

Amplification Free detection of Herpes Simplex Virus DNA from Cerebrospinal Fluid

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Supplementary Information

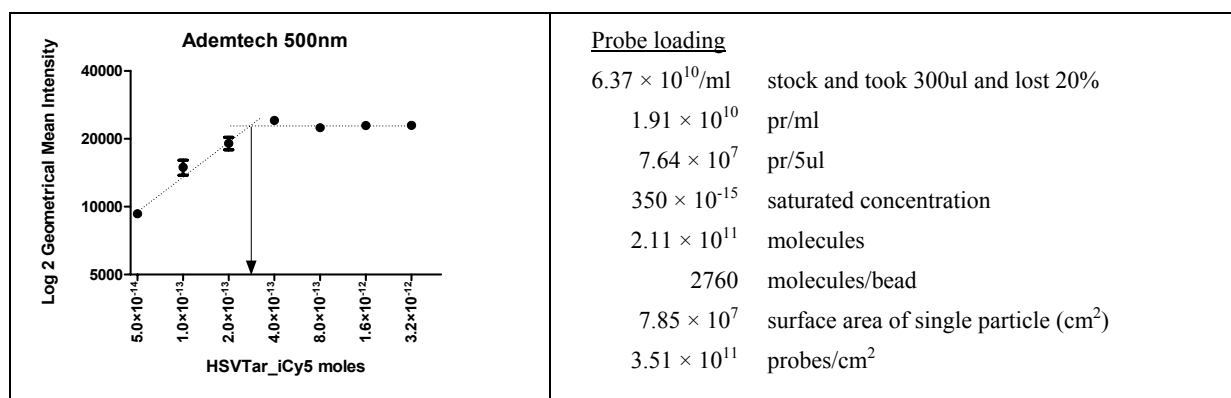
Ademtech bead calculations

Ademtech quotes surface area equals $5\text{m}^2/\text{g}$ for 500nm Masterbeads. Particles are supplied as $10\text{mg}/\text{mL}$ therefore $0.05\text{m}^2/\text{mL}$. Taking the assumption particles are 250 nm radius the number of particles per mL equals 6.37×10^{10} (total area/surface area of each particle). Beads specified to have a biotinylated oligonucleotide binding capacity $\sim 200\text{pmol}/\text{mg}$ (quote $\gg 175\text{pmol}/\text{mg}$ and so $200\text{pmol}/\text{mg}$ taken for optimisation). This equals $2000\text{ pmol}/\text{mL}$, 1.89×10^4 biotins/particle, 2.4×10^{12} sites/ cm^2 . Calculations for numbers of beads were made assuming 20% loss in coupling.

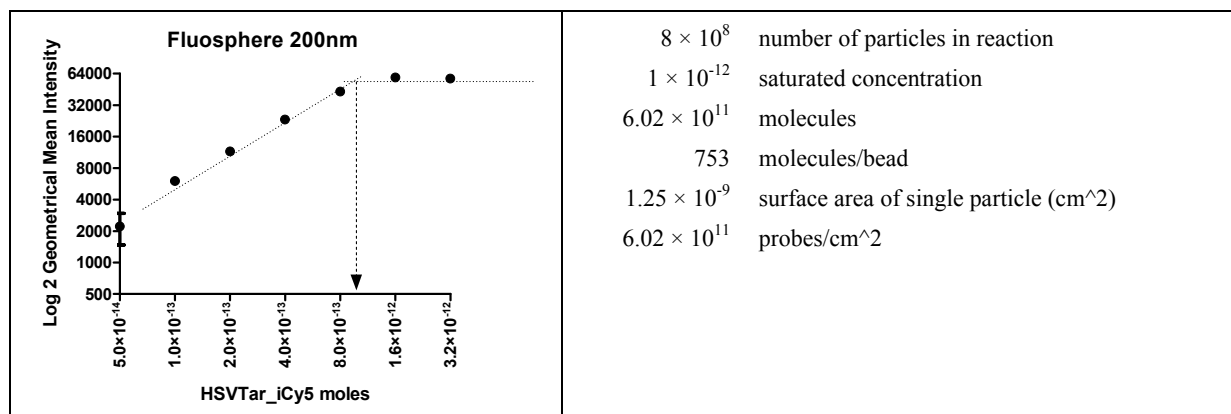
Fluosphere Calculations

Calculation of number of particles per ml calculated from equation given in Invitrogen document (mp0500.pdf). # microspheres /mL = $(6 \times \text{particle concentration} \times 10^{12}) / (\text{density} \times 3.142 \times \text{diameter}(\mu\text{m})^3)$ yielding 4.54×10^{12} particles/mL. Invitrogen quotes 0.1 – 2 meq./gram (millimol/gram) of carboxylic acid.

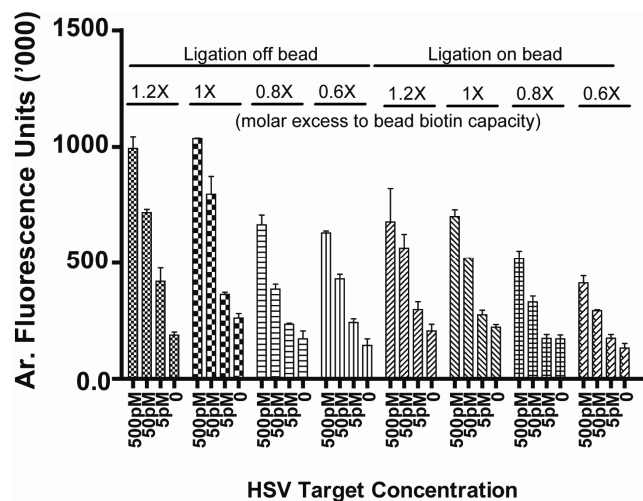
Flow cytometry data for probe density calculation



Sup Fig. 1 Flow cytometry saturation results for Ademtech 500nm beads



Sup Fig. 2 Flow cytometry saturation results for Fluospheres nanoparticles



Sup Fig. 3 Ligation of the HSV capture probe conducted off the bead prior to coupling gives a stronger and more robust signal and dose response compared with ligation conducted on the bead. The amount of linker added is also compared showing increased signal with more linker added to the coupling reaction.