The AptaReport™
Newsletter
SUMMER 2016

Aptamer Uranium Sensor

Uranium is a radioactive element best known for its utilization in weapons of mass destruction and the production of nuclear energy. Because of the immense damage that uranium is capable of causing to both human populations and the ecosystem in general, its detection in potentially contaminated water is vital to public safety. Although there are a number of highly sensitive methods for detecting Uranium in water samples, they require expensive and complicated instruments to yield results.

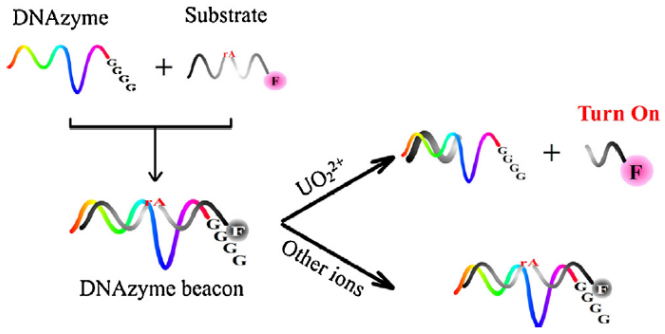

Figure (adapted)* The concept of DNAzyme sensor for trace UO_2^{2+} detection.

Thus, scientists at the East China Institute of Technology developed a DNAzyme sensor for highly sensitive uranium detection with the potential for on-site applicability. The DNAzyme binds to a fluorescently labeled substrate, and quenches the fluorescence until it is bound by uranium, at which point it cleaves the substrate, releasing the fluorophore. This fluorescent 'beacon' system enabled the scientists to detect uranium in water at 0.41 nM concentrations, well below the toxic concentration of 130 nM defined by the U.S. Environmental Protection Agency. Additionally, the scientists were able to detect and quantify uranium in ore, expanding the DNAzyme's usefulness to mining operations as well as public safety. This new sensor system displays the versatility of aptamers, and demonstrates the potential of aptamers for relatively cheap, on-site applicability in real-world situations.

-C.B.

*Reference: Apta-Index™ ID #598

Multivalent DNA Aptamer Displays Improved Binding Affinity

L-Selectin (or CD62L) is an adhesion molecule present on the surface of leukocytes that enables the cells to effectively home in on infected tissues in the body, and furthermore, recruit other leukocytes. Because of its role in facilitating inflammatory responses, inhibiting L-Selectin can serve anti-inflammatory functions. Inhibiting L-Selectin activity can be challenging due to the small size of most aptamers compared to protein targets. In addition, the length of time during which aptamers can survive an in vivo environment is limited, requiring high aptamer doses to effectively inhibit L-selectin activity. However, a multivalent complex of aptamers resolves a number of issues faced by aptamers meant for in vivo applications: multivalence increases the affinity of the aptamer for its target, and it increases stability of the aptamer in vivo. To that end, scientists have created a multivalent complex of an L-Selectin aptamer through Rolling Circle Amplification, and tested it both in vitro and in vivo for L-Selectin inhibition. Not only did the multivalent aptamer successfully inhibit L-Selectin homing functions in vivo, it also improved the affinity of the aptamer approximately a thousand-fold.

This new technique for rendering defined aptamers multivalent promises to improve the viability of aptamers as in vivo therapeutics, as well as increasing their effectiveness as diagnostic tools.

- C.B.

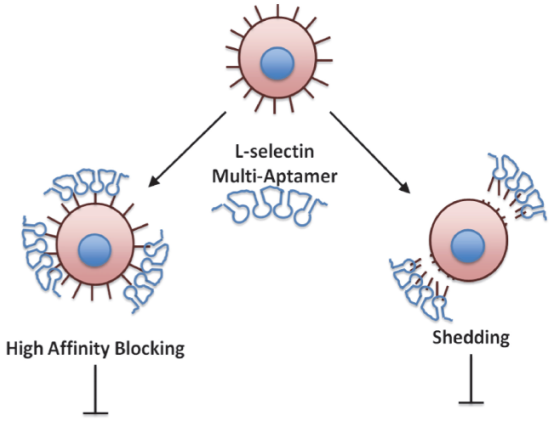

Figure (adapted)* Potential mechanisms of action of multivalent interactions with L-Selectin binding. It is hypothesized that the Multi-Aptamer may either inhibit L-Selectin function via high affinity, multivalent binding to L-Selectin, or by inducing shedding of surface L-Selectin via multivalent interactions.

*Reference: Apta-Index™ ID #599

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Colorimetric Aptamer Test for Malaria

Scientists at the Pohang University of Science and Technology, Korea, have successfully developed an aptamer-based colorimetric test for detecting a biomarker from two different species of malaria. The biomarker, known as Lactate dehydrogenase (LDH), is a protein that is highly expressed by both the infectious Plasmodium falciparum (Pf) and Plasmodium vivax (Pv) species of malaria, and so can be used to detect the presence of the parasites in humans. Using gold nanoparticles, a DNA aptamer called pL1—which specifically binds to LDH—and an additional component known as CTAB, the scientists were able to reach a limit of detection of 1.25 pM for PvLDH and 2.94 pM for PfLDH and render the results visible to the naked eye. CTAB, formally known as hexadecyltrimethylammonium bromide, is a positively charged cation that can be used to either aggregate gold nanoparticles (which then change color from red to gray), or form supramolecular nanostructures of unbound DNA aptamers. Thus, in the absence of LDH, CTAB will form complexes with the DNA aptamers in solution, leaving the gold nanoparticles dispersed and preventing any change in color.

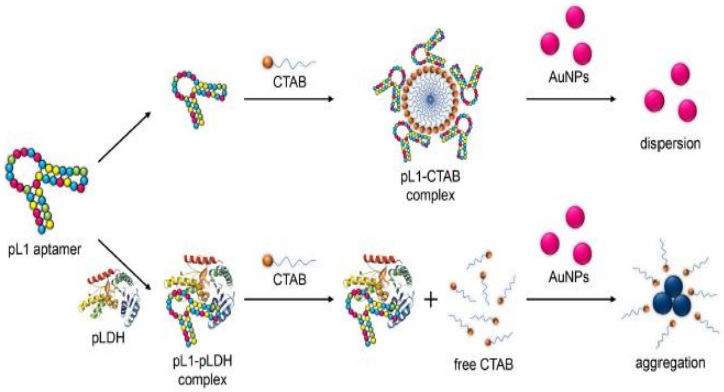

*Figure (adapted)** Schematic representation of the biosensor for the detection of pLDH based on the surfactant-induced aggregation of AuNPs.

In the presence of LDH, however, the pL1 aptamers will bind to their targets, preventing complexation with CTAB, which will then proceed to aggregate the gold nanoparticles, leading to the loss of red color, and the development of gray coloring. This new colorimetric test demonstrates the ability of aptamers and novel nanotechnology to enable scientists to rapidly and easily perform diagnostic tests at the point-of-care, and shows the tremendous potential for the use of aptamers in the future.

- C.B.

*Reference: Apta-Index™ ID #308, #309

APTAMER/Antibody Sandwich Detection System Targets Myoglobin for Heart-Attack Diagnosis

Acute myocardial infarction (AMI), or heart attacks, are one of the leading causes of death worldwide. Cardiac biomarkers are key in early diagnosis of AMI, allowing for quicker detection and response to the life-threatening event. Myoglobin is not a cardiac specific marker, but has been suggested to be a relatively good candidate for diagnosis of AMI. A biosensor was developed that uses a standard personal glucose meter to detect presence of myoglobin. This biosensor utilizes both a myoglobin antibody and a myoglobin aptamer, and relies on the fact that both can bind to their target simultaneously. The antibody was immobilized on a microplate, allowing for myoglobin in a sample to be captured and retained on the microplate. The aptamer was conjugated with the enzyme invertase, which produces glucose from sucrose. An 'antibody-myoglobin-aptamer sandwich' is formed when the aptamer-invertase complex attaches to the myoglobin-antibody complex. Therefore, when the microplate is exposed to sucrose, the amount of glucose formed is dependent on the amount of myoglobin present. A basic personal glucose meter can then be used to determine if there is a high concentration of myoglobin in the sample, simply by adding sucrose.

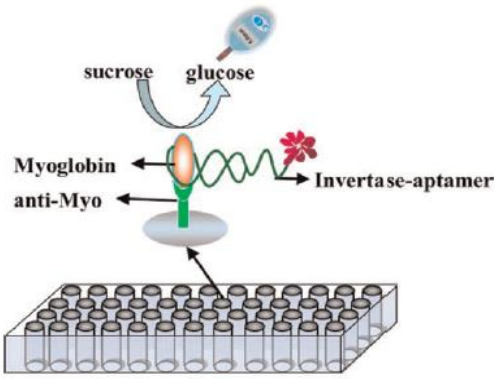

*Figure (adapted)**

-M.E.H.

*Reference: Apta-Index™ ID #602

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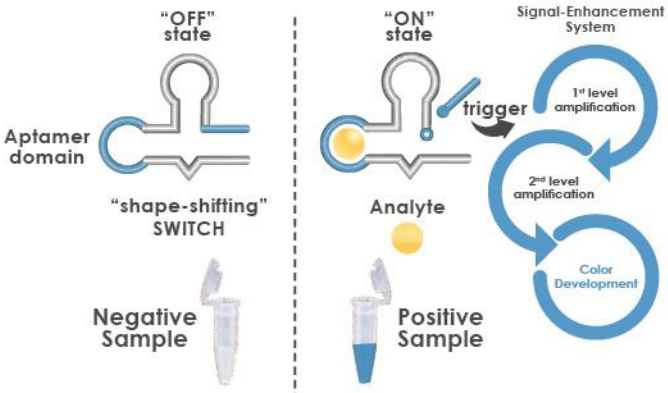

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Technology Comparison Chart

	Antibodies	Aptamers	Apta-Beacons™
Stability/ Refolding		++++	++++
HIGH Affinity	++++	++++	++++
HIGH Selectivity	+	++++	++++
Unknown Biomarkers		++++	++++
Small Target Selectivity	+	++++	++++
Difficult Target Selectivity		++++	++++
One-Step Detection			++++
In-Solution Detection			++++

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