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EDITORIAL

Chemical Synthesis and the Synthetic Cell

Analysis and synthesis are terms that have a very special meaning in chemistry. The study of matter has always required analytical tools of increasing sophistication. Over the last two centuries the successes of physics and chemistry have provided remarkable insights into the nature of matter, living and non-living. The distinctions between biology and life on the one hand and chemistry and inanimate matter on the other were first blurred in the 1820s, when Friedrich Wohler famously demonstrated the production of urea, a molecule long associated with living organisms, by heating ammonium cyanate, a common chemical that belongs undoubtedly in the realm of inorganic chemistry. This was indeed the decisive step in demonstrating the unity of matter and chemistry. Vitalism, which endowed living systems with a shadowy 'vital force', was dealt a death blow when Eduard Buchner demonstrated fermentation by the 'juices of cells', in the later years of the 19th century. Biology clearly was controlled by chemistry. In the 20th century, both chemical synthesis and chemical analysis have made astounding progress. Synthesis, especially organic chemical synthesis, has focused on strategies to build, often painfully and laboriously, molecules that are produced with remarkable ease and facility by living organisms. The 'total synthesis' of natural products is an area where the experimental campaigns have been long and hard. The targets when achieved, have permitted the development of closely related 'non-natural' structures, that have often proved immensely valuable as therapeutics. Nature's chemical virtuosity has endowed 'natural products' with a degree of structural variability that never ceases to amaze the connoisseurs of molecular structures. Organic synthesis has for long been pre-occupied with targets, which display a degree of structural intricacy, that provide an intellectual challenge to the practitioner. Stitching groups of atoms together, while exercising total control over their three-dimensional relationships, is not a task for the faint hearted. Imagination, experimental skills, patience and perseverance are key ingredients in the long and sometimes tedious campaigns of total synthesis. The field of organic synthesis, like most other fields of science, is changing with the times. Terms like 'atom economy' and 'green chemistry' may soon dominate the field. A recent essay highlighting the challenges of the field argues that, 'potentially the biggest challenge for twenty first-century synthetic chemistry' will not just be 'preparing a given target molecule, but doing so with a level of efficiency and cost-effectiveness that rivals that of nature itself' (Snyder, S. A., *Nature*, 2010, **465**, 560).

Traditional 'natural products chemistry' has always limited its attention to the diverse 'secondary metabolites' produced by living organisms, most notably microbes and plants. Nature produces these structurally diverse molecules ('biosynthesis') using groups of enzymes that catalyse chemical reactions that are often hard to mimic in the laboratory. Nature's chemistry is truly 'green', displaying a degree of efficiency that can only be envied. The enzymes (invariably proteins) are in turn produced with amazing efficiency as products of a 'DNA blueprint' that is present in the organism's genome. Sophisticated control mechanisms can turn these chemical pathways on and off. Organisms thus develop a sophisticated chemical response to environmental stimuli. Curiously, while secondary metabolites have provided synthetic chemists with many peaks to conquer, the products of primary metabolism have always been of interest only on the fringes of mainstream organic chemistry.

The central actors of biological chemistry, proteins and nucleic acids are polymers in which a limited number of structural units (monomers) are strung together into long monotonous chains. The size and repetitive nature of the chemical steps needed to assemble these polymers has always appeared to pose a technical challenge of formidable proportions. The chemistry of proteins and nucleic acids has its origins in the work of Emil Fischer in the final years of the 19th century and the early years of the 20th century. The most dramatic progress occurred in the period between the 1950s and 1970s when the methodologies for nucleic acid synthesis, DNA in particular, were established by Hargobind Khorana and the principles of 'solid phase synthesis' were developed by Bruce Merrifield. The ability to synthesize, stepwise, long chains of proteins and nucleic acids has been facilitated immeasurably by the technique of holding the growing polymer chain on a solid, insoluble polymer bead, while washing off reagents and reactants in step after step. The repetitiveness of the steps involved lends itself to automation. Automated 'synthesizers' now adorn laboratories;

sleek replacements for trained and tired chemists. Researchers today can order synthetic nucleic acids and proteins over the Internet; the credit card and a research grant being the sole requirements for acquiring material, that was completely unavailable not too long ago. Molecular biology has been revolutionized by the ability to analyse (sequence) genomes with amazing speed. Synthetic genes can also be assembled at relatively low cost and high speed. The abilities to cut, paste, modify and copy DNA have been central to the advance of biotechnology. The announcement of the 'Creation of a bacterial cell controlled by a chemically synthesized genome' seemed almost inevitable when it was first reported a couple of weeks ago (Gibson, D. G. *et al.*, *Science*, doi: 10.1126/science.1190719;2010). A team of authors led by Craig Venter describe the 'design, synthesis and assembly of the DNA corresponding to the genome of the organism *Mycoplasma mycoides*'. The task of putting together 1.08 million bases of the synthetic genome was accomplished by using 1078 'DNA cassettes' each having a length of 1080 base pairs. The synthetic sequence contains 'watermark' sequences which permit distinction from the natural genome. Laboratory 'forgeries' can be set apart from Nature's 'originals' using these cleverly placed identification marks. Venter and his colleagues successfully transplant the synthetic genome into a faster growing organism, *Mycoplasma capricolum*, in their quest to establish the ability of a cell to grow and divide under the control of a chemically synthesized genome. Specialists will undoubtedly dissect the strategies they employ enroute to their goal; but it is clear that this report constitutes a technical *tour de force*, which demonstrates the power of analysis and synthesis in contemporary molecular biology.

The paper by Venter and coworkers is yet another landmark in the triumphant advance of molecular biology. The authors note that 'our ability to rapidly digitize genomic information has increased by more than eight orders of magnitude over the past 25 years'. Their rationale for creating the chemically synthesized genome is simply stated: 'No single cellular system has all of its genes understood in terms of their biological roles. Even in simple bacterial cells, do the chromosomes contain the entire genetic repertoire? If so, can a complete genetic system be reproduced by chemical synthesis starting with only the digitized DNA sequence contained in a computer?' Have Venter and his colleagues successfully synthesized life, as represented by mycoplasma, from the bare chemical components of DNA? The authors note that they 'refer to . . . a cell controlled by a genome from chemically synthesized pieces as a "synthetic cell", even though the cytoplasm of the recipient cell is not synthetic'. They further argue that the 'phenotypic effects of the recipient cytoplasm are diluted with protein turnover and as cells carrying only the transplanted genome replicate'. They also add: 'The properties of the cells controlled by the assembled genome are expected to be the same as if the whole cell had been produced synthetically (the DNA software builds its own hardware)'.

The Venter paper has reopened an old question: 'Can life be synthesized?' The numerous commentaries that have begun to appear after the recent paper in *Science* acknowledge with admiration a major technical achievement, but are quick to dispel the notion that the 'synthesis of life' has been achieved in the laboratory starting from chemical constituents of cells. The opinions of 'eight synthetic biology experts' on the 'synthetic cell' that appear in a recent issue of *Nature* (2010, **465**, 422) make interesting reading. Steven Benner presents a provocative view: 'Synthesis is not a field. Rather, it is a research strategy that can be applied to any field in which technology allows scientists to design new subject matter. Such technology has long been available to chemistry where it allowed theory to develop faster than in fields lacking synthesis, such as planetary science and biology'. Benner notes that the real challenges of synthesis lie not in tinkering with 'natural biological parts' but in new design. He adds: 'Should . . . design strategies be flawed they will fail in ways that cannot be ignored. Thus synthesis drives discovery and technological innovation in ways that observation and analysis cannot' (p. 423). Jim Collins provides the dampener: 'Frankly, scientists do not know enough about biology to create life. Although the Human Genome Project has expanded the parts list for cells, there is no instruction manual for putting them together to produce a living cell. It is like trying to assemble an operational jumbo jet from its parts list – impossible' (p. 424). The 'synthetic cell' has reopened an old question: 'What is life', famously asked by Erwin Schrödinger in a book that drew a generation of physicists to biology. The answer has never been very clear and the debate on the synthetic cell is unlikely to yield a satisfactory resolution.

The technical triumph reported by Venter and coworkers opens up the field of engineered genomes, with many potential applications in biotechnology. Genome analysis and synthesis will undoubtedly be central in the attempt to create engineered organisms capable of performing designed chemistry. Synthetic life may still remain a distant dream. Many years ago, when one of synthetic chemistry's iconic figures, R. B. Woodward, received the Nobel Prize (1965) for his contributions to the 'art of organic synthesis' he is reported to have been asked: 'Will you next synthesize life'. His response (possibly apocryphal) will please many: 'I am quite happy with the way it is presently done.'

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