Designer DNA Bases: Probing Molecules and Mechanisms in Biology

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Standard organic fluorophores have been highly useful in biology and medicine, but they exhibit physical properties that can limit their applications. For example, common dyes have relatively short Stokes shifts and each has its unique excitation wavelength; these properties make it difficult to image multiple biological species simultaneously, especially in moving systems, and make instrumentation more complex and expensive than it needs to be.

We have been investigating a new approach for construction of fluorescent labels, by assembling single chromophores into short DNA-like oligomers built on a DNA backbone. The oligomers are termed oligodeoxyfluorosides (ODFs). The close proximity of dyes (Fig .1) results in multiple forms of energy and excitation transfer, yielding complex, emergent emission properties. The repetitive DNA synthesis cycle allows us to combine monomeric fluorophores in thousands of combinations in relatively low-molecular-weight structures (typically tetramers) that can be readily conjugated both to small molecules and large biomolecules. [1-3]

Screening libraries of ODFs has enabled us to identify sets of dyes that can be excited at a single wavelength but emit in colors across the visible spectrum. Such ODFs have large Stokes shifts and high quantum yields, and can be used in real-time imaging of moving biological systems. ODFs can be cell permeable on their own, or they can be delivered intracellularly with DNA uptake reagents such as cationic lipids. We have further developed methods for conjugating them to antibodies, and using genetic encoding methods, to other proteins of interest in living cells. [2,3]

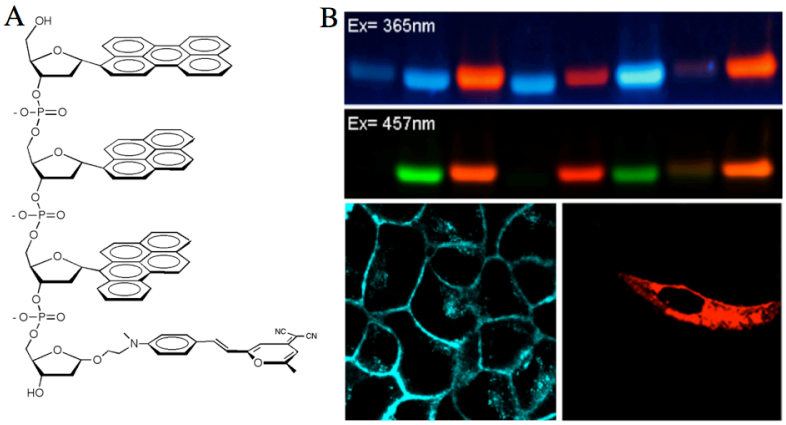

Figure 1. (A) Example of DNA-polyfluorophore. (B) Genetically encoded multispectral protein labeling in vitro and in live cells.

ODF dyes can show static fluorescence properties, or they can change in response to molecular stimuli. Fluorescent DNA bases can be combined with natural DNA bases or with designer fluorescent bases that have additional properties such as affinity for metals. Screening libraries of such ODFs has allowed us to identify sequences that respond with changes in fluorescence to organic and inorganic species in the air and in water. For example, by optical monitoring of fluorescence changes, we have used ODFs to identify different strains of bacteria by the vapors they emit as they grow. Moreover, we have used metal-binding ODFs to distinguish and quantify toxic metals in solution. In a third example, combining fluorophores with damaged DNA bases has also allowed us to design cellular reporters of enzymes that repair DNA damage. [4-6]

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