Broad-host-range Gene Transfer Particles

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Viruses or virus-like particles (VLPs) have been acknowledged to be general constituents of common aquatic ecosystems, whose postulated roles are microbial population control and gene transfer. Viruses have been assumed to have little importance because bacterial cells generally restrict their acceptance of foreign genetic materials. It is also believed that the host range of bacterial viruses (bacteriophages) is restricted to specific bacterial strains or closely related species. While knowledge of transduction and VLP-host interaction has accumulated mainly through studies of mesophilic microbial communities, there is insufficient information about the distribution and ecological roles of VLPs in thermal environments.

We have been studying marine and thermophilic VLPs, which are produced by spontaneous induction in bacteria. Such VLPs, whose original hosts are not related to Escherichia coli at least at the family level, showed mild lethal effect (reduced population up to 1/10) on E. coli recipients with gene transfer capability (10^-2 - 10^-6 cfu/VLP). It is noteworthy that the transductant acquired VLP productivity, which showed similar lethal effect and/or gene transfer ability to the recipient as well. Such VLPs showed a reduction of viable cell count 'infection' without forming plaques on original host and E. coli recipients. The findings imply that a 'broad-host-range vector particle' is prevalent in Nature. It is also suggested that at least some environmental VLPs may share similar characteristics, a phenomenon not explained by the general features of lytic or lysogenic cycles of viruses that infect microorganisms.

Samples were collected from a geothermal hot spring, Nakanoyu, Nagano, and a relatively high number of VLPs in the water (up to 10^6 particles per ml) was observed. Mature VLPs were observed inside about 12 % of the bacteria, which were classified as a novel hyperthermophilic chemolithotrophic sulphur-turf predominant bacteria belonging to the deepest-branching lineage of the domain Bacteria (Aquificales). VLP-mediated gene transfer was experimentally demonstrated using auxotrophic mutants of E. coli and Bacillus subtilis with an average efficiency of 10^-6 cfu per particle. The 'VLP' originated from these thermophilic bacteria may be a xenotrophic gene transfer particle. The particle from thermophilic, sulphur-oxidizing bacteria was able to accomplish gene transfer from Gram-negative bacteria to Gram-positive bacteria. Particle production from the generated transductants was confirmed.

From these results, it is strongly suggested that at least some of the widely distributed VLPs could be general gene transfer agents among a wide range of microbial host cells and might function as a universal vector. Considering that 'particles', which could mediate gene transfer between genetically distinct host cells, were obtained from such dissimilar environments as hot spring and the ocean, gene transfer mediated by 'VLPs' may be a ubiquitous event in the natural environment.

In order to examine such a high transduction frequency, the objective was extended to the thermal water column, and 'VLP'-mediated gene transfer experiments were conducted. VLP and cell abundance in water samples from geothermal vents in Toyoha Mine drift way (78 C), and a hydrothermal vent (255 - 308 C) in the Suiyo Seamount were ca 10^8 (Toyoha Mine) and ca 10^4 (Suiyo Seamount) particles/cells per ml, respectively. Gene transfer frequency observed for amino acid requirements restoration of auxotrophic E. coli was 10^-4 (Toyoha Mine) and 10^-8 (Suiyo Seamount) cfu per particle. Preliminary results using 'VLPs' from these geo- and hydrothermal vents supported the above hypothesis. The non-specific gene transfer by such particles from a hydrothermal vent implies that such gene transfer particles have mediated gene flux among phylogenetically diverse bacterial communities since the early age of the Earth.