Carbon Nanotubes Improve Protein Array Detection Limits

November 20 2008

(PhysOrg.com) -- To detect cancer as early as possible, dozens of research groups are developing methods to detect trace levels of cancer-related proteins and genes in blood or other biological samples. Those efforts should get a boost thanks to new research results showing that carbon nanotubes can serve as incredibly sensitive optical labels for use in a wide variety of assay systems.

Reporting its work in the journal *Nature Biotechnology*, a research team headed by Hongjie Dai, Ph.D., Stanford University and the Center for Cancer Nanotechnology Excellence Focused on Therapeutic Response, describes a new type of coating developed specifically for attaching any number of different types of targeting agents to the surface of single-walled carbon nanotubes. This coating, a branched form of the biocompatible polymer poly(ethylene glycol) (PEG), enabled the investigators to readily couple antibodies to carbon nanotubes. In the experiments reported in their current paper, the antibodies were designed to identify specific proteins immobilized on a standard protein array microchip.

Carbon nanotubes can function as bright Raman optical tags that are readily detected when irradiated with light. Experiments comparing the lower limits of protein detection using an antibody-labeled carbon nanotube tag and a standard fluorescence tag showed that the carbon nanotube-enabled assay was at least 1,000 times more sensitive than the fluorescence assay. At least part of this improvement resulted from the almost total elimination of background fluorescence that can confound

other detection schemes.

In addition, the investigators found that the Raman tags were useful over a larger range of concentrations, ranging from 10 nanomoles to 1 femtomoles. The investigators note in their paper that the coating they developed also should enable them to create Raman tags that can detect nucleic acids and other types of biomolecules.

Meanwhile, a second group of investigators, led by Beatrice Knudsen, M.D., Ph.D., Fred Hutchinson Cancer Research Center, and Selena Chan, Ph.D., Intel Corporation, has developed a mathematical technique for analyzing the specific spectral output of different Raman probes, making it possible to create highly multiplexed assays using these probes. Unlike traditional fluorescent labels that typically absorb and emit light in a very narrow band of frequencies, Raman probes generate complex frequency spectra that are chock-full of information.

The Knudsen-Chan team, which published its results in the journal ACS Nano, developed a method for sorting out the various spectral peaks associated with individual nanoscale Raman probes that were part of a mixture of these probes. Each probe was designed to bind to a different biomolecule. In one experiment, the investigators were able to decipher a complex Raman spectrum that included the optical emission from three different Raman probes and thereby determine the amount of each probe in the mixture. The researchers note that their method for spectral analysis is exceedingly simple to conduct and is amenable to high-throughput analysis in any type of multiplexed assay system.

The work by Dr. Dai and his colleagues, which is detailed in the paper “Protein microarrays with carbon nanotubes as multicolor Raman labels,” was supported by the NCI Alliance for Nanotechnology in Cancer, a comprehensive initiative designed to accelerate the application of nanotechnology to the prevention, diagnosis, and treatment of cancer.

An investigator from Tsinghua University in Beijing, China, also participated in this study. An abstract of this paper is available at the journal's Web site.

The work led by Drs. Knudsen and Chan, which is detailed in the paper "Spectral analysis of multiplex Raman probe signatures," was supported by the National Cancer Institute (NCI). An abstract of this paper is available at the journal's Web site.

Provided by National Cancer Institute