SOMAmers: Defining A New Structural Vocabulary With Modified Nucleotide DNA Aptamers In Complex With Protein Targets

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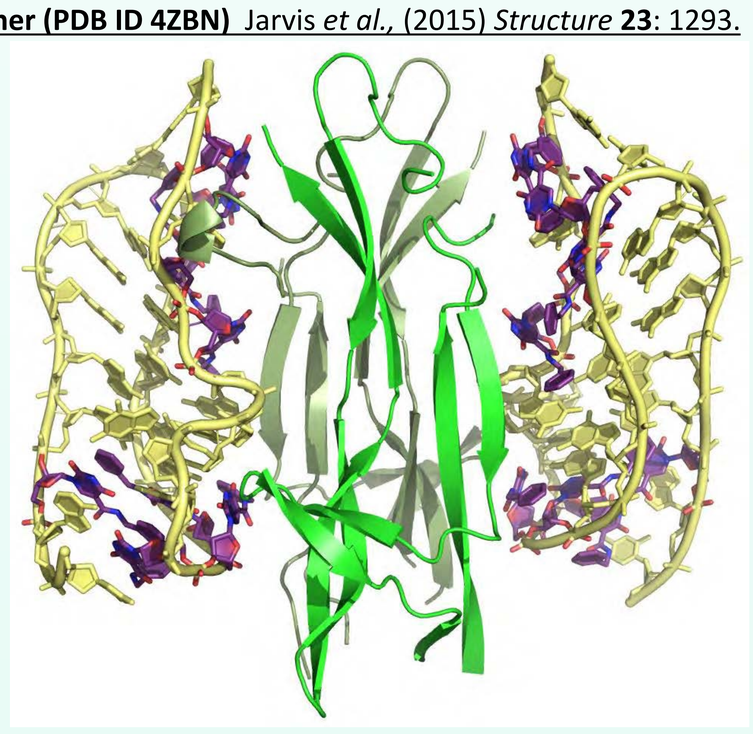
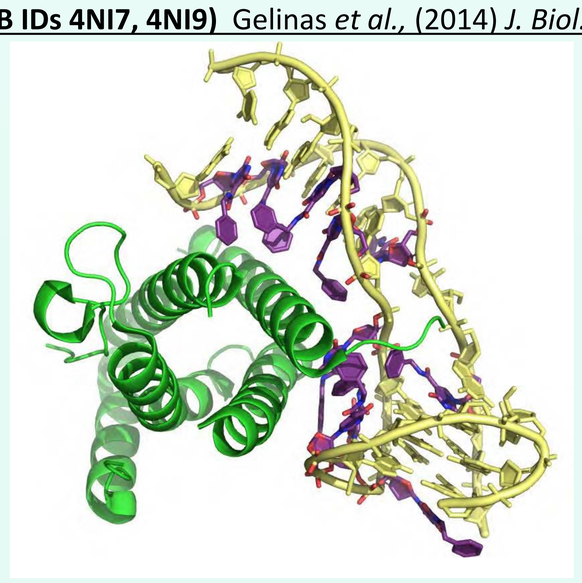
Abstract

Selection of aptamers from nucleic acid libraries by *in vitro* evolution represents a powerful method of identifying high-affinity ligands for a broad range of molecular targets. Nevertheless, a sizeable fraction of proteins remain difficult targets due to inherently limited chemical diversity of nucleic acids. We have exploited synthetic nucleotide modifications that confer protein-like diversity on a nucleic acid scaffold, resulting in a new generation of binding reagents called SOMAmers (Slow Off-rate Modified Aptamers). Here we present a review of X-ray crystal structures of SOMAmers bound to three unique protein targets: PDGF-BB, IL-6 and NGF. In all cases, the SOMAmers fold into compact structures and exhibit hydrophobic binding surfaces that mimic the interface between the protein target and its natural receptor (contrasting sharply with polar interactions seen in traditional protein-binding aptamers). Through incorporation of hydrophobic chemical groups, the modified nucleotides circumvent the intrinsic diversity constraints of natural nucleic acids, thereby greatly expanding the structural vocabulary of nucleic acid ligands.

NGF:SOMAmer (PDB ID 4ZBN) Jarvis *et al.,* (2015) *Structure* **23**: 1293.



IL-6:SOMAmer (PDB IDs 4NI7, 4NI9) Gelinas *et al.,* (2014) *J. Biol. Chem.* **289**: 8720.



PDGF-BB:SOMAmer (PDB IDs 4HQU, 4HQX) Davies *et al.,* (2012) *PNAS* **109**: 19971.

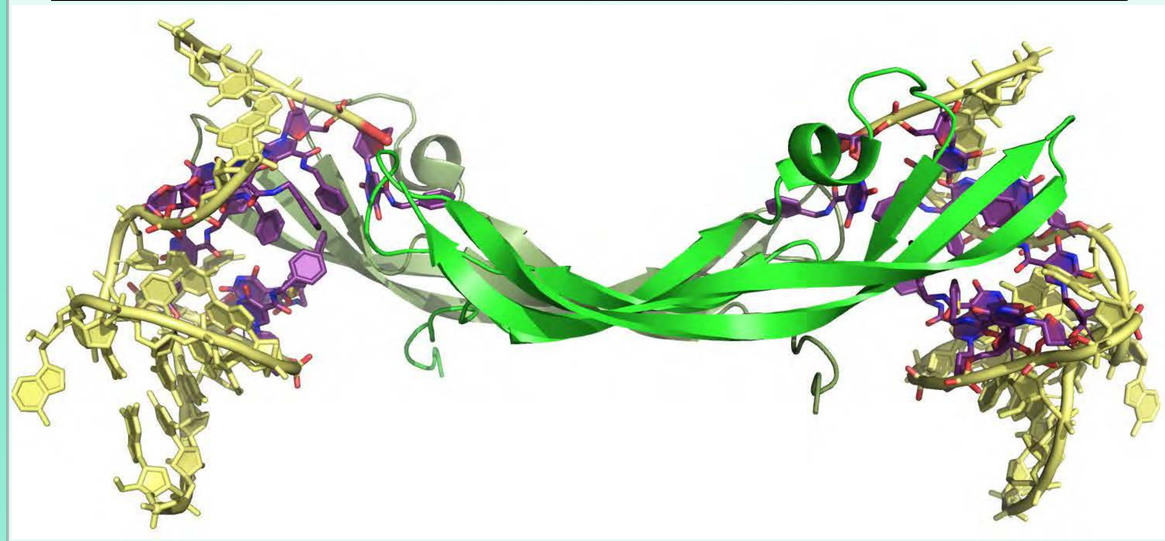


Figure 1: Cartoon Representations of SOMAmer:Protein Complexes Solved by Beryllium for SomaLogic. SOMAmers are represented as yellow stick structures with modified nucleotide residues colored purple. Protein components are displayed as green ribbon diagrams.

SOMAmers: Slow Off-rate Modified Aptamers

SomaLogic has augmented the diversity of randomized nucleic acid libraries by modifying deoxyuridine residues at the 5-position, resulting in improved success rates for SELEX [Gold, *et al.* (2010) *PLoS ONE* **5(12)**:e15004].

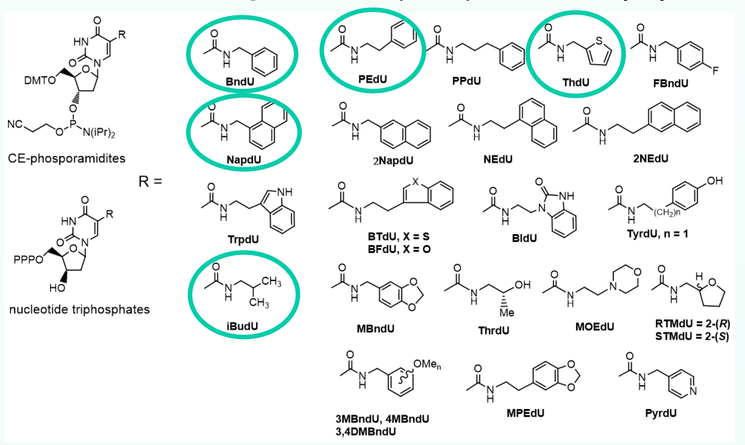
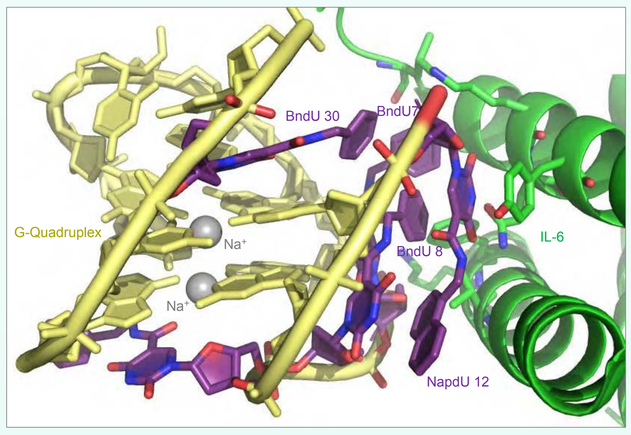


Figure 2: Examples of Modified Residues Available from SomaLogic. Modified residues introduce a "side chain" to the nucleoside base via an amide linker. "Side chain" functional groups mimic amino acid side chains or functional groups of small molecule drugs. Circled modifications have been visualized in X-ray crystal structures.

Unique Structural Features of SOMAmers

Figure 3: Hydrophobic Clusters. Although separated across the primary sequence of each SOMAmer, modified residues cluster in the folded structures, increasing intramolecular stability and presenting a hydrophobic binding surface to the protein target. In the example to the right, a G-quadruplex domain in the SOMAmer is a scaffold for four modified residues that comprise the primary binding interaction with IL-6.



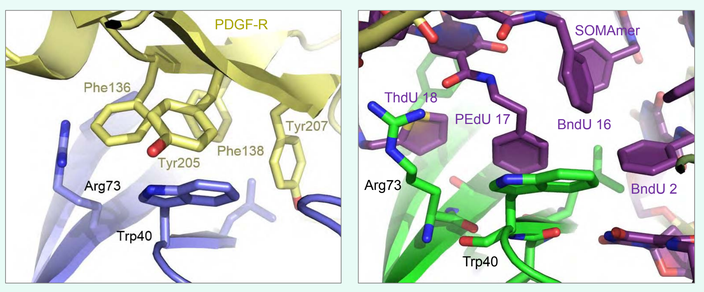
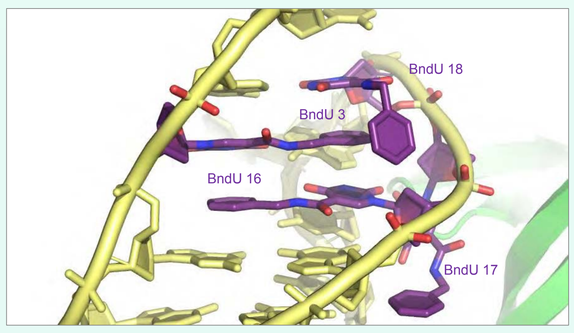


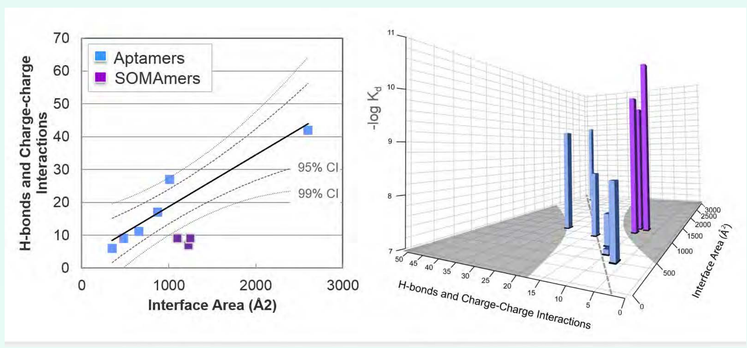
Figure 4: SOMAmer Mimicry of Natural Receptors. In all cases observed to date, the SOMAmer binding site overlaps with the natural protein receptor binding site. In the case of the PDGF-BB SOMAmer, the mimicry extends to the approximate positions in three-dimensional space of aromatic side chain functional groups.

Figure 5: Benzyl-dU "Zipper." In the NGF SOMAmer, two BndU residues mimic two DNA base pairs where each benzyl group exhibits π–π stacking with the nucleoside base of the neighboring BndU residue.



SOMAmers vs. Traditional Aptamers

Figure 6: Plot of the number of polar contacts (hydrogen bonds plus charge–charge interactions) vs. contact surface area vs. reported binding affinities for traditional DNA/RNA aptamer-target complexes (blue bars) and for the three SOMAmers described here (violet bars). **In contrast to traditional aptamers, SOMAmers have higher affinity and primarily utilize hydrophobic contacts.**



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