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Fluctuation correlation spectroscopy (FCS) in turbid media: Detection of somatic cells and bacteria in body fluids

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Fluctuation correlation spectroscopy (FCS) in turbid media: detection of somatic cells and bacteria in body fluids.

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Abstract

We seek an alternate, simple, and cheap method to analyze cells and bacteria in body fluids for clinical and biotechnological applications; a task commonly performed using a flow cytometer, a large, expensive instrument. FCS can detect and quantify single molecules in small volume. Single molecule sensitivity is obtained when small volumes (~ 0.2 fL) of excitation is achieved. Best sensitivity is when one molecule is contained in that volume, i.e., for concentrations of ~ 10 nM and the range spans ~ 1 uM to ~ 10 pM. Volume of observation can be increased for better sensitivity, but background fluorescence or impurities limit the lowest detectable concentration. For analysis of body fluids, sensitivity on the order of 100-100000 particle/ml is needed. Bacteria or somatic cells stained with fluorescent dyes give large fluorescence signals, but volume must be increased by $\$10^8$ to have about one particle in the observation volume. Also, diffusion does not produce fast enough fluctuations. We constructed a small, portable, easy to use, and relatively inexpensive FCS instrument (designed to measure somatic cells in milk) with high sensitivity and high dynamic range. The sample is rotated at high speed to effectively scan a large volume. Background signal is analyzed to provide accurate values of the number of cells in the physiological range. Support: NIH, PHS P41 5 RR03155.