Darwin in Vitro

The quest to make synthetic self-replicating molecules

By RICHARD LIPKIN

Consider, for a moment, what it would take for a chemical system — any chemical system — to evolve to the point where its chief informational molecules (such as RNA and DNA) could copy themselves, spawn an army of little replicators, and spark a runaway reaction that populated an entire planet.

The improbability of such an event is unfathomably high. And yet, at least once, it happened. This type of chain reaction led to life on Earth.

Given the appearance of terrestrial life, it seems only natural to wonder what mechanisms, or principles of nature, enabled this phenomenon to occur. Or, as Julius Rebek Jr., a chemist at the Massachusetts Institute of Technology, puts it: "What are the minimum features necessary for a chemical system to self-replicate?"

As part of a small coterie of chemists whose struggle with this question dates back nearly half a century, Rebek — in contrast to most of his colleagues — has looked at the problem from a nonbiological point of view. Rather than picking apart the molecules of living systems and experimenting with what may have been the chemical precursors of Earth's primordial soup, he is building self-replicating molecules from scratch, by rational design and synthesis.

"In a very distant sense, I am thinking about the origin of life," he says. "But none of the molecules I work on were in the prebiotic soup. These molecules were carefully contrived to help us understand the basic rules of replication."

Rebek and his coworkers seek nothing less than the fundamental causes that prompt some chemical systems to make copies of themselves. In essence, these scientists highlight the fact that life based on replicating nucleic acids such as RNA and DNA is only one example — albeit a powerful one — of a general chemical phenomenon. His goal is to figure out the underlying "rules" behind chemical self-replication.

Rebek contends that by synthesizing simple, self-replicating molecules that behave according to a primitive chemical logic, he can gain insights into the overarching principles. Indeed, to achieve his objective, he must decode the essential physical chemistry behind molecular self-recognition and replication.

"People have been after this problem ever since the structure of DNA was revealed in 1953. Just by looking at that structure you can see how DNA can replicate," Rebek says. "But what I want to show is that you don't need such a complicated molecule to achieve self-replication. You can do the same thing with simple, synthetic molecules. I believe that if you can get molecules to recognize each other chemically, then you can build a replication cycle."

Four years ago, Rebek and his coworkers successfully produced a primitive self-replicating molecular system (SN: 2/3/90, p.69). Though the system turned out to be slower and less efficient than they had hoped, the molecules did copy themselves by way of complementarity. In other words, each molecule served as a template for an identical copy of itself. As a steady cycle of autocatalysis took hold, each template molecule attracted smaller molecular pieces from solution and fit them neatly into place.

Unlike life, which uses water as the basis of its chemistry, Rebek performs his experiments in organic solvents. This decision, he says, arose from the fact that such solvents speed reactions and allow the general principles of replication to be observed more readily.

Recently, Rebek and his colleagues reported on a second generation of self-replicators in the Oct. 5 JOURNAL OF THE AMERICAN CHEMICAL SOCIETY.

"We've improved the structure by tweaking some fine points," says M. Morgan Conn, a chemist at the University of California, Berkeley, who helped in the new design. The first generation of molecules showed that, for a template to reproduce itself, it must hold molecular pieces firmly enough in place so that they can bind together without wiggling around. "Position is very important," adds Conn. "So we tightened up the molecule's receptors," where self-assembly takes place.

Another problem arose in the first set of experiments because of "alternate reaction pathways," says MIT chemist Edward A. Wintner, who aided in the redesign. Rather than follow the desired pathway of chemical steps, the molecules tended to wander off into nonproductive reactions that did not produce replication. Rebek's group solved that problem in its more recent work by building in safeguards that effectively block those extraneous reactions. In their current incarnation, the self-replicating molecules have fewer available choices. They will either replicate or do nothing.

Another advantage of the new replicators is their more "general-purpose scaffolds," the core skeletons onto which functional chemical groups can later be attached, Conn says. "This will give us more flexibility for modifying the [molecule's] structure in the future."

Though Rebek is not the first person to create an artificial system of replicating molecules, he has pushed furthest in the direction of making explicitly nonbiological self-replicators. Indeed, most previous efforts came out of molecular biology, primarily from chemists studying the chemical precursors of RNA and DNA.

"There are two distinct approaches to this game," says Leslie Orgel, a chemist at the Salk Institute in La Jolla, Calif. "One is to start with synthetic organic chemistry and the other is to start with molecular biology. Each approach has its advantages. Molecular biologists have an edge right now. And yet what they achieve will be more limited, in the sense that they will be repeating what nature does. If the organic chemists succeed, they will be doing something quite different from what nature does."

In 1986, Gunther von Kiedrowski, a chemist then at the Salk Institute and now at the University of Freiburg in Germany, created the first artificial self-replicators, derived from models of RNA and DNA. Shortly thereafter, Orgel and his colleagues devised an alternate scheme, also rooted in the chemistry of the two nucleic acids. Subsequently, Gerald F. Joyce, a chemist at the Scripps Research Institute in La Jolla, and Jack Szostak of Harvard University have each independently come up with biologically based self-replicating systems.

Rebek, though, has moved increasingly in the opposite direction.

"What makes Rebek's work so interesting is precisely his generalized approach to replication," Joyce says. "He's saying that self-replicating molecules don't have to be nucleic acids. They can be whatever will replicate in a complementary fashion. He's not getting too parochial about specific molecules and so his work is very imaginative."

While some researchers want primarily to know how life on Earth began, leading them to water-based replication, says Joyce, others want to know how genes evolved, prompting them to study nucleic acids. "But I think there's great value in opening up one's mind to the general problem of replication," Joyce adds. "On the practical side, by abandoning the historical constraints of life on Earth and just trying to get molecules to replicate, Rebek may find more interesting pathways to replication."

Rebek remains somewhat bothered by the fact that many molecular biologists see no point in deviating from nature. "They say we already have living systems that replicate," he notes, "so why bother with model systems?"

"With DNA and RNA, we can feel our way back to the origins of life by looking down the narrow groove of DNA into the past," he points out. "Maybe that approach will work. But maybe it won't. What if the road's washed out, so to speak, after billions of years of evolution? It's possible that RNA is an advanced molecule that came later, after earlier self-replicating forms."

"As a chemist, I think the world of possibilities for self-replicating molecules is much larger than just RNA and DNA," he observes. "It may be possible, for example, to make self-replicating molecules out of many types of molecules."

While approaching molecular self-replication with the origin of terrestrial life in mind, Rebek's overall goal is to abstract the underlying principles and then test them.

What would it take to develop a living system — any living system?

First, Rebek notes, one needs either a molecule that "lives" forever or one that makes copies of itself. Second, that molecule must be able to recognize and distinguish itself from its surroundings, a requirement that calls for a boundary or capsule — such as a cell membrane — to "keep good things in and bad things out," he says.

The replicators also must be able to harness energy. "In our experiments, we have to add material to keep the products growing. But we'd like to have a way to take sunlight, for example, and use it to generate pieces to be incorporated into replication. This would be a form of artificial metabolism. If we can put all of these elements together, then maybe we can cross the boundary from chemistry to biology."

Taken to its extreme, an explanation of the general principles of self-replication — freed of the constraints of nucleic acids — might give some clues about the possibilities for and limitations of extraterrestrial life.

Using evolution on Earth as a model, Rebek wants eventually to incorporate Darwinian competition and natural selection into his artificial system as a way of selecting the best self-replicators from among various types.

"Clearly he's gotten self-replication to go," Joyce says. "But Darwinian evolution is what's needed next. If many different molecules replicate at the same time, all competing for limited resources, then the most 'fit' molecule will be the one that replicates the fastest. That's what we'd all like to see next: Darwinian behavior in an artificial replicating system.

"Whether this is easier to achieve by liberating oneself from nucleic acids is unclear," observes Joyce. "But it's certainly worth exploring both paths."

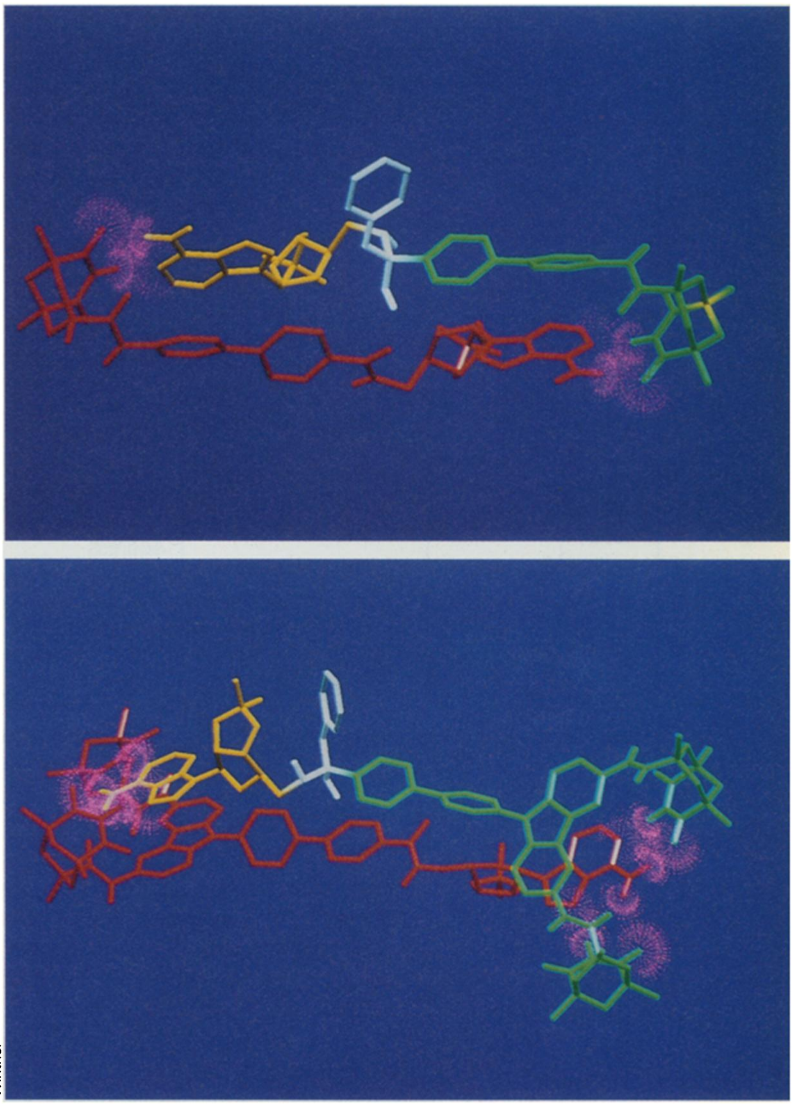
Andrew D. Ellington, a chemist at Indiana University in Bloomington, agrees.

"We tend to regard DNA and RNA as special because they are the materials of life. But, really, they're not that special. They just exhibit complementarity."

"In the primordial soup, I suspect that the best replicators probably won out, as a result of competition and natural selection. And those winning sequences eventually became the building blocks of life. But these concepts are somewhat foreign to [most] synthetic chemists. They tend to think in terms of a different paradigm than biologists do."

Ellington believes that bringing biological concepts — such as population diversity, competition, and natural selection — to bear on synthetic chemical systems will accelerate the process of generating viable self-replicating molecules. "What if you need competition between different molecules to make a good self-replicator?" he wonders. "What if the results emerge not from a single species of molecule but from a population?"

"When the viewpoints of biology and synthetic chemistry eventually merge," Ellington adds, "I have no doubt that it will produce something astounding." □



Wintner

Self-replicating molecules: In the first generation (above), a template molecule (red) copies itself by holding an amine (yellow) and an ester (green) in place with four hydrogen bonds (purple), two on each end. This allows a covalent bond (white) to form, fusing them. In the second generation (below), the ester (green) is branched; it has eight hydrogen bonds. The new configuration holds molecular components more firmly in place.