The AptaReport

Newsletter

SPRING 2019

CAIX Aptamer Used to Target Tumor Tissue in Ultrasound Molecular Imaging

In clinics currently, ultrasound contrast agents (UCAs) are used to take ultrasound images of blood vessels in tumors. The images obtained from this procedure are lacking because the contrasting agents do not accumulate in the tumor tissue but instead are equally dispersed throughout every tissue. Targeted UCAs are one method to counteract this problem.

Zhu et al developed a targeted UCA by attaching a carbonic anhydrase IX (a membrane protein that is prevalent in many cancers and allows them to grow) DNA aptamer to the surface of UCAs to specifically target tumor cells. These new nanoparticles allow for the sequestering of UCAs on tumor tissue causing this tissue to produce a higher contrast intensity and producing better images (ultrasound molecular imaging).

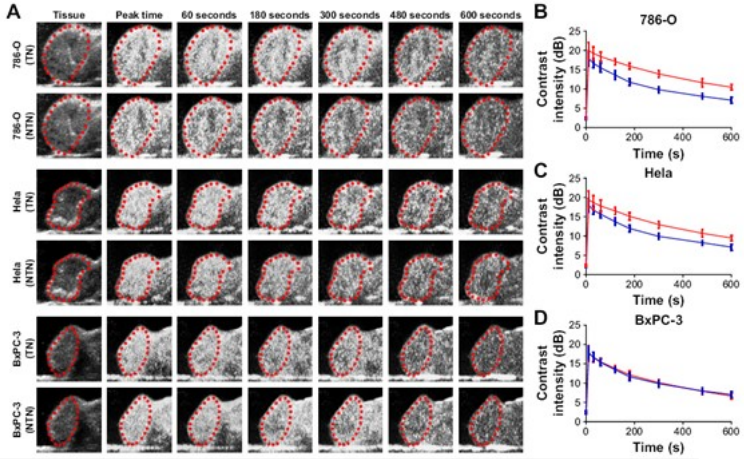

Figure (adapted) Figure: Ultrasound images of targeted and non-targeted nanobubbles in 786-O (positive 1), Hela (positive 2), and BxPC-3 (CAIX-negative) xenograft tumor tissues. Contrast intensities were increased in the positives while the negative control stayed the same.*

-J.S.

*Reference: Apta-Index™ ID #641

Selective Release of Growth Factors to Promote Wound Healing using Aptamers

Growth factors such as TGF-β1 are present in human wounds but they are inactivated by a cage like complex (Large Latent Complex or LLC). During wound healing these cage-like complexes are unfolded through traction forces, releasing the growth factors and stimulating cellular processes to heal the wound.

Stejskalová et al have developed an aptamer complex that mimics the LLC cage allowing for localized application of growth factors that can speed up the healing process for wounds. They call these complexes TrAPs or Traction Force-Activated Payloads because traction forces are what is needed to release the growth factor similar to the LLC in nature. These TrAPs allow for localized release of growth factors that can speed up wound healing. They have also verified the ability to put the TrAPs on materials various materials while keeping their function.

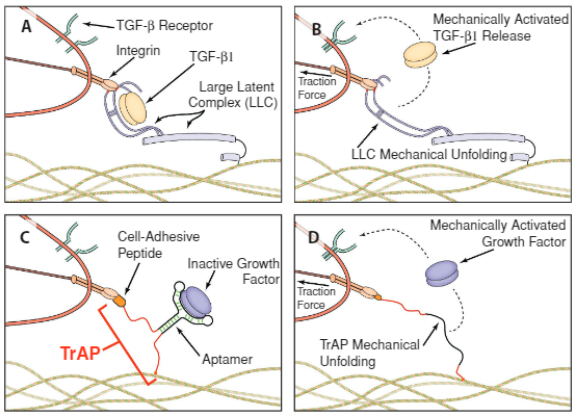

Figure (adopted). Parts A and B show the normal LLC complex being pulled apart releasing the growth factor TGF- β1 during wound healing. Parts C and D show the TrAP complex using an aptamer for TGF- β1 and how it is unfolded in the same fashion.*

-J.S.

*Reference: Apta-Index™ ID #368 & #372

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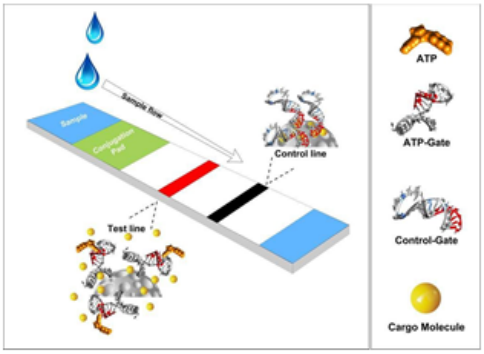
Detecting Small Molecules Using Aptamer Gated Silica Nanoprobes and LFA

Aptamer-based lateral flow assays (LFAs) are an emerging field with numerous applications, however, targeting small molecules remains a challenge. Ozalp et al. has designed a novel LFA using aptamer-gated silica nanoparticles loaded with fluorescein to detect small molecules. The concept is a simple one. When the target molecule is present in the assay solution the aptamer gate molecules will undergo a conformational change which results in the opening of the nanopores releasing the fluorescein dye [1]. Detection can be achieved without modification on the target molecule.,

The aptamer used was modified to have a hairpin conformation when not activated and forms a G-quadruplex when the target is present. The target used was Adenosine-5'-triphosphate (ATP) which is a purine nucleotide present in all living cells. The LFA was designed by immobilizing the ATP aptamer gated nanoparticles on the test line and a mutated aptamer sequence that is non-responsive to ATP on the control line [1]. When samples containing ATP are added to the sample pad the solution will flow down the strip to the test and control lines. The ATP will cause the aptamer gates to open releasing the fluorescein which are washed away by flowing buffer. The release can be monitored by quantifying the decrease in fluorescent signal at the test line [1]. This simple assay can be a powerful tool in detection of small molecules using a lateral flow device.



ATP-Gate	5'-CACCTGGGGGAGTATTGCGGAGGAAGGTTCCAGGTG-SH-3'
Control-Gate	5'-CACCTAGGAGAGTAATGCCGAGGAAGGTTCCAGGTG-SH-3'

Figure (adapted) [1] Özalp, V. C., Çam, D., Hernandez, F. J., Hernandez, L. I., Schäfer, T., & Öktem, H. A. (2016). Small molecule detection by lateral flow strips via aptamer-gated silica nanoprobes. The Analyst, 141(8), 2595–2599. doi:10.1039/c6an00273k*

*Reference: Apta-Index™ ID #642

-L.S.

Fluorogenic Peptide Aptamer used to Track and Measure Individual Metabolites in Algae Cell

The ability to track individual particles and measure their amount in real-time has always been a highly demanded method in molecular and cellular biology research and in the field of biotechnology. There exist drawbacks in current characterization methods with molecular beacons, with one being cell wall protection of bacterial and plants cells, which are both increasingly important in modern biotechnology for producing a lot of useful products such as the antibiotic penicillin and biofuels.

A team in Japan recently developed a laser photoporation delivery strategy for a synthesized peptide aptamer to be used to accurately characterize both spatial and temporal expression of paramylon, a carbohydrate compound, in a microalgal species *Euglena gracilis*. This technique is clearly translational for other cellular organisms and is one of the first to demonstrate both spatially and temporally resolved quantitative characterization of up to single-cell resolution. It could be used to dissect the anatomical kinetics within each living cell and has great value to the biotechnology industry in optimizing various production strategy.

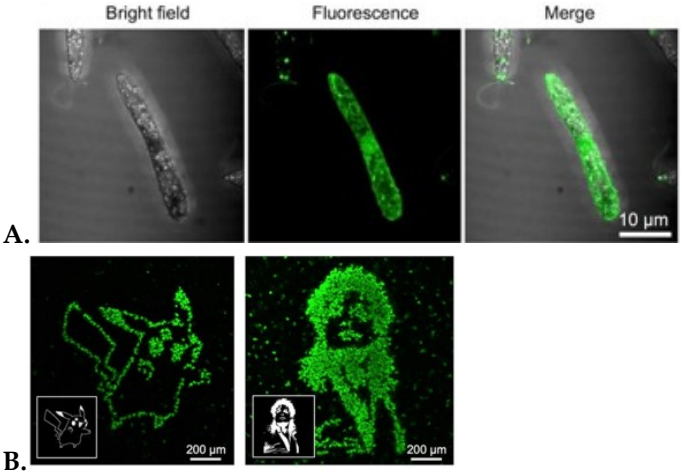


Figure (adapted) (A) Bright-field and fluorescence images of an E. gracilis cell 20 min after the photoporation with the FPBP. (B) Fluorescence images of E. gracilis cells 20 min after the spatially patterned photoporation with the same aptamer. The patterned photoporation was performed on the cells in the black and white patterns of Pikachu (left) and Michael Jackson (right) as shown in the insets. Each fluorescent dot corresponds to a single E. gracilis cell into which the aptamer was injected and bound to intracellular paramylon.*

-M.M.

*Reference: Apta-Index™ ID #643

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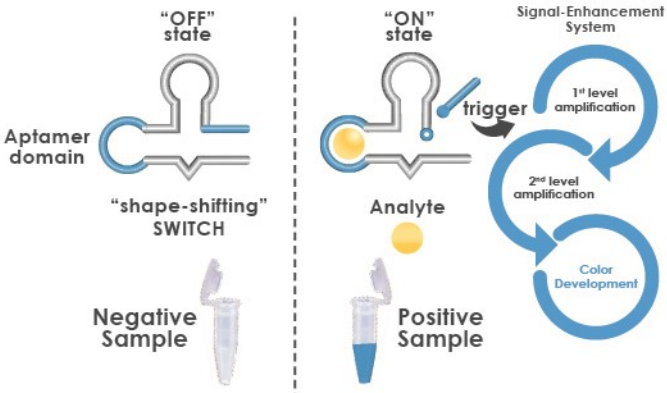

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Technology Comparison Chart

	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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