The Development of DNA-Templated Organic Synthesis

Of primary importance to the organic chemist is the synthesis and discovery of functional molecules. While tremendous strides have been made towards this end through improvements in synthetic methodology, high-throughput screening, and rational design, the general philosophy governing the application of these technologies to chemical synthesis and discovery has remained largely unchanged. Recognizing certain basic differences between synthetic and cellular means of assembling and optimizing molecular structures, my co-workers and I turned to the cell for inspiration for a fundamentally new and general method for both the synthesis and discovery of functional organic molecules.

In the realm of synthesis, organic chemists wield a powerful arsenal of chemical transformations, the utility of which has been demonstrated through the synthesis of a dizzying array of natural products, drugs and other functional molecules. For the most part, however, these transformations are applied in a single step to all molecules in a reaction flask. In addition, these transformations rely upon kinetic differences between various reaction pathways for specificity, and require the use of toxic and or specialty solvents to aid in the dissolution of the high concentrations of material necessary to drive reactions to completion. These three points, judging by their near universal application, have at the very least hindered our ability as chemists to approach problems from new perspectives.

A typical cell assembles the preponderance of its molecules using a fundamentally different philosophy and a considerably smaller chemical arsenal. Yet, these limited numbers of transformations are sufficient to generate the incredible functional diversity of molecules observed in nature. Unlike the synthetic chemist, cells assemble many different molecules with similar reactivities, at the same time, in a small number of enveloped compartments. This is achieved by maintaining the molecular pool at low concentrations, driving reactions only between desired molecules by localizing them to common macromolecular templates. Such co-localization increases their effective molarity, and allows a chemical reaction to proceed. Incredibly, a cell conducts transformations in this manner on an array of substrates with wildly varying solubilities using a single universal solvent: water.

From the perspective of an organic chemist, a cell provides an inspiring lesson in the control of molecular reactivity. Cells, however, have another remarkable property from which chemists interested in discovering functional molecules can take a cue – the ability to evolve. Evidence for the power of molecular evolution abounds, and the basic process has been recapitulated in vitro and applied to biological macromolecules with great success. Strictly speaking, evolution is the iterative application of four manipulations: diversification, synthesis, selection and amplification. In order to apply these four steps, a method must exist for linking the chemical structures from which functional molecules will be selected to an amplifiable information carrier (such as DNA or RNA) that can be both amplified and mutated. In addition, a method must exist for translating the genetic information stored within DNA into the chemical structures to be evolved. Cells accomplish this feat by maintaining both DNA and its downstream products within the same vesicle. Information is translated by an array of polymerases, the ribosome, and the various protein catalysts which synthesize and maintain the cellular
metabolome. However, because chemists lack the machinery to translate DNA into arbitrary chemical structures, the evolution of wholly synthetic molecules has thus far proven elusive.

My thesis first describes the in vitro adaptation of the cell’s method of controlling chemical reactivity to many powerful and general synthetic transformations developed by chemists. The method relies upon the rapid kinetics of annealing of complementary DNA strands under dilute conditions to bring together covalently tethered reactants to a common site, increasing their effective molarity, and allowing them to undergo a chemical reaction. The method, referred to as “DNA-Templated Synthesis” (DTS), has proven surprisingly general. My results and those of my coworkers illustrate a variety of new approaches to synthesis and discovery enabled by DTS. These include the parallel assembly of molecules using traditionally incompatible transformations together in a single flask, novel purification strategies which are independent of the properties of the synthesized molecules, multi-step syntheses, novel DNA architectures to fine-tune reactivity of specific transformations, multi-component reactions, and synthetic libraries of functionally rich macrocycles. Aspects of the method have subsequently been elaborated on by others to discover powerful transition metal-mediated cross-coupling reactions that are impossible or difficult to perform in a traditional flask-based synthetic format. These results suggest that an array of chemical reactivity awaits discovery which has previously been obscured by problems inherent to solution or solid phase synthesis.

In addition to presenting a fundamentally new approach to synthesis, my thesis describes the application of DNA-Templated Synthesis to the evolution and discovery of functional synthetic molecules. By the nature of DTS, the products of DNA-templated reactions are synthetic organic moieties linked to amplifiable information carriers that both encode and direct their own synthesis. As a result, these molecules are amenable to in vitro selections as well as the other three manipulations integral to the evolutionary process (selection, amplification, and diversification). My coworkers and I show that starting with a diverse pool of DNA templates functionalized at one end with an unnatural amino acid, we can synthesize a structurally and functionally diverse collection of macrocyclic peptide fumaramides, select from them a single molecule exhibiting protein binding properties, and amplify the resulting pool of DNA-linked small molecules to reveal the DNA “recipe” for the synthesis of the active molecule.

Taken together, my thesis presents a fundamentally new approach to the synthesis and discovery of functional molecules inspired by the synthetic and evolutionary machinery found in every living cell. In addition to the advances outlined above, DTS may also provide the foundation for a more facile ‘bottom-up’ approach to the synthesis of supra-molecular compounds, molecular electronics, and nanostructures containing true organic, covalent bonds. It should be feasible to construct these structures in a more carefully controlled manner than has been previously possible using non-covalent interactions that are neither as easily engineered, nor as precise as those provided by DNA.
Publications:


