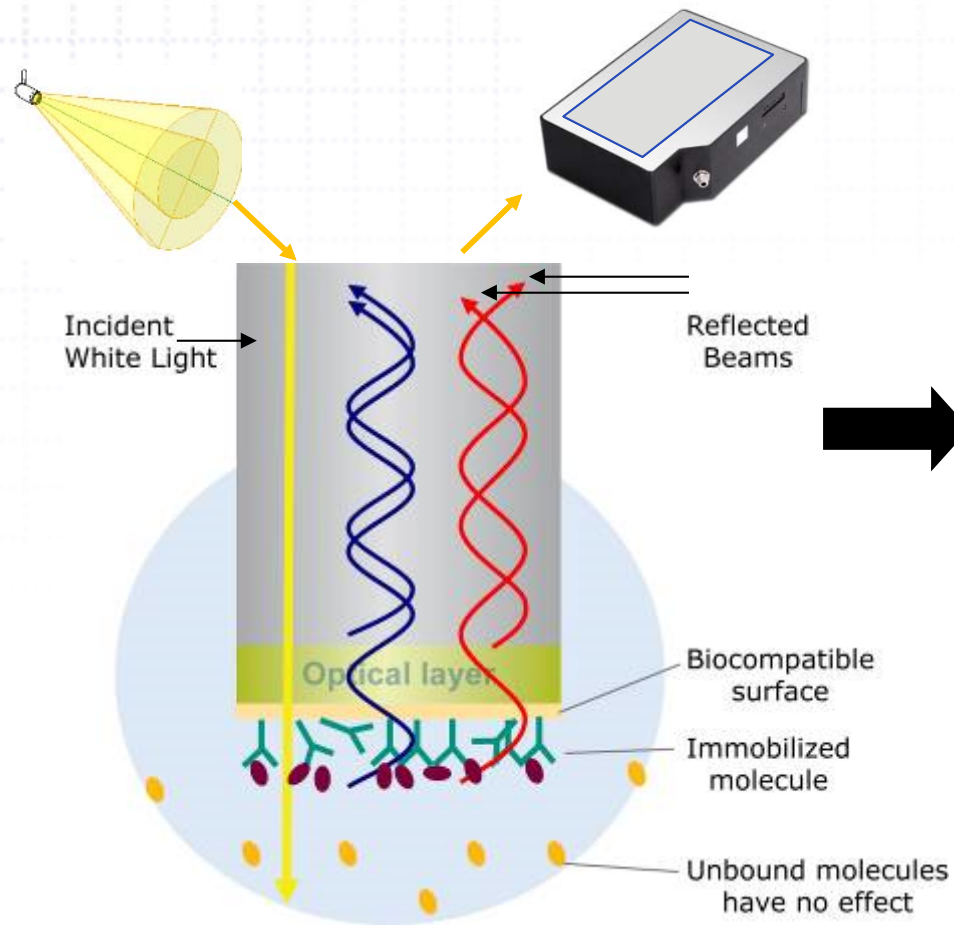


Two waves in phase
→ Constructive interference

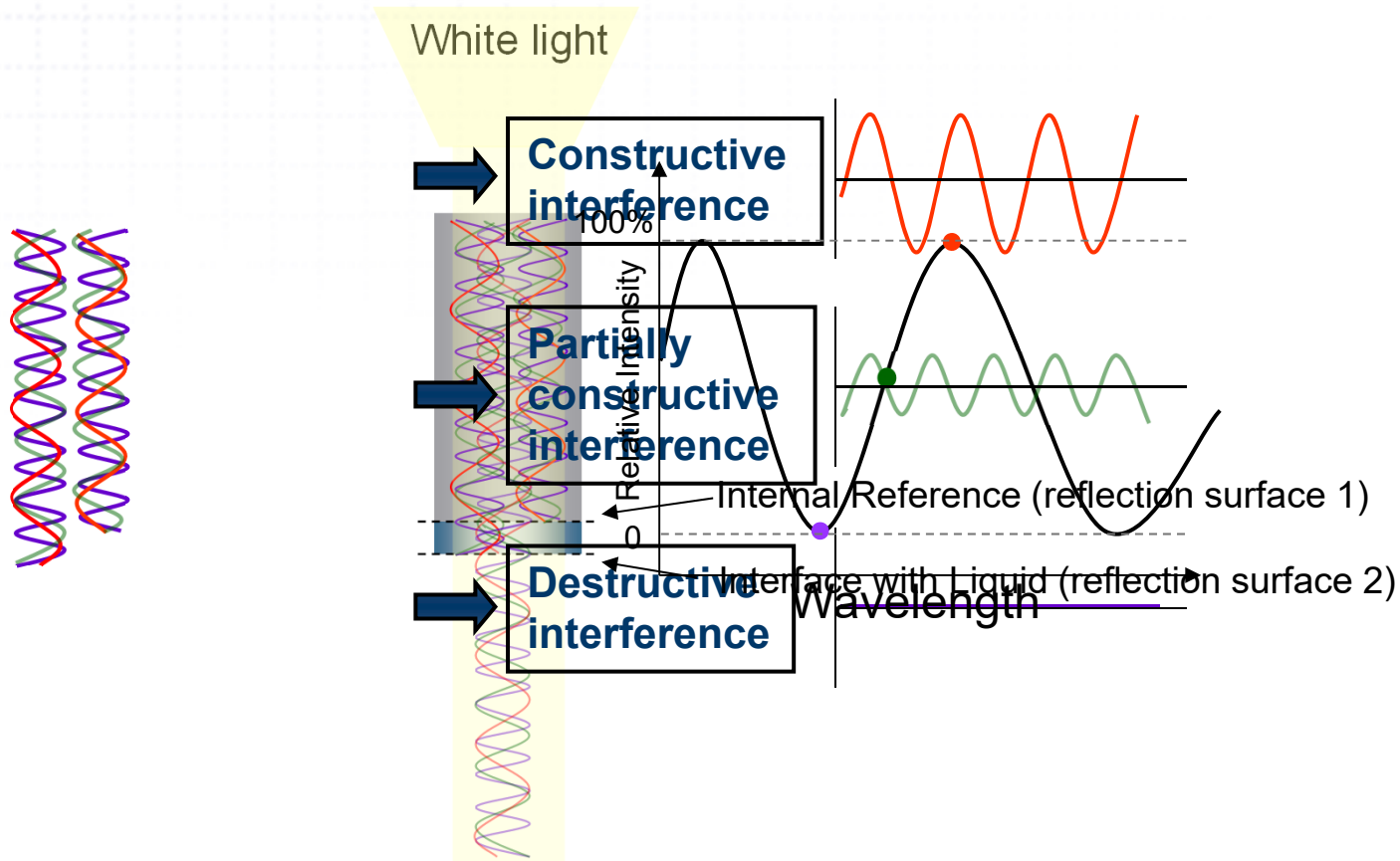


Two waves 180° out of phase
→ Destructive Interference

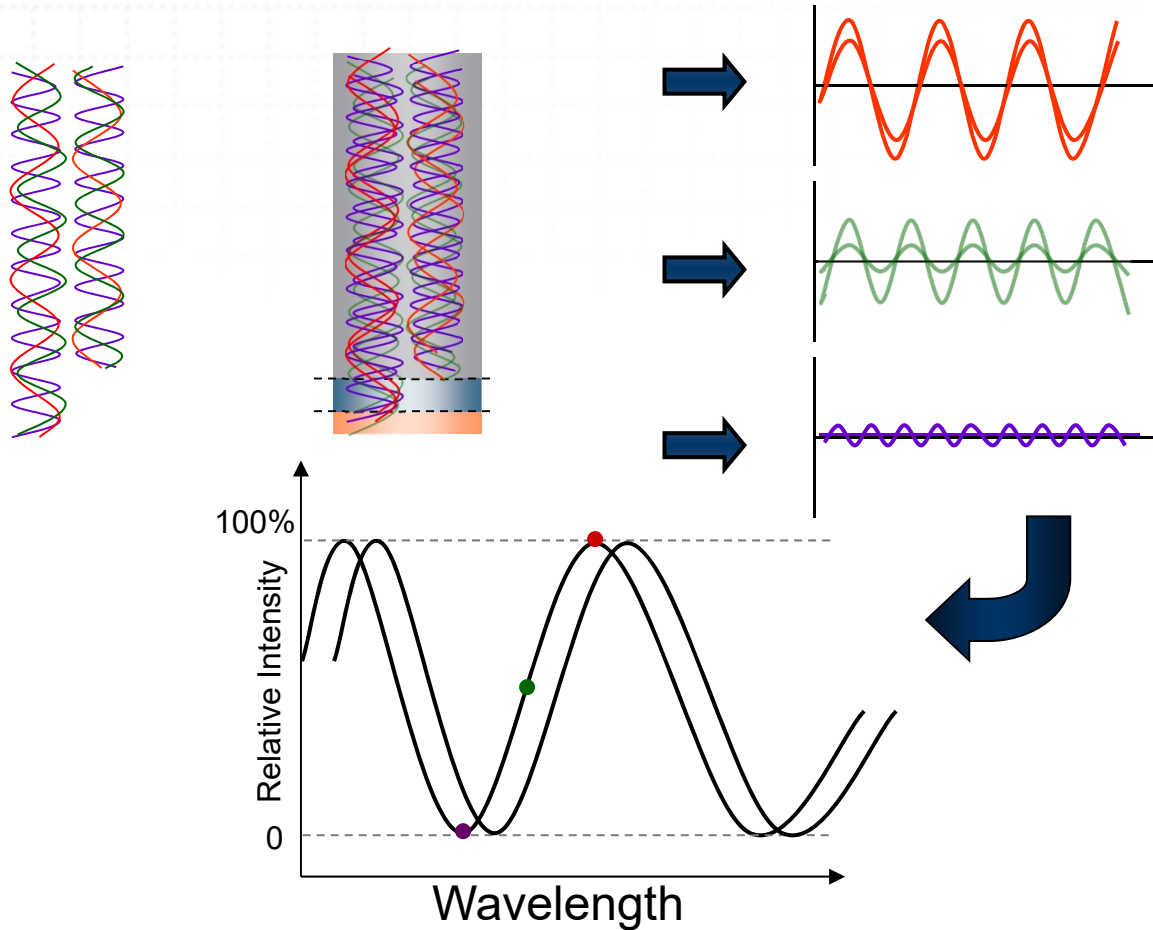
Measuring of interactions between waves



Biolayer Interferometry Principle



Biolayer Interferometry Principle

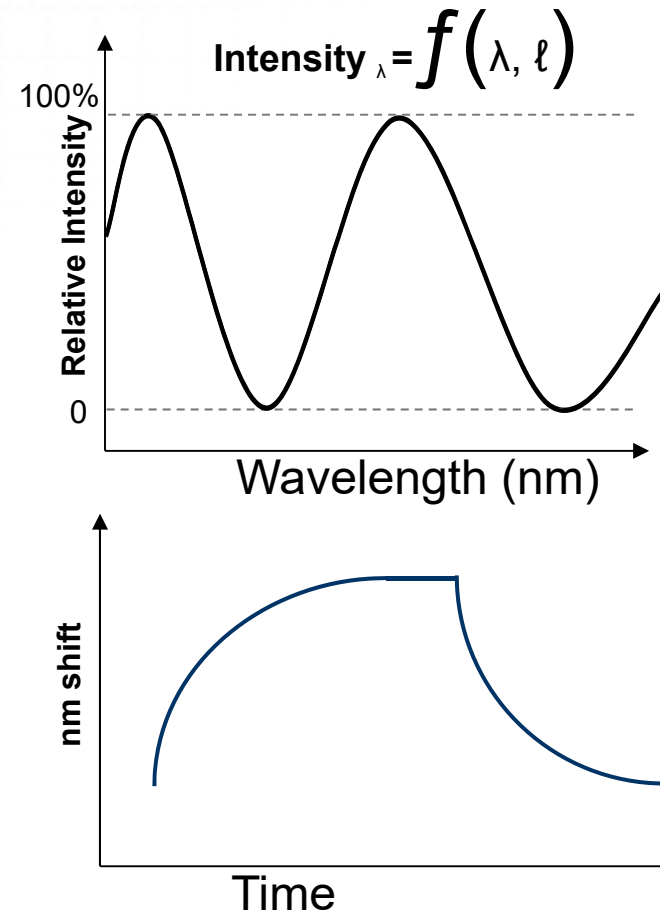


Monitoring the Interference Pattern vs. Time Provides Kinetic Data

Distance
between the
two reflecting
surfaces = ℓ



- Kinetics
- Affinity
- Quantitation



Generic Biosensors:

- Streptavidin (SA)
- Super Streptavidin (SSA)
- High Precision Streptavidin (SAX)
- Amine Reactive 2nd Generation (AR2G)
- Aminopropylsilane (APS)

Tag Biosensors:

- Anti-Penta-HIS (HIS)
- Ni-NTA (HIS)
- Anti-GST

Current Biosensor Kits:

- Immunogenicity (ADA)
- Residual Protein A
- CHO HCP

Antibody Biosensors:

- Anti-Human IgG Fc (AHQ)
- Anti-hIgG Fc Capture Surface (AHC)
- Anti-Murine IgG Fv (AMQ)
- Anti-Murine IgG Fc Capture (AMC)
- Protein A
- Protein G
- Protein L
- Anti human IgG Fab

Custom Sensors

- Custom Capture Sensor
- Custom HCP kit



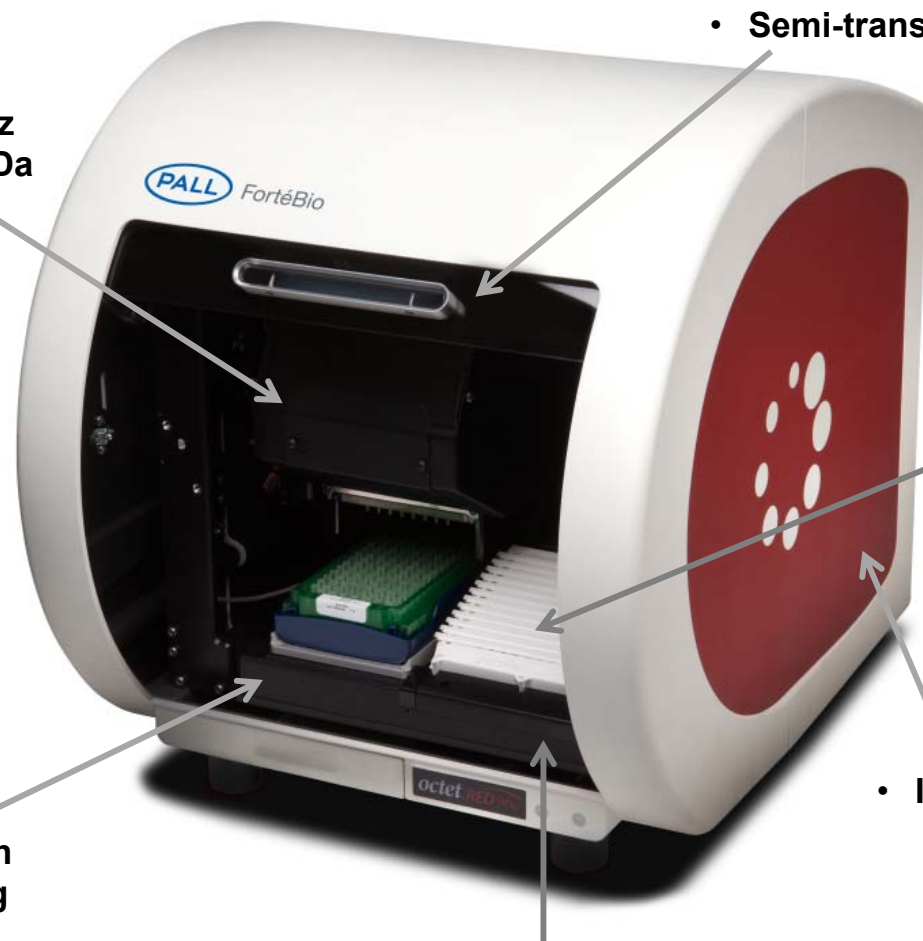
Octet RED96e System

- 8-channel BLI system with an updated look to Octet enclosure and a semi-transparent door
- Expanded assay temperature control range 15°C-40°C
- Optional evaporation cover to minimize sample evaporation rate, extending unattended run time to 12 hours
- Installed with the new Data Analysis HT v10.0 software for multi-experiment analysis and customizable experiment report
- Lift bar addition for ease of installation
- Integrated power supply for a more compact footprint



Octet RED96e System Features

- 8-channel read head
- Acquisition rates: 2, 5 or 10 Hz
- Analyte MW for kinetics: 150 Da
- K_D range: 1 mM – 10 pM



- Semi-transparent cover

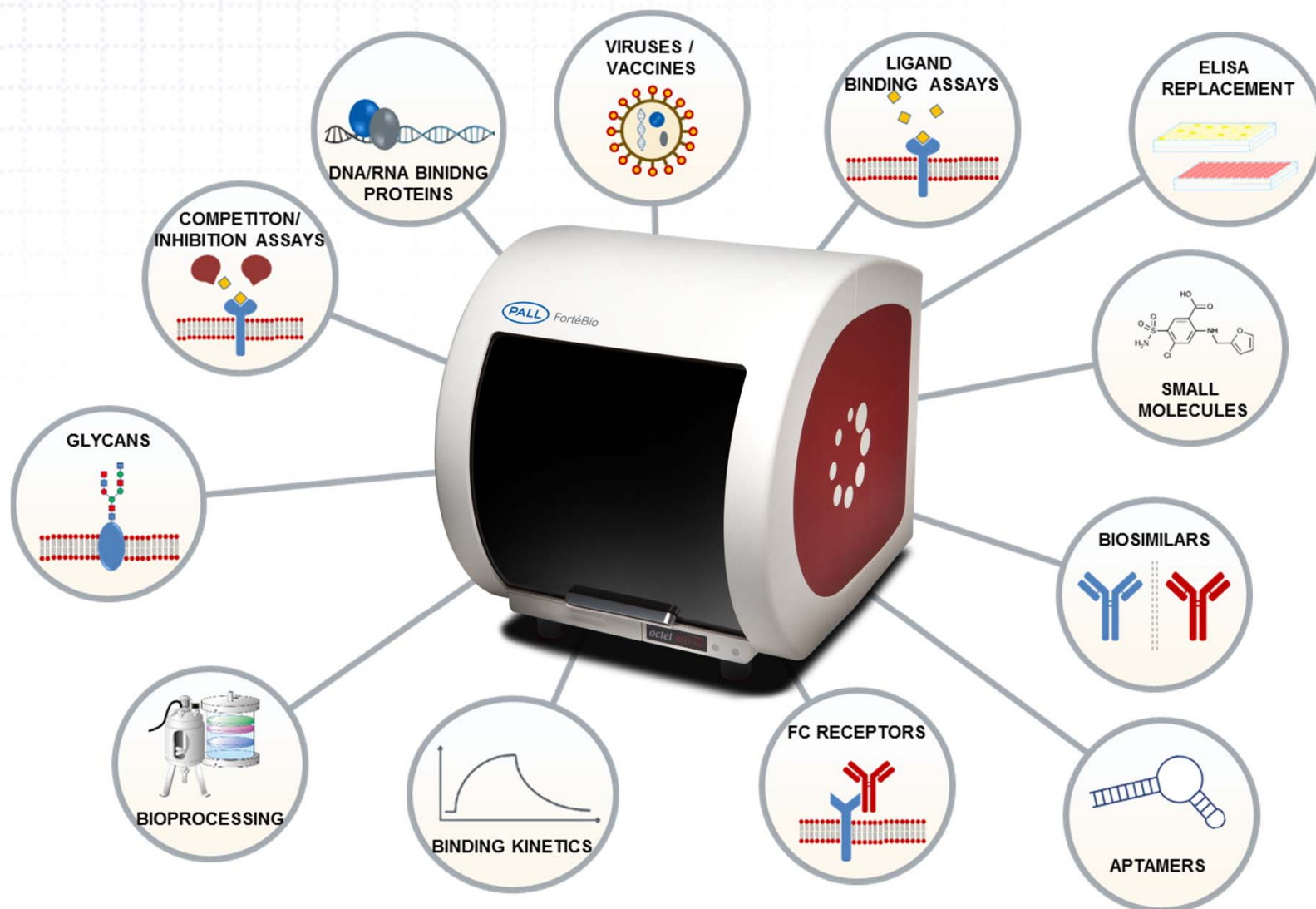
- Sample evaporation control

- Integrated power supply

- 1 Biosensor tray position
- Allows sensor re-racking

- Temperature control: 15 – 40 °C
- Orbital flow: 0 – 1500 rpm

Broad Utility of Octet RED96e System



- **Higher throughput, fast workflow**
 - Concentration measurements **faster than ELISA or HPLC**
 - Screen more samples in less time than SPR
 - **Little** dilution or **sample preparation required**
 - **Full sample recovery**
 - Fixed volume allows long on/off rate determinations
- **Unparalleled ease-of-use**
 - Multiple users are easily trained to operate the system
 - **Microfluidics-free** format and disposable biosensors
 - **No maintenance**, cleaning, or instrument prep time required
- **Versatility**
 - **Crude samples** can easily be measured
 - **Wider solvent** tolerance and assay flexibility
 - **Non-destructive** analysis
- **Price**
 - **Lower capital** acquisition cost than traditional SPR systems
 - Lower usage/maintenance costs SPR systems
 - **Lower cost per sample** than Protein A HPLC or commercial ELISA kits



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