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THE BEAST WITH FIVE GENOMES

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*Inside a termite's gut lives *Mixotricha paradoxa*, a microscopic organism comprising hundreds of thousands of smaller life-forms. *M. paradoxa* is an extreme example of how all plants and animals—including ourselves—have evolved to contain multitudes.*

By Lynn Margulis and Dorion Sagan

The hullabaloo over mapping the human genome—the sum of all the genes in an individual—might lead one to think that each species has only a single genome and that the genetic makeup of individual organisms is discrete and unitary. Such is far from the case. Paraphrasing Walt Whitman, we multicellular beings contain multitudes. All animals' cells have at least two interacting genomes. One is the DNA in the cell nucleus; this is the genome that has recently been "mapped." The other is that of the DNA in the mitochondria—the cell's multiple oxygen-breathing organelles that are inherited only through the maternal line. For more than a century, some scientists have known that every organism is in fact a multiple being, but until recently these unorthodox researchers were ignored.

In most of the animals we think we know best (mammals, reptiles, insects), the genomes that determine limbs, eyes, and nervous systems, for example, are very similar to our own. These animals, like us, are doubly genomic. Even some unicellular beings that do not have eyes, limbs, or nervous systems—such as amoebas and paramecia—contain both nuclear and mitochondrial genomes. Plants and algae have these double genomes as well, plus a third genome, of symbiotic origin. During their evolutionary history, they ingested (but did not digest) photosynthetic blue-green bacteria. Therefore, all visible photosynthetic organisms have at least three genomes. But many organisms—such as the protists that inhabit termites—contain within them up to five or more genomes.

The great nineteenth-century naturalist Joseph Leidy, one of the founders of the Academy of Natural Sciences in Philadelphia, was the first to take a close-up look at the contents of a termite's gut. "In watching the Termites from time to time wandering along their passages beneath stones," he wrote, "I have often wondered as to

what might be the exact nature of their food." What he saw under his microscope amazed him. If the termite's intestine is ruptured by the experimenter, he wrote, "myriads of the living occupants escape, reminding one of the turning out of a multitude of persons from the door of a crowded meeting-house." Leidy immediately realized that what he knew as "white ants" were actually composed of dozens of different kinds of tiny life-forms, including bacteria and what we now call protists. (Protists are microbes with nuclei; more complex than bacteria, the group includes amoebas, slime molds, and algae.) We now recognize that the immense and motley crew that Leidy observed within a termite is in no way a gratuitous add-on or a pathological infection. Rather, it is a necessary part of the termite's digestive system and is organized as a particular tissue: an aggregate working mechanism that turns the refractory compounds lignin and cellulose (the main constituents of wood) into food. This composite fabric, or living consortium, has evolved in the nearly oxygen-free closed system of the termite's abdomen for probably 100 million years; without the living, wood-degrading factories that have become their digestive systems, these termites starve.

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The pioneering biologist Konstantin S. Merezhkovsky first argued in 1909 that the little green dots (chloroplasts) in plant cells, which synthesize sugars in the presence of sunlight, evolved from symbionts of foreign origin. He proposed that "syntrophogenesis"—a term he coined for the merger of different kinds of life-forms into new species—was a major creative force in the production of new kinds of organisms. A Russian anatomist, Andrey S. Famintsyn, and an American biologist, Ivan E. Wallin, worked independently during the early decades of the twentieth century on similar hypotheses. Wallin further developed his unconventional view that all kinds of symbioses played a crucial role in evolution, and Famintsyn, believing that chloroplasts were symbionts, succeeded in maintaining them outside the cell. Both men experimented with the physiology of chloroplasts and bacteria and found striking similarities in their structure and function. Chloroplasts, they proposed, originally entered cells as live food—microbes that fought to survive—and were then exploited by their ingestors. They remained within the larger cells down through the ages, protected and always ready to reproduce. Famintsyn died in 1918; Wallin and Merezhkovsky were ostracized by their fellow biologists, and their work was forgotten. Recent studies have demonstrated, however, that the cell's most important organelles—chloroplasts in plants and mitochondria in plants and animals—are highly integrated and well-organized former bacteria. Using new methods, scientists have been able to raise and resolve the question of how these bacteria became permanent symbionts.

Like other animals, we harbor in our intestines an assortment of specific microbes that help us digest food, although some are also able to live outside humans. Few of our microbes are organized as layers of tissue,

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as they must be in termites. Nevertheless, without these hitchhikers to help digest fiber and produce vitamins, we—like termites—weaken and even die. Entirely integral to our bodies, however, are the mitochondria in our nucleated cells. These tiny entities use oxygen to generate the chemical energy needed to sustain life. They reproduce on their own, independently of the nuclear

DNA, and multiply more quickly after short bursts of muscular exercise, leading to stronger, more mitochondria-packed muscles. Because mitochondria are so genetically integrated into each of our cells, no one has yet succeeded in growing them in test tubes.

We believe that Wallin and Merezhkovsky were fundamentally correct when they claimed that all nucleated living things evolved by syntrophogenesis, generally because of preexisting bacterial genomes physically associated with other organisms. Reef-building corals, for instance, are now known to have five different genomes of once independent organisms. And *Mixotricha paradoxa*, a compound beauty found in a termite's gut, also has five genomes. Indeed, *M. paradoxa* could well be the "poster animal" for syntrophogenesis.

In 1933 Australian biologist J.L. Sutherland first described and named “the paradoxical being with mixed-up hairs” (she mistakenly thought it was the only microbe that swims by simultaneously using both flagella and cilia). Studies done by A.V. Grimstone of Cambridge and the late L.R. Cleveland of Harvard in the 1950s with the electron microscope showed that *M. paradoxa* was a hundred times larger than its close relatives, that four different kinds of bacteria were part of its body, and that it lacked mitochondria.

For many years, we have studied and photographed this organism. Under low magnification, *M. paradoxa* looks like a single-celled swimming ciliate. With the electron microscope, however, it is seen to consist of five distinct kinds of creatures. Externally, it is most obviously the kind of one-celled organism that is classified as a protist. But inside each nucleated cell, where one would expect to find mitochondria, are many spherical bacteria. On the surface, where cilia should be, are some 250,000 hairlike *Treponema spirochetes* (resembling the type that causes syphilis), as well as a contingent of large rod bacteria that is also 250,000 strong. In addition, we have redescribed 200 spirochetes of a larger type and named them *Canaleparolina darwiniensis*.

Acceptance of the composite nature of individuals, we predict, will soon revolutionize evolutionary biology. Bacteria are exemplary genetic engineers: splicers and dicers and mergers of genomes par excellence. Devoid of immune systems, always reproducing without mate recognition, bacteria are supremely promiscuous beings in which infection and sex—that is, gene flow—are virtually the same thing. The sexual proclivities of bacteria include (when their survival is threatened) rampant exchange of genes—next to which our own species’ most bacchanalian orgies look like rather subdued affairs.

Biologists have always puzzled over why there are so many kinds of beetles. Perhaps symbionts beneath the surface, generating variety at the genomic level, account for nature’s beetlemania. Insects have integrated bacterial genomes to an extraordinary degree. In many cases, bacteria reside in all the tissues, accumulate in the eggs, and are inherited. Beetles have developed partnerships with an extremely diverse assortment of bacteria; many more kinds live inside their tissues than live in most other groups of animals.

Eventually we may well realize that natural selection operates not so much by acting on random mutations, which are often harmful, but on new kinds of individuals that evolve by symbiogenesis. Scrutinizing any organism at the microscopic level is like moving ever closer to a pointillist painting by Georges Seurat: the seemingly solid figures of humans, dogs, and trees, on close inspection, turn out to be made up of innumerable tiny dots and dashes, each with its own attributes of color, density, and form.

As a graduate student in genetics at the University of Wisconsin, **Lynn Margulis** learned about “cytoplasmic genes”—such as those that determine green color in plants—and recognized that they belonged to microbes that were once free living. Now a Distinguished University Professor at the University of Massachusetts Amherst, she is still exploring the implications of such ancient mergers. Margulis has collaborated with writer **Dorion Sagan** on numerous books, including *Slanted Truths: Essays on Gaia, Symbiosis and Evolution* (Springer-Verlag, 2001). Their collaborative work is accessible at www.sciencewriters.org. Sagan is now coauthoring a book on the evolution of intelligence.

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