

# NANO BIOSENSORS

FOR

## IN-VIVO ANALYSIS OF SINGLE CELLS

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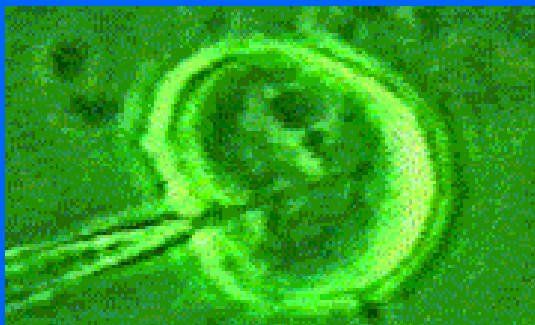
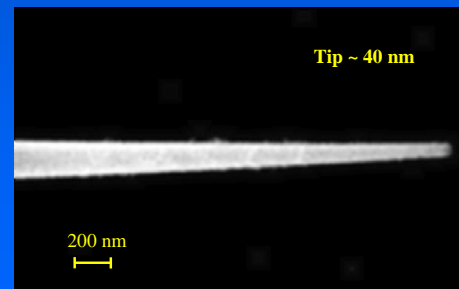
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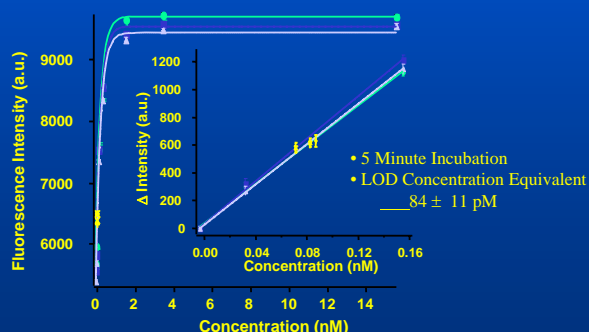
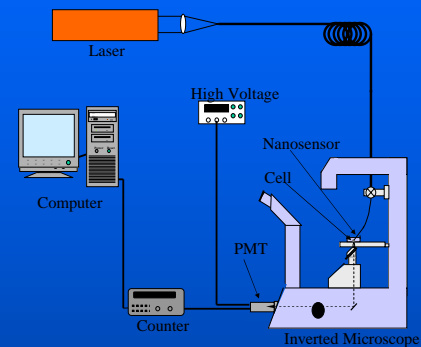
We have developed nanometer-scale fiber optic biosensors capable of being inserted into individual living cells and obtaining quantitative measures of toxic chemicals using fluorescence detection. These nanosensors are constructed by pulling an optical fiber to approximately 40-nm in diameter at the tip, and then coating the sides with silver. The tips of the fibers are silanized and bioreceptors (e.g., antibodies, DNA, peptides) to target species of interest are attached. The nanosensors have been used to reproducibly measure concentrations of targeted biospecies (e.g., benzopyrene tetrrol, (a metabolite of the carcinogen benzo[a]pyrene) in single cells at picomolar concentrations.

- Pulled nanosensors have tip diameters of approximately 40 - 50 nm
- Final coated fibers are approximately 200 nm diameter
- Antibody coated tips for specificity in binding
- Nanometer diameter tip provides near-field excitation



- Nano-size fibers are small enough to be placed within subcellular compartment (e.g. nucleus, mitochondrion)
- Small size of nanosensor allows for insertion into typical mammalian cells with no apparent damage
- After insertion and removal of nanosensor from individual cells, they continue to perform routine functions such as mitosis

- 325-nm line of a HeCd laser used for excitation of BPT and BaP
- Inverted microscope used for monitoring insertion of fiber optic probe into single cell
- Fluorescent light is collected by microscope objective and detected using a photon counting photomultiplier tube (PMT)



- Capable of detecting only a few molecules of BPT in a single cell
- Near-field excitation allows measurements to be performed directly inside the cell with negligible background
- Excellent sensitivity and reproducibility from fiber to fiber
- Picomolar detection sensitivity

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