Quantitative microbial community analysis of hydrothermal fluids from Suiyou-seamount

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In the caldera of Suiyo seamount, highly hydrothermal activities have been observed through the existence of many hydrothermal vents. Hydrothermal chemical components of end members showed that varied temperature of hydrothermal fluid was just diluted by surrounding seawater. Even in the high temperature hydrothermal fluid, more than 10000 microbial cells per ml have been detected. However, roles of microbes in hydrothermal fluid, and relationship between the density of microbial cells and the concentration of hydrothermal fluid are still unclear. In this study, we investigated the microbial community structures and distributions in various hydrothermal fluids through quantitative fluorescent in situ hybridization methods, to refer to the microbial world under Suiyo seamount hydrothermal system.

The hydrothermal fluid for microbial analysis was sampled with hybrid particle & fluid sampler using Shinkai 2000 during NT-01-09 cruise and NT-02-09 cruise in Suiyo Seamount caldera. The temperature of hydrothermal fluid samples was varied from below 10 C to 310 C. After one tenth volume of 38% nutrilized formalin was added to 100-200 mL of the samples, those were kept in the refrigater (4C) overnight to fix microbial cells. Some of the samples were filtrated onto PLL-filter with optimum cell numbers for microscopic observation, and then the PLL-filters were kept and brought back to the laboratoty under -80 C. The rest of the samples were also frozen and brought back to the laboratory under -80 C. These samples were used for the measurement of the number of each microbial group such as Bacteria, Archaea, and some sulfur oxidizers by FISH-DC method. Addition to microscopic analysis, microbial phylogeny based on 16S rDNA was investigated using particle trapped filter samples which obtained by in situ filtration with the hybrid particle & water sampler.

In general microbial cell counting method, microbial cells are stained with DNA fluorochromes such as DAPI and SYBR Green. However, it is difficult to count microbial cells in the hydrothermal fluid, because the hydrothermal fluid contains not only microbes but also many inorganic mineral particles those have fluorescence. In FISH, the specific sequences of microbial rRNA were specifically recognized and stained by fluorescent labeled DNA probes. By using FISH method, microbial cells are specifically detected and easily counted. Most of microbial cells in the hydrothermal fluids were belonged to Domain Bacteria, and cells belonged to Domain Archaea were only a few. The numbers of Bacteria cells varied from under detection limit, e.g. 5 x 10^3 cells per ml, to 10^6 cells per ml. The cell numbers were not related with the temperature and Si concentration, which suggested the purity of the original hydrothermal fluid. From some low-temperature hydrothermal vents, SUP05 cells, which are the dominant microbial group in hydrothermal plume, were detected. No hyperthermophilic-bacterial sequence was found from the microbial phylogenetic analysis using the particle entrapped filter sample. From these results, the original high temperature hydrothermal fluid contains no microbes and most of microbial cells detected in hydrothermal vent fluids were suggested to originate from the surrounding sand or mytilids colonies.