

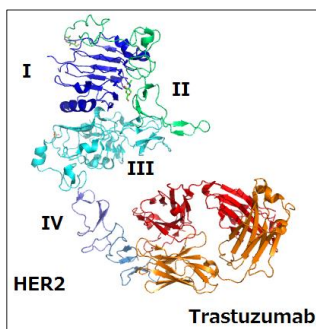
Affinity and Kinetics of Antibodies in Serum

Creoptix™ WAVE



Summary

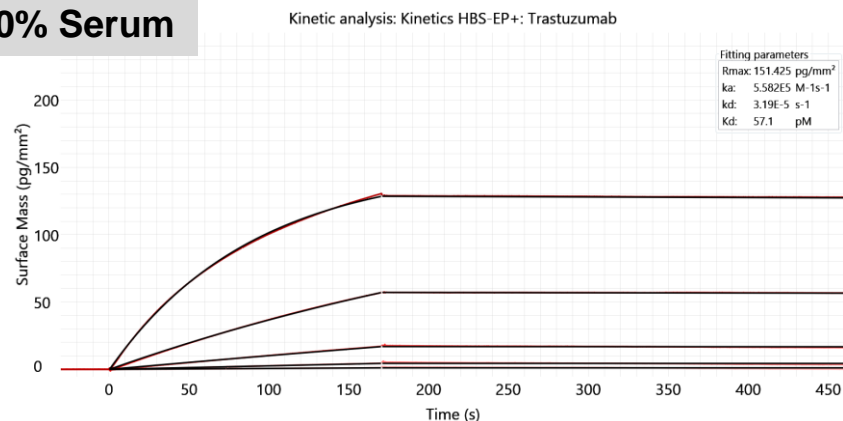
Analysis of molecular interactions in complex matrices like blood serum is of great interest, especially for therapeutic antibodies. Label-free surface-based biosensors are typically put under stress by such complex matrices due to potential deleterious effects on the microfluidics, and high non-specific binding of the matrix components (e.g. serum albumines) to the surface. Here we show that the innovative disposable microfluidics of the **Creoptix™ WAVE** system provides very high tolerance against blood serum both with regard to clogging but also regarding kinetic analysis. This enables the accurate determination of eg. antibody kinetics in nearly pure serum.



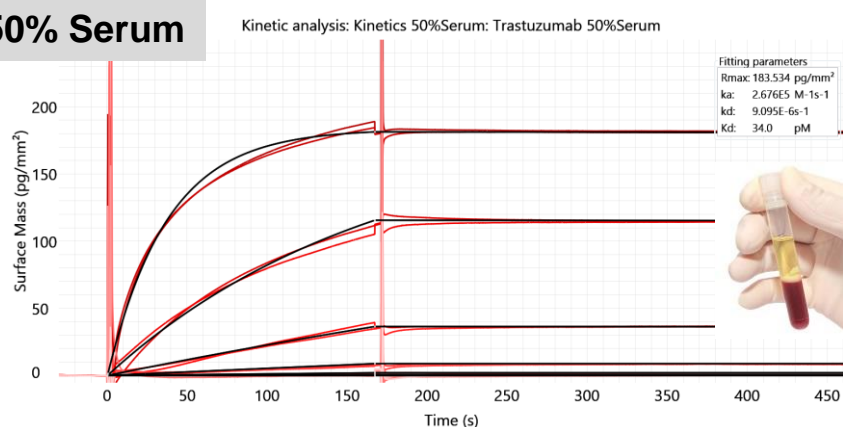
Legend: Sensorgrams of the interaction between HER2 and Trastuzumab in different matrices.

HER2FC fusion (Sino Biological, CN) was immobilized on the reference channel at 975pg/mm² via amine coupling followed by BSA (Roche, CH) on both channels to reach ~4200pg/mm² of total protein per channel. After few startup cycles injecting running buffer (RB; HBS-EP+ 0.5%BSA for all the matrices used), Trastuzumab (Absolute Antibody, UK) was injected at different concentrations on both channels at 150µl/min for 170s followed by 300s of dissociation. Regeneration was achieved with a pulse of 50mM NaOH, 1M NaCl, followed by one RB injection (plus one Serum injection at the respective concentration for 50% Serum and 90% Serum). Trastuzumab was injected in a 1:4 dilution series (5 dilutions from 20nM for 0% Serum, 8 dilutions from 100nM in duplicate for 50% and 90% Serum). Trastuzumab dilutions were prepared in RB for 0% Serum, in 1:1 Serum:RB for 50% Serum or 9:1 Serum:RB for 90% Serum. Measurements were adjusted using DMSO calibration, double referencing (blanks of the respective Serum dilution were injected just before each Trastuzumab injection in 50% and 90% Serum) and bulk correction during evaluation.

0% Serum



50% Serum



90% Serum

