

Competitive-SELEX Discovery of DNA Aptamers Selective for Neurofilament Light Chain in Human Plasma

Neurofilament light chain (NfL) is a cytoskeletal protein released into cerebrospinal fluid and blood following neuronal injury, making it an important biomarker for neurodegenerative diseases such as Alzheimer's disease, Parkinson's disease, and multiple sclerosis. With Competitive-SELEX, the authors identified DNA aptamers that recognize NfL fragments in plasma.

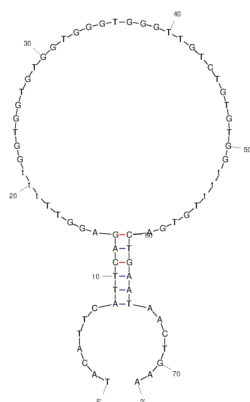


Figure 1. Predicted structure of aptamer MN711. Note that secondary structure may be inaccurate due to changes resulting from interactions with the target.

To improve specificity during later rounds of selection, the researchers introduced a competitive selection strategy using purification tags. Both recombinant NfL and competitor were immobilized separately during selection. Aptamers that bound non-target areas were captured by the competitor, while sequences that specifically recognized true NfL epitopes were recovered. This approach increased epitope-level specificity and reduced enrichment of false-positive binders commonly encountered during recombinant protein SELEX.

Following seven rounds of selection, two lead aptamers, MN711 and MN734, were identified and characterized. Binding affinity and specificity were evaluated using aptamer blotting assays and ELONA, revealing nanomolar dissociation constants comparable to previous antibody-based assays conducted. Both aptamers selectively recognized clinically relevant NfL fragments, showed minimal cross-reactivity with amyloid β 40, amyloid β 42, and phosphorylated tau181. The aptamers demonstrated successful binding to NfL spiked into human plasma, supporting their future use in neurodegenerative disease biosensors and diagnostic platforms.

*Reference: Apta-Index™ ID# 9940

R.S.

Selective Removal of Soluble FLT-1 Using a High Affinity DNA Aptamer for Potential Apheresis Treatment of Preeclampsia

Preeclampsia is a pregnancy-specific disorder characterized by hypertension and proteinuria. A modified DNA aptamer was discovered that specifically binds to soluble fms-like tyrosine kinase-1 (sFLT-1), an antiangiogenic protein.

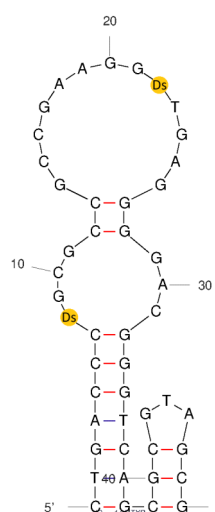


Figure 1. Predicted structure of aptamer TXB-0080. Note that secondary structure may be inaccurate due to changes resulting from interactions with the target.

DNA aptamers were generated using libraries containing the hydrophobic artificial base Ds (7-(2-thienyl)-imidazo[4,5-b]pyridine) to achieve sub-nanomolar binding affinity toward protein targets. The authors utilized a proprietary Xenolingo technology platform based on artificial base pairing to enhance aptamer affinity and specificity.

Binding characterization using surface plasmon resonance demonstrated high specificity, with negligible binding to off-target proteins such as fibrinogen and neuropilins 1 and 2. The aptamer lost some binding activity in human serum. When tested on samples from three patients with preeclampsia, sFLT-1 removal through the apheresis platform ranged from 64% to 94%, demonstrating variable but significant efficacy.

Overall, this study demonstrates the successful development and validation of TXB-0080. This approach represents a promising therapeutic strategy for preeclampsia, complementing existing treatments such as antihypertensive and anticonvulsant therapies.

*Reference: Apta-Index™ ID# 9945

-H.R.

Enriching Higher Affinity Aptamers by Addressing the Kinetic Aspect of the DNA Strand-Displacement Reaction

Ampicillin is one of the most widely used β -lactam antibiotics in clinical medicine and agriculture.

AMP-L2 was identified using a modified capture-SELEX strategy designed to overcome the kinetic limitations of DNA strand-displacement reactions during aptamer enrichment. In this study, the incorporation of an additional 10-minute incubation step dramatically altered the selection outcome, enabling enrichment of higher-affinity aptamers based on equilibrium binding properties rather than kinetic advantage alone.

The aptamer has a molecular weight of 12,731.3 g/mol. Binding studies conducted at room temperature demonstrated a dissociation constant (K_d) of 1.8 μ M, representing a substantial improvement over the lower-affinity AMP-H1 aptamer (K_d = 12.7 μ M).

Mutation analysis revealed that residues A10 and C13 play particularly important roles in ampicillin recognition, while kinetic measurements demonstrated that AMP-L2 dissociates more slowly from the capture strand despite possessing higher equilibrium affinity for ampicillin. This finding established that kinetic bias during SELEX can dominate enrichment outcomes and potentially suppress the recovery of superior aptamers.

In summary, AMP-L2 represents an important advancement in small-molecule aptamer engineering and capture-SELEX methodology. The study provides a rational framework for improving future SELEX strategies by combining low target concentration with extended incubation times to enrich thermodynamically superior aptamers for biosensing, environmental monitoring, and analytical biotechnology applications.

*Reference: [Apta-Index™ ID# 9833](#)

-P.J.K.

DNA Aptamers Mediated Inhibition of Pathogenic Erm42 Enzyme Involved in Antimicrobial Resistance

Erythromycin-resistant methyltransferase 42 (Erm42) is a pathogenic rRNA methyltransferase that contributes to antimicrobial resistance, preventing the binding of macrolide, lincosamide, and streptogramin B (MLSB) antibiotics to the peptide exit tunnel. This allows pathogenic bacteria to evade multiple clinically important antibiotics, making Erm enzymes significant therapeutic targets in efforts to combat the growing global threat of antimicrobial resistance.

Erm42 belongs to the broader Erm family of methyltransferases, including ErmA, ErmB, and ErmC, which confer antibiotic resistance through ribosomal modification. Monomethylation of A2058 results in moderate resistance, while dimethylation produces strong resistance across the MLSB antibiotic class. Highly specific DNA aptamers capable of selectively targeting and inhibiting Erm42 were developed from a randomized 73-mer ssDNA library containing a 40-nucleotide variable region. To improve specificity, the researchers incorporated counter-selection using KsgA, a related rRNA methyltransferase.

Apt-E1 was identified as the strongest aptamer. Gel-monitoring assays and surface plasmon resonance (SPR) analysis demonstrated nanomolar binding affinities, with Apt-E1 exhibiting a K_D of 14.5 nM. Functional inhibition studies using an in vitro methylation assay with radiolabeled 3H-SAM revealed potent inhibition of Erm42 activity, with Apt-E1 producing an IC_{50} value of 12 nM and respectively. DNA aptamers can function as highly specific inhibitors of Erm42, making them antimicrobial resistance therapeutics and diagnostic tools.

*Reference: [Apta-Index™ ID# 9927](#)

-R.S.

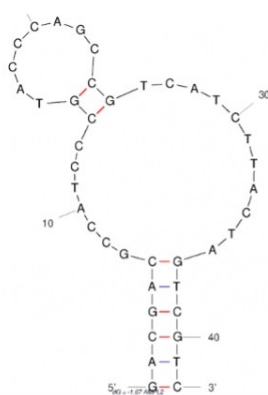


Figure 1. Predicted structure of aptamer AMP-L2. Note that secondary structure may be inaccurate due to changes resulting from interactions with the target.

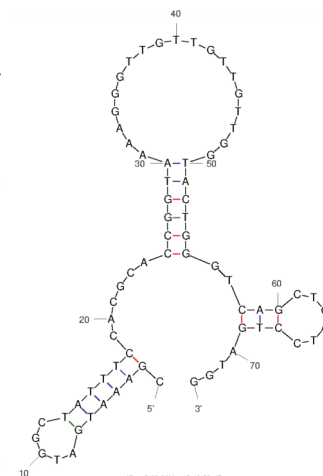


Figure 1. Predicted structure of aptamer Apt-E1. Note that secondary structure may be inaccurate due to other modifications and/or changes resulting from interactions with the target.



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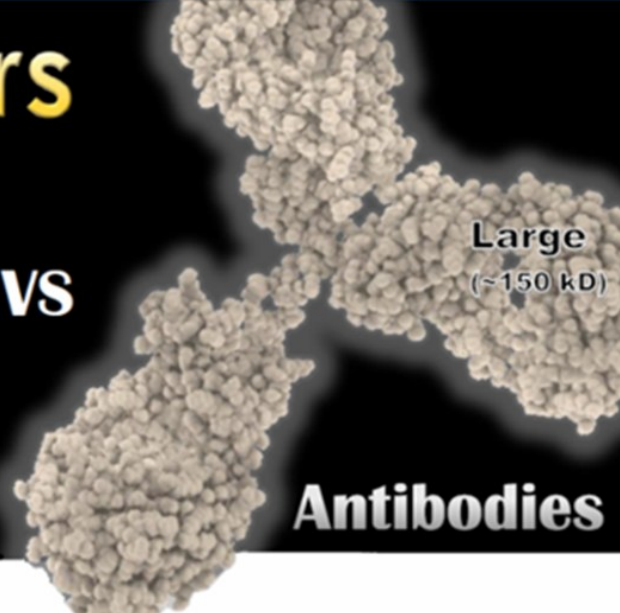
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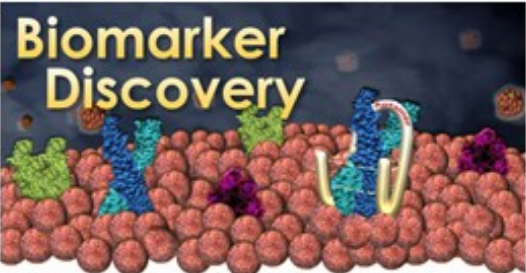


Large
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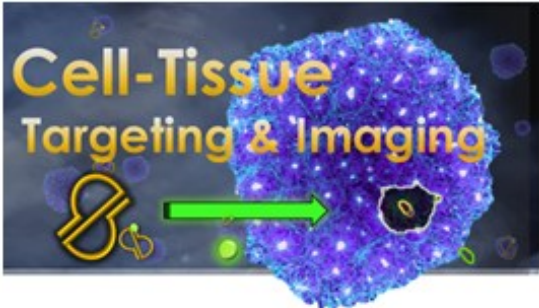
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