

## Phylogeny: Are Methanogens a Third Class of Life?

Modern biology's concept that living organisms split into two major lines of descent more than 3.4 billion years ago may need some revision. Recent evidence from a team of investigators at the University of Illinois headed by Carl R. Woese and George Fox (who is now at the University of Houston) suggests that there is at least a third line of descent, and perhaps more.

The two lines of descent are prokaryotes, organisms such as bacteria and blue-green algae whose cells do not have a well-defined nucleus, and eukaryotes, more complex organisms whose cells do have a nucleus. Organisms can also be placed in these categories on the basis of similarities in metabolism and genetics. Members of the proposed third class, the Illinois group says, superficially resemble bacteria, but have genetic and metabolic characteristics that make them unique. These organisms are methanogens, anaerobic, methane-producing microorganisms that occur in places as disparate as the gastrointestinal tract of man and animals, sediments of natural waters, sewage treatment plants, and hot springs. They participate in the terminal stages of the degradation of organic matter, living on the carbon dioxide and hydrogen produced by anaerobic bacteria and converting them to methane.

Three main lines of evidence indicate that the methanogens are no more related to the bacteria than bacteria are related to higher organisms:

- ▶ The cell walls of methanogens do not contain muramic acid, the characteristic constituent of the peptidoglycans that form bacterial cell walls. This has been demonstrated chiefly by Otto Kandler of the University of Munich, in collaboration with Ralph Wolfe of the University of Illinois.

- ▶ The metabolism of methanogens is substantially different from that of bacteria. Drawing on preliminary work by Horace A. Barker of the University of California at Berkeley and Robert E. Hungate of the University of California at Davis, Wolfe and his colleagues have so far identified several coenzymes that appear to be unique to methanogens. The best characterized of these is 2-mercaptoethanesulfonic acid. It is involved in methyl transfer reactions, including the formation of methane, and is one of the smallest coenzymes that has been discovered. A second coenzyme is a yellow-fluorescent heterocyclic ring system

that participates in electron transfer reactions, a function that is performed in most other organisms by flavin coenzymes or nicotinamide adenine dinucleotide. The other coenzymes in the methanogen metabolic pathway have been only preliminarily identified.

A further difference is the way in which carbon dioxide is fixed into cellular carbon. Several investigators, including J. G. Zeikus of the University of Wisconsin, have been unable to demonstrate in methanogens the carbon fixation pathways that are shared by other organisms. The way in which carbon is fixed has not yet been determined.

- ▶ The methanogens have RNA sequences that are markedly different from those in other organisms. Transfer RNA's in nearly all other organisms previously investigated, for example, share the common base sequence thymidine-pseudouridine-cytidine-guanosine in one arm. Woese has shown that in at least one of the methanogens, the thymidine has been replaced by pseudouridine and both cytidine and guanosine have been modified in an as yet undetermined manner. Similar modifications also occur in the RNA sequences of methanogen ribosomes. To evolutionists Woese and Fox, who trace the genealogical history of organisms through such sequence similarities and differences, these results suggest that the methanogens share a common ancestor with prokaryotes and eukaryotes, but that they branched off as an independent line of descent about the same time the other two diverged.

The methanogens seem to have changed very little since that divergence. They are ideally suited to the conditions that are thought to have existed then, particularly an atmosphere that lacked oxygen and was rich in hydrogen, ammonia, and carbon dioxide. Their relatively simple metabolic requirements, furthermore, could easily have been met in the primeval oceans even if those oceans lacked the sugars and amino acids required by other organisms.

That methanogens are unusual is not, of course, a new discovery. As long ago as the mid-1950's, Barker argued that they represent a radically different class

of bacteria. The new findings about their cell walls and metabolism accentuate the difference, but are probably not strong enough evidence in themselves to warrant reclassification of the methanogens. The crux of the matter, then, is the significance of the differences in RNA.

Determining the phylogeny of organisms by examining molecular differences is now a well-accepted field of study. Subtle differences between the cytochromes or ferredoxins of various species, for example, have clarified the relationships between these species and the course of their evolution. The Illinois group has been studying the RNA sequences of the 30S ribosome subunit because it is universally distributed, whereas cytochromes and ferredoxins are not present in many species, including most methanogens. Few evolutionists have had time to evaluate Woese's most recent work on methanogens, which is to be published in the October and November issues of *Proceedings of the National Academy of Sciences*, but he has a reputation as a careful, accurate investigator. His results are thus not likely to be questioned, only their interpretation.

Marvin Bryant of the University of Illinois—a leading expert on methanogens and methane fermentation, but not an evolutionist—suggests that Woese's results are sufficient to classify the methanogens only as an early form of bacteria and not as a class separate from prokaryotes. He argues that the structural and molecular similarities between methanogens and bacteria may be more important than the differences. Evolutionist Walter Fitch of the University of Wisconsin, however, says that if all of the Illinois group's evidence is correct, then their interpretation is probably also valid. Ultimately, it would appear that acceptance of the Illinois group's arguments will depend on the strength of one's faith in phylogenetics.

Even if Woese and Fox are right, recognition of a third line of descent will have little effect on practical problems in biology. But that recognition may have a profound effect on our understanding of the nature of life during the earliest stages of its development and of the evolution of more complex organisms. It might also give us a better idea of what man might expect to find when he ventures toward other planets that resemble Earth in its formative years.

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*Erratum:* Due to a last minute printer's error, the 18 November Table of Contents omitted mention of a Research News article on "The 1977 Nobel Prize in chemistry" by I. Procaccia and J. Ross.