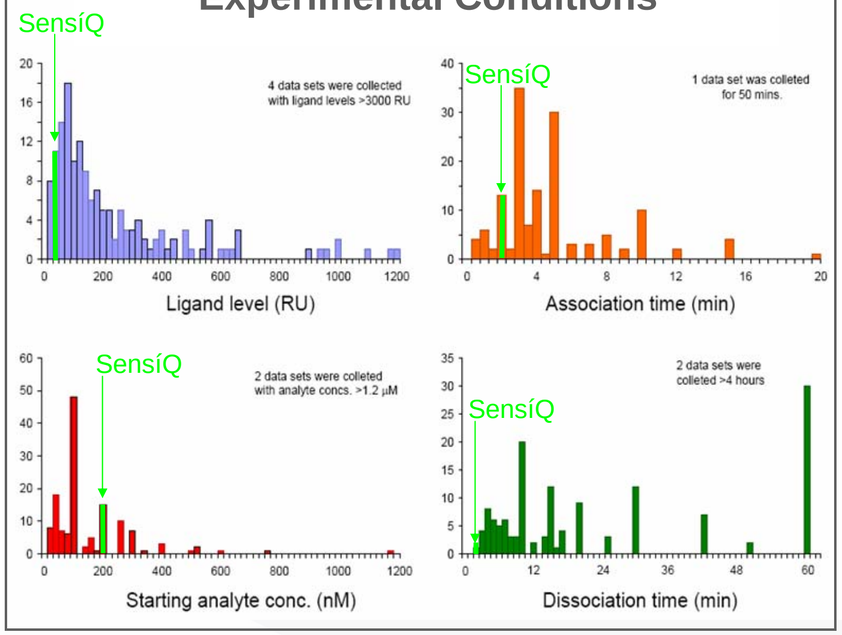
# Myszka's Benchmark Study: Over 100 Participants

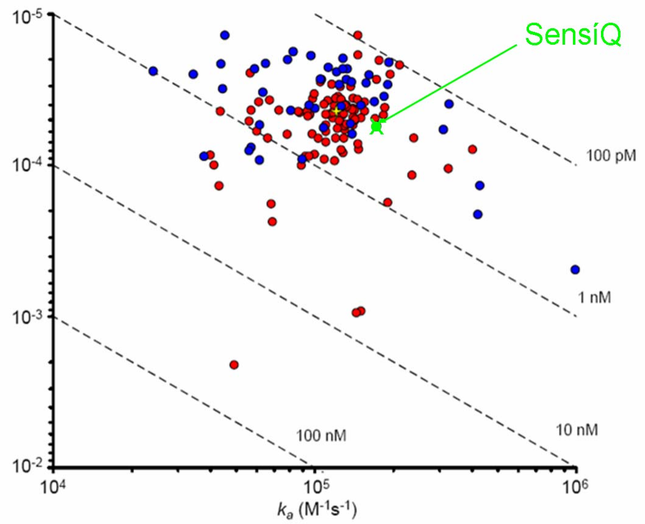
Presented at DiPIA, 2007


## Main Findings

### Experimental Conditions



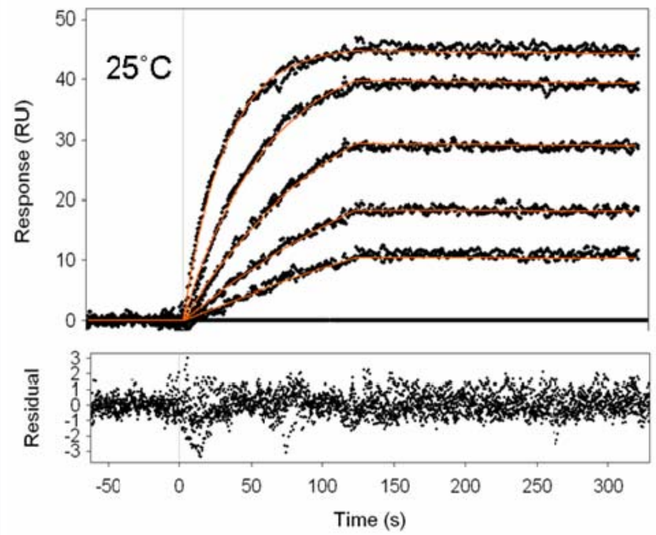

### Distribution of Kinetic Constants




| | Ligand = Fab | | Ligand = GST-antigen | |
|---|---|---|---|---|
| | Mean | 95% | Mean | 95% |
| $k_a$ ($M^{-1}s^{-1}$) | $1.1x10^5$ | 1.7 to $0.9x10^5$ | $1.2x10^5$ | 1.33 to $1.11x10^5$ |
| $k_d$ ($s^{-1}$) | $3.0x10^{-5}$ | 6.1 to $2.0x10^{-5}$ | $4.9x10^{-5}$ | 14 to $2.0x10^{-5}$ |
| $K_D$ (pM) | 305 | 430 to 180 | 470 | 652 to 210 |

### SensíQ

**Global fit to a Simple 1:1 Interaction Model**

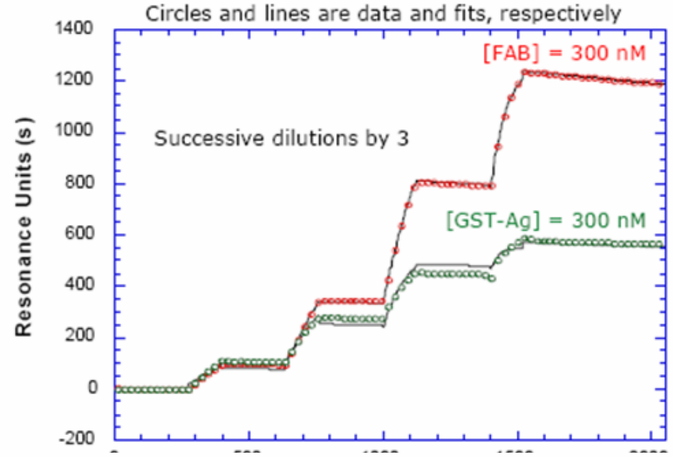



| | |
|---|---|
| $k_a$ | $1.70 \times 10^5$ ($M^{-1}s^{-1}$) |
| $k_d$ | $5.4 \times 10^{-5}$ ($s^{-1}$) |
| $K_D$ | 0.313 nM |
| $R_{max}$ | 45 (RU) |
| ResSD | 0.74 (RU) |

### BIACORE 3000

- GST-antigen 2954 RU on channel 1
- FAB 551 RU on channel 2
- regeneration 10 mM glycine pH 2

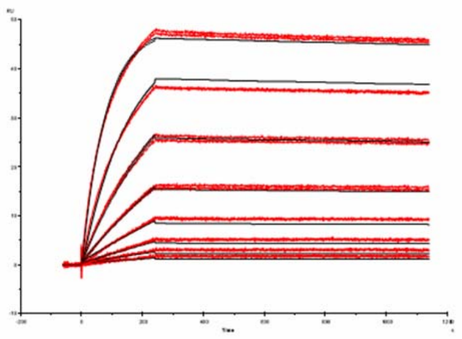
Circles and lines are data and fits, respectively




| Protein injected | | $k_{on}$ $M^{-1}s^{-1}$ | $k_{off}$ $s^{-1}$ | Kd nM |
|---|---|---|---|---|
| FAB | 1st exp | $6.6\ 10^4$ | $6.6\ 10^{-5}$ | 1.0 |
| | 2nd exp | $5.7\ 10^4$ | $7.6\ 10^{-5}$ | 1.3 |

### T100

Full Concentration Series



- **Instrument**: Biacore T100, CM5 chip
- **Method**: FAb immobilized at 43RU, GST-Ag in solution, regen with10mM glycine pH2.0
- **Analyte Conc.**: 1.56 -200nM (8) tested in replicates
- **Model**: 1:1 Langmuir with mass transport limitation
- **Model Fit**:

| | |
|---|---|
| **Rmax** | 48.6 RU (83% $R_{max,theoretical}$) |
| **ka** | $6.33x10^4$ $M^{-1}sec^{-1}$ |
| **kd** | $3.28x10^{-5}$ $sec^{-1}$ |
| **KD** | $5.19x10^{-10}$ M |
| **kt** | $4.65x10^{13}$ RU $M^{-1}sec^{-1}$* |
| **chi²** | 0.737 $RU^2$ |

**Select Data Sets**