530-51
ABS ONLY
188877
18.

N89-26364

PHYLOGENETIC PERSPECTIVE AND THE SEARCH FOR LIFE ON EARTH AND ELSEWHERE

Norman R. Pace
Department of Biology and
Institute for Molecular and Cellular Biology
Indiana University IE 449333
Bloomington, IN 47405

Studies in molecular phylogeny over the past decade have provided a quantitative outline of the evolutionary relationships among known lifeforms. Because of their ubiquity and constancy of function, the ribosomal RNA (rRNA) sequences are particularly useful for molecular phylogenetic analyses. Employing rRNA sequence similarities, C. R. Woese and colleagues have defined the three primary lines of evolutionary descent: the eukaryotes, the eubacteria, and the archaebacteria.[1,2] The rRNA structures of the three primary lineages share many features that are identical or demonstrably homologous, proving that all known, extant organisms are derived from a common ancestor, the progenote. The rates of change of sequences in different lineages, at various times during their evolution, differ significantly. Thus, phylogenetic trees cannot be interpreted in terms of absolute time. It is nonetheless clear that the eukaryote nuclear line of descent arose as early as the two prokaryotic lineages. The major organelles, the chloroplasts and mitochondria, are unequivocally of eubacterial origin. The nature of the progenote in principle may be inferred by the identification of biochemical features that are common to the three primary lineages.

Because terrestrial organisms were derived from a single ancestor, we have no perspective on the potential diversity available to lifeforms that might arise elsewhere. Thus, the discovery of living or fossil organisms on Mars would be of profound importance. Their commonalities with and differences from terrestrial organisms would provide entirely new insight into the origins of life and the fundamental properties of biological systems.

Any search for microbial life on Mars cannot rely upon cultivation of indigenous organisms. Only a minority of even terrestrial organisms that are observed in mixed, naturally-occurring microbial populations can be cultivated in the laboratory. Consequently, we are developing methods for analyzing the phylogenetic affiliations of the constituents of natural microbial populations without the need for their cultivation.[3,4] This is more than an exercise in taxonomy, for the extent of phylogenetic relatedness between unknown and known organisms is some measure of the extent of their biochemical commonalities. In one approach, total DNA is isolated from natural microbial populations and 16S rRNA genes are shotgun cloned for rapid sequence determinations and phylogenetic analyses. A second approach employs oligodeoxynucleotide hybridization probes that bind to phylogenetic group-specific sequences in 16S rRNA.[5] Since each actively growing cell contains about 104 ribosomes, the binding of the diagnostic probes to single cells can be visualized by radioactivity or fluorescence. The application of these methods and the use of *in situ* cultivation techniques is illustrated using submarine hydrothermal vent communities.[6,7]

Regarding planning toward future Mars missions:

1. A specific recommendation that derives from this work is that future intensive searches for life on Mars must employ *in situ* procedures. It is unlikely that Martian microorganisms, if

they exist, will be cultivatable using standard techniques. The capabilities of *in situ* analysis should, as much as possible, focus toward retrieving biochemical information from a single cell. The required technology would be an enormous boon to terrestrial biological studies, as well.

2. Any future consideration of oases for life on Mars must treat not only the surface environment, but also the possibility of subsurface ones. Possible scenarios would include convection-driven aquifers associated with volcanic features.

3. A previously-discussed design concept that was amplified in the discussions should receive further attention. This is the notion of a robust, detector-laden probe, perhaps a cane-like device, that could be used as a prod for inspecting (and sniffing) surfaces and cavities.

REFERENCES

1. Woese, C. R. 1987. Bacterial evolution. *Microbiol. Rev.* **51**:221-271.
2. Pace, N. R., G. J. Olsen, and C. R. Woese. 1986. Ribosomal RNA phylogeny and the primary lines of evolutionary descent. *Cell* **45**:325-326.
3. Pace, N. R., D. A. Stahl, D. J. Lane, and G. J. Olsen. 1986. The analysis of natural microbial populations by ribosomal RNA sequences. *Adv. Microbial Ecol.* **9**:1-55.
4. Olsen, G. J., D. J. Lane, S. J. Giovannoni, D. A. Stahl and N. R. Pace. 1986. Microbial ecology and evolution: a ribosomal RNA approach. *Ann. Rev., Microbiol.* **40**:337-365.
5. Giovannoni, S. J., E. F. DeLong, G. J. Olsen, and N. R. Pace. 1988. Phylogenetic group-specific nucleic acid probes for identification of single microbial cells. *J. Bacteriol.* **170**:720-726.
6. Stahl, D. A., D. J. Lane, G. J. Olsen, and N. R. Pace. 1984. The analysis of hydrothermal vent-associated symbionts by ribosomal RNA sequences. *Science* **224**:409-441.
7. Karl, D. M., G. T. Taylor, J. A. Novitsky, H. W. Jannasch, C. O. Wirsen, N. R. Pace, D. J. Lane, G. J. Olsen, and S. J. Giovannoni. 1988. A microbiological study of Guaymas Basin high temperature hydrothermal vents. *Deep Sea Research*, in press.