COMPARISON OF OPTICAL AND ACOUSTIC WAVE PHAGE BIOSENSORS

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A dual channel SPREETA ™, a scaled down surface plasmon resonance (SPR) sensor and a thickness shear mode sensor (TSM) were used to study binding of the β-galactosidase (β-gal) to the phage immobilized to the gold surface by a physical adsorption. [1] Landscape β-gal-binding phage [2] and β-gal from *E.Coli* were used as probe/analyte system. While batch mode sensing was employed for the TSM sensor surface, a flow through mode was used to deliver the solutions to the surface for the SPR sensor.

The SPREETA ™ sensor is a highly integrated unit based on Kretschman geometry containing several components such as a light-emitting diode with a wavelength of 825 nm, a polarizer, a thermistor and two silicon photodiode arrays, all closely integrated with each other. A cleaned, gold surface of the sensor was exposed to a phage suspension at a concentration of 3.2×10^{11} virions/mL until saturation was achieved (in approximately 3 hours) and followed by washing with Dulbecco's phosphate buffered saline (DPBS). Bovine serum albumin (2 mg/mL) was utilized to block the uncovered sensor surface. The sensor was then exposed to graded concentrations of β-gal solutions with intermediate washes of DPBS, and the changes in the refractive index were recorded. Binding studies of phage to β-gal using TSM sensor was conducted as described earlier. [1]

Dose response plots from SPR and TSM sensor experiments are shown in Fig 1. Curve A represents the mean values of steady state refractive indices change as a function of increasing concentrations of β-gal obtained from an SPR sensor. The signals were normalized at the maximal refractive index change of 3.6×10^{-5}. The smooth curve is the sigmoid fit to the experimental data ($\chi^2=8.2\times10^{-4}$, $R^2=0.99$). Curve B represents the mean values of steady-state output voltages as a function of increasing concentrations of β-gal obtained from a TSM sensor. The smooth curve is the sigmoid fit to the experimental data ($\chi^2=2.3\times10^{-3}$, $R^2=0.99$). The signals were normalized at the maximal voltage change of 0.43 Volts.

Hill plots obtained from binding isotherms [2] for a SPR and TSM sensors are shown in Fig 2. The ratio of occupied and free phages is shown as a function of β-gal concentrations. [2] The upper straight line is the linear least squares fit to the SPR sensor data (R=0.99, slope=0.73±0.03). The bottom straight line is the linear least squares fit to the TSM sensor data (R=0.98, slope =0.32 ±0.03).

EC_{50} for the SPR sensor is about 5 times smaller than that for the TSM sensor, while apparent dissociation constants (K_d) are not significantly different (Table 1). The Hill coefficients and time constants of signal responses (τ) are larger for SPR sensors. This may be due to the flow through mode employed on an SPR sensor. Results obtained for binding of β-gal with immobilized phage compare well with those for β-gal-specific antibodies (data not shown).

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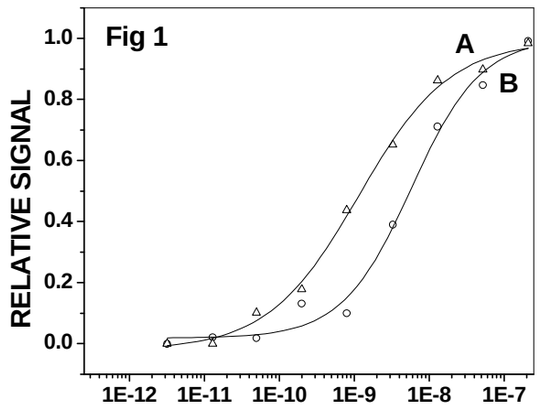
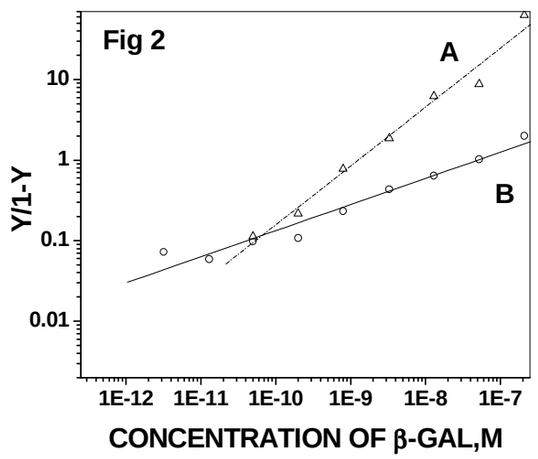
Table 1

Method	EC_{50}, nM	Hill coef.	K_d, nM	τ, min
SPR	1.2±0.2	0.73±0.05	1.1±0.2	45
TSM	5.8±1.4	0.32±0.03	1.7±0.5	22





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2. V. Petrenko, V. Vodyanoy, *J. of Microbiological Methods*, 53, 253-262(2003).