**DNA Meta-Molecules: Synthetic Biology via DNA Nanostructures & Hybridization Reactions**

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The goal of synthetic biology is to design and assemble synthetic systems that mimic biological systems. Synthesizing synthetic systems for artificial cells forms perhaps the most fundamental challenges in synthetic biology. The impact of developing such synthetic biology systems is to enable a better understanding of the basic processes of natural biology, and also to enable re-engineering and programmability of synthetic versions of biological systems.

*Prior Methods:* One of the key aspects of modern nucleic acid biochemistry is its extensive use of protein enzymes that were originally evolved in cells to manipulate nucleic acids, and then later adapted for laboratory use. This practice provided powerful tools for manipulating nucleic acids, but also limited the extent of the programmability of the available chemistry for manipulating nucleic acids, since it is very difficult to predictively modify the behavior of protein enzymes. Hence methods for synthetic biology based on synthesis of novel proteins enzymes are very difficult.

*Our General Approach of DNA-based Meta-Molecules:* Our approach is to instead synthesize artificial biochemical systems that provide the same functionality of nucleic acids and their enzymes and other proteins. but using a very limited number of types of base molecules (we call these Meta-Molecules) with a very limited chemistry. Meta-Molecules are molecules that are constructed of DNA, but have the properties of natural biological molecules such as proteins and nucleic acids(DNA and RNA). Meta-Molecules act as a type of programmable matter that simulates a number of the most basic and important biochemical reactions that act on DNA. Meta-Molecules have reactions that have an affect similar to protein-based reactions (such as enzyme reactions such as polymerase and restriction enzyme reactions), but are entirely based on DNA hybridization reactions.

*meta-DNA:* We discuss our work to develop a biochemical system termed meta-DNA (abbreviated mDNA), based entirely on strands of DNA as the only component molecule. The work leverages extensive prior work on self-assembled DNA nanostructures. Our mDNA offers the possible advantage of being far easier to re-engineering and program for desired functionality since it entirely DNA-based. Each base of mDNA is a DNA nanostructure. These mDNA bases are paired similar to DNA bases, but have a much larger alphabet of bases, so providing increased power of base addressability. The mDNA bases self-assemble assembled to form flexible linear assemblies (single-stranded mDNA, abbreviated as ssmDNA) analogous to single stranded DNA, and can be hybridized to form stiff helical structures (duplex mDNA, abbreviated as dsmDNA) analogous to double stranded DNA, and also can be denatured back to ssmDNA. We discuss experimentally demonstrations (by Yao Yan's group at ASU) of the self-assembly of ssmDNA and dsmDNA from mDNA bases.

*meta-DNA Reactions using DNA Hybridization:* We also describe on-going work to experimentally demonstrate isothermal hybridization reactions that manipulate mDNA in powerful ways analogous to DNA-DNA reactions and the action of various enzymes on DNA. These mDNA reactions operate without the use of enzymes, are based only on hybridization reactions, and are largely isothermal and autonomous. The reactions already designed to operate on mDNA include (i) hybridization of ssmDNA into dsmDNA and heat denaturation of a dsmDNA into its component ssmDNA (analogous to DNA hybridization and denaturation), (ii) strand displacement of one ssmDNA by another (similar to strand displacement in DNA), (iii) restriction cuts on the backbones of ssmDNA and dsmDNA (similar to the action of restriction enzymes on DNA), (iv) polymerization chain reactions that extend ssmDNA on a template to form a complete dsmDNA (similar to the action of polymerase enzyme on DNA), (v) isothermal denaturation of a dsmDNA into its component ssmDNA (like the activity of helicase enzyme on DNA) and (vi) an isothermal replicator reaction which exponentially amplifies ssmDNA strands (similar to the isothermal PCR reaction).

We discuss have potential applications of mDNA and their reactions for in vitro biochemical systems like transport devices, molecular motors, detection, signaling and computing systems. **Finally, we** discuss extension to structures (termed para-peptides) that allow for side binding of para-peptides, and hence potentially allow for more complex nano-assemblies akin to proteins.