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## **Microbial Diversity and Ecology in the Natural Ecosystem**

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Earth was born 4.6 billion years ago as a planet of the solar system. Then, after the chemical evolution of organic compounds 4.0 to 3.5 billion years ago life was created by accident. The ancient cells are presumed to have RNAs as a hereditary element. The RNA was substituted by DNA, and the present forms of living cells (with protein-synthesizing ability) are the DNA organisms. The DNA organisms evolved to presently existing various descents. Motive forces for the explosive evolution are environmental changes of the biosphere. The dropping temperature of the biosphere was one of the most important environmental changes for the early evolution. The ancient organisms living before 3.0 billion years ago are presumed to be thermophiles which liked higher temperature than 70. The temperature of the surface of the earth dropped gradually under 70 after 3.0 billion years ago, and the thermophilic organisms were drove into hot springs or hydrothermal vents of sea bottom volcanoes. The second important environmental change of the biosphere was the generation of oxygen. Oxygen was produced by cyanobacteria in the sea, and after saturated in the sea it was released into the atmosphere. The generation of oxygen drove away the primitive anaerobic bacteria and created aerobic bacteria. The primitive forms of anaerobic bacteria are presumed to be archaebacteria. As the concentration of atmospheric oxygen increased, the aerobic bacteria diversified explosively about 2.0 billion years ago, the eukaryotes were created by endosymbiosis about 2.0 billion years ago, and various multicellular organisms were created up to 0.7 billion years ago.

### **Evolution of microorganisms**

Phylogenetic relation between microorganisms could not be revealed by fossil informations. Information in macromolecules, namely, amino acid sequences of proteins or nucleotide sequences of genes could reveal the phylogenetic relation between microorganisms. Covering most of living organisms the macromolecule

should be the molecule which exists ubiquitously in organisms. The phylogenetic relation drawn by ribosomal small subunit RNA between presently living organisms is the most familiar and reliable tree. Very similar phylogenetic trees were drawn by many other macromolecules such as F1 ATPase and elongation factor for the peptide synthesis.

The primitive life created 4.0 to 3.5 billion years ago diversified into three major lineages at the very early stage of its evolution. The major three lineages are called domains Bacteria (Eubacteria), Archaea (Archaeobacteria), and Eucarya (Eukaryotes). The Bacteria diversified into about 12 groups, and the Archaea into three groups. The Eucarya consists of animals, plants, fungi, algae, and protozoa, however, this classification does not necessarily reflect their phylogenetic relationships. Some fungi (the Oomycetes) are apparently related to yellow algae, and some protozoa (Flagellata) are related to the Euglenophyta.

### **Importance of endosymbiosis for the evolution of microorganisms**

Endosymbioses played very important role for the evolution of life. Energy producing organs of eukaryotes, mitochondria were derived from the endosymbiosis of a proteobacteria, and organs for photosynthesis of plants and algae, chloroplasts were from cyanobacteria. Algae and plants showed heterogeneity in their morphology of chloroplasts and nuclei. The Rhodophyta and Chlorophyta have chloroplasts surrounded with two membranes, the Dinophyta and Euglenophyta with three membranes, and Chromophyta and Cryptophyta with four membranes. The Cryptophyta have second nucleus in the chloroplasts, and molecular phylogenetic studies showed that the two nuclei had different origins. The heterogeneity in the morphology of chloroplasts and nuclei in algae implies that many kinds of endosymbioses occurred between eukaryotes.

### **Microbial symbioses in animals and plants and their ecological roles**

Microorganisms with photosynthetic, chemoautotrophic, nitrogen fixing or some special ability have been targets of symbioses in the evolution of animals and plants. In lichens algae are captured by fungi. Fungi receive energy sources from algae, and algae extend their living territory to dry environments. In corals various kinds of algae are captured as energy producers. Invertebrates living around hydrothermal vents of sea bottom volcanoes keep chemoautotrophic bacteria in their gills, and the bacteria supply energy for the

hosts. In plants root nodules were induced by many kinds of nitrogen fixing bacteria. The most famous nodulating bacteria were rhizobia and frankia. Rhizobia induce nodule on leguminous plant roots, and frankia on forest trees such as *Alnus*, *Myrica*, and *Casuarina*. Root nodule forming bacteria are the major nitrogen suppliers in the soil. Plant root symbiotic fungi, mycorrhizae play very important roles for plant absorption of phosphate. There are many other forms of symbioses and association of microorganisms with animals and plants in the environments. Such symbioses and association came of the evolution and diversity of microorganisms.

### **Diversity of unculturable microbes in the natural ecosystem**

The three domain system has been widely accepted since the establishment of the concept of Archaeobacteria (=Archaea). However, many of microorganisms living in natural environments have not been cultured. It is possible to find out extremely ancient cells in the microbial community. Recent developments in the analysis of both DNA and RNA recovered from natural environments are leading the way to a new stage of microbial ecology which elucidates phylogeny and the ecological roles of uncultured microorganisms. In the course of flora analysis of soil Archaea, we found very strange 16S rDNA clones, which could possibly constitute a sister clade from known two archaeal, Crenarchaeota and Euryarchaeota, lineages. Overall signature sequences showed that the clones were closely related to domains Archaea and Eucarya. However, at least nine nucleotides distinguished the novel clones from domains Archaea and Eucarya. Phylogenetic trees drawn by maximum parsimony, neighbor joining and maximum likelihood methods showed unique phylogenetic position of the clones. Both signature-sequence and phylogenetic analysis strongly suggest that the novel organisms may constitute a new group and their phylogenetic position may be distant from Crenarchaeota and Euryarchaeota. A very specific primer set was synthesized to detect the presence of the novel group of organisms in terrestrial environments. A specific DNA fragment was amplified from all of paddy soil DNAs, and this fact suggests that the novel organisms are anaerobic. The number of novel cells per gram of wet paddy soils is presumed to be between  $10^3$  to  $10^5$ .

### **Future of the human race, the global environment, and microbial ecology**

Human beings by their instincts made a very convenient society, and are

taking tasty foods. However, human beings apparently are destined to become extinct because of the explosive growth of population and of the demolition of environments. Now, important things are how to delay the extinction.

Major global environmental issues are globally warming temperature by green house effect and demolition of ozone layer. These two subjects are very difficult to handle relating to politics, economics, life style, etc. In the fields of environmental sciences, microbiology will not play so important role. Microbiology will be of use for remediation of chemically polluted environments, wastewater treatments, etc.

In the agricultural fields it is important to develop sustainable, highly productive, and energy saving agriculture. Nitrogen and phosphate adsorptions are the most important factors for plant growth. The adsorptions of the two elements are related to the microbial activities in the soil, and high productivity of crops can not be given without assistance of microorganisms. Plant diseases can be reduced to some extent by microbial flora control without using agricultural chemicals.

In 20th century applied microbiology played very important roles in human disease control, developing genetic engineering, etc. In 20th century microbial ecology have not made a rapid progress compared with applied microbiology. As mentioned above, microbial ecology is related to crop production and environmental sciences. As in 21st century crop production and environmental problems will be major subjects, progresses in microbial ecology will be strongly expected.

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