When and how did the command and control center of the eukaryotic cell arise?

The Birth of the Nucleus

LES TRELLES, FRANCE—What stands between us and *Escherichia coli* is the nucleus. Eukaryotic cells—the building blocks of people, plants, and amoebae—have these specialized, DNA-filled command centers. Bacteria and archaea, the prokaryotes, don’t. The nucleus’s arrival on the scene may have paved the way to the great diversity of multicellular life seen today, so the membrane-bound organelle fascinates scientists probing the evolution of modern organisms. “The question of the origin of the cell nucleus is intimately linked to the question of our own origin,” says Patrick Forterre, a molecular biologist at the University of Paris-Sud in Orsay, France.

Last month, Forterre and two dozen micro-biologists, evolutionary biologists, cell biologists, and others met here to hash out leading theories about the origin of the nucleus. One camp holds that the organelle is the result of a microbial merger. Another contends that residual nuclei hidden away in some bacteria indicate that the crucial innovation is far older than commonly thought. Perhaps the most radical theory of all puts viruses at the center of this cellular development.

At the meeting’s end, the discussions of the origin of the nucleus had left biologists with a key insight: They had underestimated the complexity of the eukaryotic cell’s 1.5–billion-year-old precursor. The data presented indicated that this ancestral cell had more genes, more structures, and more diverse biochemical processes than previously imagined.

But when it came to accounting for how the nucleus was born, no single hypothesis bubbled to the top. “It’s like a puzzle,” says Forterre. “People try to put all the pieces together, but we don’t know who is right or if there is still some crucial piece of information missing.”

Biologists have long considered the nucleus the driving force behind the complexity of eukaryotic cells. The Scottish botanist Robert Brown discovered it 180 years ago while studying orchids under a microscope. In his original paper, Brown called the novel cellular structure both an areola and a nucleus, but the latter name stuck. Now, as then, the organelle’s complexity inspires awe. The nucleus is a “huge evolutionary novelty,” says Eugene Koonin of the National Center for Biotechnology Information in Bethesda, Maryland.

Each nucleus in a eukaryotic cell consists of a double lipid-based membrane punctuated by thousands of sophisticated protein complexes called nuclear pores, which control molecular traffic in and out of the organelle. Inside, polymerases and other specialized enzymes transfer DNA’s protein-coding message to RNA. Other proteins modify the strands of RNA to ensure that they bring an accurate message to the ribosomes outside the nucleus. The nucleus also contains a nucleolus, a tightly packed jumble of RNA and proteins that are modified and shipped out of the nucleus to build ribosomes.

The picture is far different in bacteria, in which DNA, RNA, ribosomes, and proteins operate together within the main cell compartment. It’s a free-for-all in that as soon as the DNA code is transcribed into RNA, nearby proteins begin to translate that RNA into a new protein. In eukaryotes, “the double membrane [of the nucleus] uncoupled transcription and translation” and resulted in better quality control, says John Fuerst, a microbiologist at the University of Queensland, Australia. As a result, RNA is modified as needed before it comes into contact with a ribosome outside the nucleus.

The nuclear distinction between prokaryotes and eukaryotes shaped early speculations about the development of complex life. Until the 1970s, two competing theories dominated the debate over early eukaryotic evolution. According to one, a subset of bacteria slowly developed eukaryotic features, such as the nucleus. In the other, eukaryotes came first, but over time, some of them lost the nucleus and evolved a cell wall, spawning modern-looking bacteria.

Then the Woesean revolution struck. By looking at DNA sequence differences in the same gene across hundreds of microorganisms, Carl Woese, a microbiologist at the University of Illinois, Urbana-Champaign, showed that “bacteria” were actually two kingdoms, the bacteria proper and the archaea, which apparently arose some 2 billion years ago, millions of years before eukaryotes. The initial genetic analyses indicated that archaea were more closely related to eukaryotes than were bacteria. This kind of hinting that eukaryotes came from the seemingly simple archael stock.

Recent comparisons of fully sequenced microbial genomes have, however, added a twist to this story: Eukaryotes contain both archael and bacterial genes. Archaeal genes tend to run processes involving DNA and RNA, so-called information functions; the bacterial genes are responsible for metabolic and housekeeping chores. From the jumble of genes, some evolutionary biologists have concluded that this division of labor arose from the ancient symbiotic partnership between bacteria and archaea, a partnership that gave rise to eukaryotes.

*The Origin of the Nucleus* was held in Les Trelles, France, from 7 to 13 July.
Friendly mergers

Such a partnership may have been enough to create the nucleus, according to Purificación López-García and David Moreira of the University of Paris-Sud. The two evolutionary biologists speculate that the original union between bacteria and archaea grew from metabolic requirements. The nucleus, they further argue, arose as a way for these endosymbionts to keep their metabolic chemistries from interfering with one another. “You needed the [nuclear] membrane because you have two competing pathways,” López-García explains.

In 1998, she and Moreira proposed that in life’s earliest days, methane-making archaea sometimes lived within bacteria that depended on fermentation for sustenance: the so-called syntrophic model. The relationship worked for the archaea because fermentation yielded a resource they needed, namely hydrogen. The bacterium may have benefited because fermentation requires that hydrogen concentrations remain low.

López-García and Moreira hypothesize that Earth’s changing environmental conditions ultimately prompted a shift in the symbiosis. The archaea gradually lost its appetite for hydrogen, ceased making methane, and instead relied more on the bacterial host for other nutrients. The archaea’s membrane, which had been critical for methanogenesis, became superfluous. At the same time, the outer bacterial membrane invaginated the cellular compartment, eventually surrounding the archaeal DNA but excluding the ribosomes. The change was advantageous to the bacteria, because in separating ribosomes from the microbial chromosomes, it helped ensure more accurate conveyance of the DNA’s message. This set-up persisted and ultimately evolved into the eukaryotic nucleus, says López-García. And what remained of the archaeal cytoplasm became the nucleolus.

The researchers suggest that modern methanogenic archaea bearing a resemblance to eukaryotes are possible descendants of the ancient methanogens that entered into the nucleus-generating symbiosis with bacteria. These archaea and eukaryotes have similar genes encoding proteins involved with DNA and RNA. For example, they share genes for histones, proteins that help stabilize chromosomes. In contrast, bacteria don’t have histones.

Another modern microbe, the myxobacterium, may resemble the ancient bacterial host in which the nucleus evolved. Like eukaryotic cells, myxobacteria communicate with other cells, move, and can form multicellular complexes. Myxobacteria “have complex structures that are very striking” and reminiscent of eukaryotic cells, López-García notes. These bacteria also have cell-signaling molecules, such as kinases and G proteins, in common with eukaryotes.

Self-starters

López-García and Moreira’s proposal assumes that bacteria and archaea appear earlier on the tree of life than eukaryotes, but Fuerst holds that the reverse is true. He is convinced that eukaryote-like cells were around before bacteria and archaea or emerged right at the time when these prokaryotes split off to form separate kingdoms of their own. Fuerst points to an unusual group of bacteria that he’s studied for years, now known to be planctomycetes, to support his hypothesis.

In 1984, researchers had suggested that some planctomycetes also have nuclei, says Fuerst, and there are likely more, yet-to-be-discovered microbes with similar features. Bacteria with nuclear pores and internal membranes, features typically considered eukaryotic-specific, suggest that the nucleus was born much earlier than traditionally thought. If Fuerst’s scenario is correct, “then the nucleus actually preceded eukaryotes,” says Koonin.

In fact, this compartment could date back to the last universal common ancestor (LUCA), a putative organism from which eukaryotes, bacteria, and archaea eventually emerged, says Fuerst. If that’s the case, certain LUCA features, such as the nucleus, were retained in eukaryotes but lost to some degree in most archaea and bacteria. Indeed, that seems to be the case, as eukaryotic cells possess features now seen in each of these groups.

Hostile takeover

A third option for the origin of the nucleus revolves around viruses. “Viruses predated the divergence between the three domains of life,” says David Prangishvili, a virologist at the University of Regensburg, Germany. He argues that viruses were already quite common in the primordial soup and only later became dependent on cells to survive. When these early cells came along, “viruses played...
a critical role in the evolution of the complex [eukaryotic] system,” adds Forterre.

Viruses do have the ability to set up permanent residency in a cell, infecting but not killing the host. Thus they and their genes can stay around and influence a cell’s evolution. Bell, Forterre, Prangishvili, and Luis Villarreal, a virologist at the University of California, Irvine, each have a different proposal for how viruses were important to the evolution of the nucleus. Their supporting data are provocative, but circumstantial and controversial. “I do not believe [it],” says Jacomine Krijnse-Locker of the European Molecular Biology Laboratory in Heidelberg, Germany. “The idea of the viruses ‘inventing’ [eukaryotic cells] from scratch is hard for me to conceive.”

When viruses persist in cells instead of killing them, cells “can acquire a whole new set of genes in one event,” counters Villarreal. While in residence over millions of years, the new viral genes could have supplanted bacterial or archaeal genes, replacing, for instance, proteins that process DNA. These extra genes could also evolve to play new roles in the cell.

Villarreal points out that there are intriguing similarities between nuclei and viruses, which are basically packets of DNA surrounded by a protein coat—and often by a membrane. In red algae, for example, a nucleus can move from cell to cell, much like an infectious virus. And in general, cell nuclei and viruses lack protein- and lipid-producing pathways within their borders. Both contain linear chromosomes, whereas most bacterial chromosomes are circular. Both disassemble their “membrane” during replication. Both transcribe DNA but don’t translate mRNA within their boundaries. As they replicate within a cell, some poxviruses even make a membrane around their DNA using the endoplasmic reticulum of the infected cell. The eukaryotic cell uses this same material to build its nucleus.

Large, complex DNA viruses, which include poxviruses and the African swine fever virus, likely bear the closest resemblance to the putative viral ancestor of the nucleus, Bell suggests. DNA strands in these viruses have primitive telomeres, protective DNA sequences found at the ends of eukaryotic chromosomes.

Bell speculates that a virus living in an archaeum set the stage for the nucleus. Ultimately, viral DNA and archaeal DNA merged inside the virus, and the new genome later shed genetic material from both. In the end, “the unique genetic architecture of the eukaryote is a result of superimposing a viral genetic architecture on an archaeal genetic architecture,” Bell argues.

“If this is true, then we are all basically descended from viruses,” remarks Forterre.

Did a virus provide the first nucleus? Or was it something an early bacterial cell evolved, either on its own or in partnership with an archaeum? To resolve the origin of the nucleus, evolutionary biologists are exploring new techniques that enable them to determine relationships of microorganisms that go much further back in time. And as new genome sequences become available, such as those of several planctomycetes, Fuerst and others plan to search for more genetic similarities between these bacteria and eukaryotes. Meanwhile, García-López anxiously awaits sequenced genomes of myxobacteria and plans to compare them with the genes of eukaryotes.

Overall, says Forterre, it’s “a really exciting time to tackle questions which were previously only considered seriously by a few theorists.”

---ELIZABETH PENNISI

### Profile: Edward Hammond

**Activist Throws a Bright Light on Institutes’ Biosafety Panels**

Edward Hammond’s aggressive sleuthing has triggered a debate on the oversight of the growing field of biodefense research.

**AUSTIN, TEXAS—**In late January, Edward Hammond sent out a blizzard of faxes to almost 400 research institutes from Honolulu to New York. His request was straightforward enough: He asked for the minutes of the last two meetings of each organization’s Institutional Biosafety Committee (IBC).

Hammond, who directs the Sunshine Project, a small nonprofit organization based in Austin, wondered whether the IBCs fulfill their oversight role for certain types of biology experiments as prescribed by guidelines from the National Institutes of Health (NIH). In particular, he questioned whether they would publicly share their deliberations. Such openness, he says, is vital to prevent biodefense research from going astray.

Today, Hammond is fighting testy e-mail battles with his targets over their tardy responses. How to answer his query has become a hot topic among biosafety officers and university lawyers. Some universities have sent him minutes, but with almost every detail blanked out, arguing that the redacted information is private, proprietary, or security-sensitive. More important, Hammond has concluded that the IBC system, designed in the 1970s to review recombinant DNA research, is in disarray. He claims that dozens of IBCs, many of them at the nation’s research powerhouses, aren’t staffed properly, don’t seriously review proposals, or never meet at all. Outraged, he has filed complaints with NIH, asking it to cut off funding retroactively to 19 institutions. Dozens more complaints are on the way.

NIH officials are investigating the charges, but there’s no reason to assume that the entire system is broken, says Allan Shipp of NIH’s Office of Biotechnology Activities (OBA), which oversees IBCs. Most IBCs are “very earnest in their attempts and desire to fulfill their responsibilities,” he says.

Some researchers who have followed Hammond’s quest—he posts alleged violations frequently on his Web site—disagree. “Frankly, I’ve been surprised by the number and magnitude of the deviations from the