Detection of the subsurface hot biosphere by microbial molecular thermometer (MMT)

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Subsurface environments of deep-sea hydrothermal vents and terrestrial hot springs are considered extremely hot environment. The hydrothermal fluid includes a variety microorganism having high growth temperatures from a subsurface hot environment. To date, the microbial diversity in various hydrothermal fluids has been revealed by the 16S rRNA gene-based analysis. However, the molecular approach cannot elucidate the real temperature of where these thermophilic prokaryotes inhabit, because most of the obtained rRNA gene fragments are derived from yet-uncultivated archaea and bacteria. To estimate growth temperatures of these yet-uncultivated prokaryotes, we propose a new method, microbial molecular thermometer (MMT), based on the strong correlation between the guanine-plus-cytosine (G+C) contents of the 16S rRNA gene sequences and the growth temperatures of prokaryotes. Based on G+C contents of the obtained 16S rRNA gene sequences, we can easily estimate the optimal growth temperatures of indigenous prokaryotes. Archaeal 16S rRNA gene fragments in the bulk DNAs extracted from various hydrothermal and geothermal fluid samples were amplified, and the sequences of 16S rRNA genes were determined. The fluid samples were collected in the deep-sea hydrothermal vents in Suiyo Seamount, southern Mariana Trench and Kagoshima Bay, and the terrestrial hot spring area in Nakabusa, Nagano. In some fluid samples of southern Mariana Trench (117^{o}C) and Nakabusa hot spring (84^{o}C), the growth temperatures estimated by MMT almost agreed with their observational temperatures. On the other hand, some archaeal 16S rRNA gene fragments obtained in the Suiyo Seamount hydrothermal fluid (40^{o}C) showed a high G+C content (63-66%) indicating a high growth temperature of 70-85^{o}C. The same finding was suggested in the terrestrial hot spring fluid (74^{o}C) of Nakabusa hot spring. Many archaeal 16S rRNA gene fragments with a high G+C content (65-67%) were found, and high growth temperatures of 83-89^{o}C were inferred from these high values. The obviously higher growth temperature estimated by MMT than the actual fluid temperature suggested that the fluid came from a hotter subsurface environment and the detected hyperthermophilic archaea probably inhabited such hot subsurface biosphere.

Microbial Molecular Thermometer (MMT)

In recent years, international deep drilling project, such as Integrated Ocean Drilling Program (IODP) and International Continental Scientific Drilling Program (ICDP), have been expanded for the purpose of scientific study, including microbiological research. Many microbiologists have become very interested in microorganisms in the subsurface hot environment, because of the number and variety of unknown microbes that it harbors. Here, we were able to suggest the presence of the subsurface hot biosphere in the deep-sea hydrothermal vents and the terrestrial hot spring area based on microbial molecular thermometer (MMT). It is likely that MMT will play an important role to select the drilling sites in the scientific drilling program targeting subsurface hot biosphere.

References

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