Exploring the Systems of Life

No longer content to inventory cells' molecular parts, biologists are teaming up with physicists and engineers to study how these parts work together

A rule of thumb among drugmakers is that the more tightly a compound binds to its molecular target, the more potent it will be. But not always, it turns out. Take cytokines, natural protein messengers that bind to receptors on cells and cause them to proliferate during wound healing or an immune response. A cytokine molecule follows a complex life history before and after it binds to its receptor. It shuttles in and out of cells, risking destruction by proteases, and eventually finds its way into a recycling bin once its work is done. These steps interact, adding to the complexity. When proteases destroy a cytokine molecule, for example, they can also wipe out its receptor, in a feedback that further reduces the compound's effectiveness.

By modeling these and other interactions on a computer, Douglas Lauffenburger and his colleagues at the Massachusetts Institute of Technology have found that in many cases the best way for genetic engineers to boost the potency of a cytokine drug is not by remodeling it to bind more tightly to its receptor but by altering other steps in the chain. Tweaking the structure to help it avoid destruction within the cell, for example, increases its chances of being recycled. "You would think that the stronger the binding, the more potent it would be," says Lauffenburger. "But that's often not the case."

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—Lucy Shapiro

As he and his colleagues have realized, understanding how parts of a biological system—genes or molecules—interact is just as important as understanding the parts themselves. It's a realization that's beginning to spread. Leading research universities around the United States have begun shelling out tens of millions of dollars to set up new interdisciplinary institutes and departments that will bring together specialists from physics, chemistry, engineering, computer science, mathematics, and biology to document how all the different cellular players work together in complex tasks such as determining when a cell divides and how gene expression is regulated. Says Lucy Shapiro, a developmental biologist at Stanford University: "The convergence of chemistry, physics, biology, and engineering is upon us."

The new centers will take a variety of approaches to exploring the complex systems of life. A proposed center at Stanford, for example, is likely to focus on biophysics, while one at Princeton will lean toward probing networks of genes and proteins. Drug companies, too, such as the Palo Alto, California–based startup Entelos, are turning to computers in the hope that "in silico" biology will lead to improved therapeutics. All these efforts are a response to the growing sense that gene sequencing and other techniques will soon have isolated all the cell's individual parts and

Building Working Cells 'in Silico'

Cells provide living proof of that old saw about the whole being greater than the sum of its parts. "Even if you construct a complete list of all the processes known to occur within a cell, that won't tell you how it works," says Masaru Tomita, a professor of bioinformatics at Keio University in Fujisawa, near Tokyo. But Tomita, who is a computer scientist as well as a biologist, has a scheme for exploring the effects that only emerge when those many processes interact: a simulation program that can reproduce, in simplified form, a cell's biochemical symphony.

His group's E-CELL simulation software will go on the Web for public "beta" testing this June (www.e-cell.org). Other computer models of the cell are being developed, but they often try to reproduce individual cellular processes in detail. E-CELL, in contrast, is designed to paint a broad-brush picture of the cell as a whole. Such efforts "are a next logical step" now that genome sequencing is giving biologists the complete parts lists for living things, says Peter D. Karp, a bioinformaticist at Pangea Systems, a bioinformatics software company in Menlo Park, California.

E-CELL is actually a model-building kit: a set of software tools that allows a user to specify a cell's genes, proteins, and other molecules, describe their individual interactions, and then compute how they work together as a system. It should ultimately allow investigators to conduct experiments "in silico," offering a cheap, fast way to screen drug candidates, study the effects of mutations or toxins, or simply probe the networks that govern cell behavior.

Written to run under the UNIX or Linux operating systems, the software relies on the user to input a cell's molecules, their locations and estimated concentrations within the cell, and the reaction rules that govern them. E-CELL then computes how the abundance of each substance at a particular location changes at each time increment. With a single mouse click, the user can knock out particular genes or groups of related genes, expose the cell to a foreign substance or deprive it of a nutrient, and then run the simulation again. Graphical interfaces allow the user to monitor the cell's changing chemistry.

Tomita's group has used early versions of E-CELL to construct a hypothetical cell with 127 genes, which they figured was a minimal set for a self-sustaining cell in their system. Most of the genes were based on those of *Mycoplasma genitalium*, a microbe that has the smallest known gene set of any self-replicating organism. But the genes for some vital cellular processes still have not been identified in the mycoplasma, so the group added genes from other organisms. The virtual cell "lives," maintaining a simple, stable metabolism: It takes up glucose from the virtual culture medium, generates the enzymes and proteins to sustain internal cell processes, and exports the waste product lactate.

This bare-bones cell has already delivered one surprise. As expected, starving it of glucose causes a drop in levels of adenosine triphosphate (ATP), a key compound that provides the energy for many intracellular processes. But unexpectedly, before ATP levels drop they briefly rise. The reason, Tomita suspects, is that the early part of the ATP-producing pathway itself consumes ATP. Cutting the supply of glucose shuts down the early stages of the pathway, stopping ATP consumption there even while ATP continues to be produced from intermediary metabolites further down the pathway. Tomita thinks the effect may eventually be confirmed in living cells.

More surprises could be forthcoming when E-CELL is eventu-

spelled out their isolated functions. Now, it's time to move beyond reductionism.

"We have generated an enormous mass of information on the molecular events that occur in cells," says Marvin Cassman, director of the National Institute of General Medical Sciences (NIGMS) in Bethesda, Maryland. "Now we need to know how all these things are integrated." John Doyle, an electrical engineer at the California Institute of Technology in Pasadena who is turning his attention toward biology, puts it this way. "Biology has spent decades trying to be like physics," trying to understand complicated systems by understanding each part at its most basic level. "Now they're interested in putting it all back together."

Doing so, says Shapiro, will take "physicists, engineers, and biologists at lab benches next to one another working on the same problem." Foremost among these problems, say Shapiro and others, will be understanding the complex chemical networks that govern cell functioning. Genome analysis, for example, has already isolated hundreds of genes that code for transcription factors, proteins that help regulate the expression of other genes. "The expression of individual genes is not being regulated by one, two, or five proteins but by dozens," says Shirley Tilghman, a molecular biologist at Princeton University. Some regulate specific genes; others work more broadly. Some sit on DNA all the time, while others bind temporarily. "The complexity is becoming mind numbing," says Tilghman.

Simply determining the individual role of each protein only gets you so far. "Even if you have all the chemistry, it's hard to understand how the cell functions," says Adam Arkin, a physical chemist at Lawrence Berkeley National Laboratory (LBNL) in California. That's because interactions between different molecules can have a feedback effect that increases or decreases the expression of other compounds. "When we get to a certain network complexity, we completely fail to understand how it works," says Arkin.

BUILDING BOOM IN MULTIDISCIPLINARY CENTERS

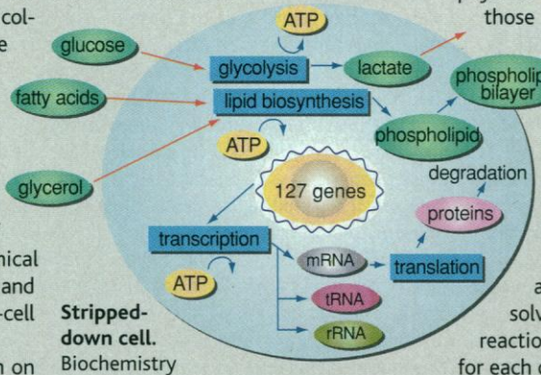
	Institution	Plan	Cost (in $ millions)
PROPOSED	Stanford	BioX (biophysics research)	??
	UC Berkeley/LBNL	Physical Biosciences	>100
	Univ. of Washington	Institute for Quantitative Systems Biology	??
APPROVED	Harvard	Center for Genomics and Proteomics	~50
	Princeton	Inst. for Genomic Analysis	70
	Caltech	Biological Sciences Initiative	100
	Univ. of Chicago	Biophysics Institute	110
	Univ. Pennsylvania	Inst. for Medicine & Engineering	20
	Stowers Institute for Medical Research	Genomics and Biotech Center	125
	Johns Hopkins	Whitaker Biomed. Eng. Institute	34
	Georgia Tech/ Emory University	Biotechnology Initiative	~75

Such complexity is well known in fields such as engineering. Take the latest Pentium chip in your desktop computer. The chip contains millions of individual elements, such as transistors, connecting wires, and gate arrays. The behavior of each element is understood to many decimal places. But for the engineers designing the chip, predicting how all the different elements would interact was a trickier proposition. Chip designers have to rely on sophisticated modeling programs to simulate how different collections of the elements interact and predict their collective behavior, so that they can iron out bugs in advance.

Now researchers are hoping to bring similar types of analyses to bear on understand-

ally put to work simulating whole cells of real organisms. Tomita admits that because building model cells with E-CELL depends on understanding the functions of large numbers of genes, the software is not likely to prove really useful for molecular biologists for some time. But he and his colleagues designed the program so that it should easily scale up to simulating the thousands of genes in a real cell. "Tomita and his group have done a fantastic job of engineering a 'graphical cockpit' for initializing and monitoring a whole-cell simulation," says Karp.

For greater realism on a smaller scale, users can turn to a different model-building kit: the Virtual Cell developed by physiologist Leslie Loew and computer scientist James Schaff of the University of Connecticut Health Center in Farmington. Rather than downloading software to run on their own computer, Virtual Cell users will simply run their simulation on Loew's host computer via the Internet. More important, rather than simulating an entire cell at once, as a biochemical system, Virtual Cell will eventually enable cell biologists to study how a cell's shape, volume, and other physical features affect individual biochemical processes.



Stripped-down cell. Biochemistry simulated by E-CELL software.

Loew's team builds its Virtual Cell models using precise measurements of how molecules diffuse and react within living cells, which they make by labeling key molecules and observing them with a video microscope. The result is a computerized cell with physical properties resembling those of real cells—a framework in which users can unleash specific biochemical reactions. For example, a researcher can add a certain amount of calcium—a key intracellular messenger—and then sit back and let the Virtual Cell solve equations describing reaction and diffusion rates for each of the molecular participants affected by calcium. Then the program generates a movie of the process. "The simulations are comfortable for the biologists to use because they are based on real image data," Loew explains.

In the case of calcium, the simulation not only looked much like the calcium waves measured in actual cells—indicating that the simulation was realistic—but it also predicted the dynamics of an intermediary molecule called IP3, which cannot be monitored inside the cell itself. (Demonstrations of Virtual Cell can be accessed at www.nrcam.uchc.edu)

"These two approaches can complement each other very well," Tomita says. And both are attracting growing interest from other biologists. Tomita says that when he first starting describing his plans for E-CELL, "I was dismissed as a naïve computer scientist." Now he gets e-mail requests for information on his simulation software nearly every day. Loew, too, has found that "interest has begun to mushroom." He adds, "[Cell biologists] are getting to the point that they are realizing that without computers we are never going to be able to organize all this information."

–DENNIS NORMILE

With reporting by Elizabeth Pennisi.