Magnetic Nanotags Spot Cancer in Mice Earlier Than Current Methods

November 17 2009

(PhysOrg.com) -- Searching for biomarkers that can warn of diseases such as cancer while they are still in their earliest stage is likely to become far easier thanks to an innovative biosensor chip developed by Stanford University researchers. The sensor is up to 1,000 times more sensitive than any technology now in clinical use, is accurate regardless of which bodily fluid is being analyzed, and can detect biomarker proteins over a range of concentrations three times broader than any existing method, the researchers say.

The nanosensor chip also search for up to 64 different proteins simultaneously and has been shown to be effective in early detection of tumors in mice, suggesting that it may open the door to significantly earlier detection of even the most elusive cancers in humans. The sensor also can be used to detect markers of diseases other than cancer. The researchers published their description of their magnetic nanosensor in the journal *Nature Medicine*.

"In the early stage [of a cancer], the protein biomarker level in blood is very, very low, so you need ultra-sensitive technology to detect it," said Shan Wang, Ph.D., of Stanford University and a member of the Center for Cancer Nanotechnology Excellence Focused on Therapy Response (Stanford CCNE). "If you can detect it early, you can have early intervention and you have a much better chance to cure that person."

Wang said the nanosensor technology also could allow doctors to rapidly determine whether a patient is responding to a particular course of chemotherapy. "We can know on day two or day three of treatment whether it is working or not, instead of a month or two later," he said.

The sensor that Wang and his colleagues created, which uses magnetic detection nanotechnology they had developed previously, can detect a given cancer-associated protein biomarker at a concentration as low as one part out of a hundred billion (or 30 molecules in a cubic millimeter of blood). Although the basics of the magnetic detection technology used in the new biosensor were described last year in a paper in the *Proceedings of the National Academy of Sciences*, the new sensor is not only more sensitive than the previous one by several orders of magnitude, it also outperforms its predecessor, as well as detection methods now in clinical use, in several other ways.

The most impressive performance gain detailed in the Nature Medicine paper is that the researchers have now demonstrated that the magnetic-nano sensor can successfully detect cancerous tumors in mice when levels of cancer-associated proteins are still well below concentrations detectable using the current standard methodology, known by the acronym ELISA. "That is a critical finding for us because it says that in a realistic biological application - that of tumor growth in mice - we can actually see tumors before anything else could have detected them," said Sanjiv Gambhir, M.D., Ph.D., principal investigator of the Stanford CCNE.

"I would say that the [first paper was] proof of concept of the technology, and the Nature Medicine paper is proof of concept of the technology working in a real-world application," he said. "It is one thing to have the technology show that it can work in principle; it is quite another to actually utilize it with real mouse blood samples from a real mouse growing a real tumor."

In the Nature Medicine paper, the researchers show that the new magnetic-nano sensor has a broad range of sensitivity, from part-per-billion levels to concentrations six orders of magnitude, or a million times, greater. The best existing analysis methods, or assays, in clinical use can detect proteins over a range of concentrations of at most two orders of magnitude.

Most of the sensing platforms currently in use are also limited to performing a single analysis at a time. To create a multiplexed assay, Wang and his colleagues attached the magnetic-nano sensors to a microchip in an array of 64 sensors, each of which can be set up to detect a different protein. As a result, the researchers can search for dozens of different proteins simultaneously during a single analysis. The new method is also faster than standard ELISA assays, with results typically available in one to two hours.

The researchers also demonstrated that the sensor is equally effective in every likely biological fluid, or matrix, that a doctor would want to analyze for cancer-associated proteins. Those fluids include urine, saliva, blood plasma (blood with the blood cells removed), serum (blood plasma with the factors that promote clotting removed) and cell lysates (the name applied to the cellular stew produced by dissolution of cells).

The key to the versatility of the magnetic-nano sensor and the broad range of concentrations it can detect lies in its use of magnetism and the wide range of ultra-sensitive magnetic detectors developed for the computer industry. The basic mechanism of detection employed in the magnetic-nano sensors is to capture proteins and disease markers using antibodies that naturally tend to bind to these molecules, known as antigens. The antibodies, dubbed "capture antibodies," are applied to a sensor, so that when the matrix of interest is placed onto the sensor chip, the appropriate antigens bind.

While the antigens are held fast, another dollop of antibodies is applied.

These antibodies are attracted to a different molecular region of the antigens held on the sensors, and when the second set of antibodies binds to antigens, they effective seal them in an antibody sandwich. The researchers then apply a wash containing magnetic nanoparticle tags that have been tailored to fit specific antibodies. The magnetic nanotags attach themselves to the outer antibody on the sandwich, where they alter the ambient magnetic field in a small but distinct and detectable way that is sensed by the detector.

Another virtue of the technology, Wang said, is that it uses existing technology already in use in the data storage and semiconductor industries. Because of that, "It can be made relatively cheaply. It is [very similar to] the same sensor you are using in a hard disk drive to read a hard disk back," he said.

One of the next steps in the research, Wang said, is to test the magnetic-nano sensors on human blood samples taken from a long-term study in which researchers took blood from subjects prior to any of them being diagnosed with cancer. To this end, the Stanford team will be collaborating with the Fred Hutchison Cancer Research Center in Seattle and the Canary Foundation, a nonprofit organization that focuses on early diagnosis of cancer.

This work, which is detailed in a paper titled, "Matrix-insensitive protein assays push the limits of biosensors in medicine," 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. Investigators from MagArray Inc., also participated in this study. An abstract of this paper is available at the journal's Web site.

Provided by National Cancer Institute (news : web)