Separation of DNA by Free Flow Electrophoresis in Space

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Free flow electrophoresis of a nematode *C. elegans* DNA was carried out on the space shuttle flight in International Microgravity Laboratory No.2 (IML-2), 1994.

We selected the *C. elegans* DNA as the sample of the space experiment for free flow electrophoresis separation. This worm is a useful animal for the study of development and behavior by genetic analysis, and is a good candidate for a complete DNA sequence analysis because the haploid genome size consists of approximately 100 Mb (megabase) distributed on the six chromosomes, only 1/30 of human genome size. (Recently, the entire genomic DNA (97 Mb) sequences of *C. elegans* have been completed at the last December, 1998, as the first organism of multicellular system. In addition, many of the genes in *C. elegans* have extensive similarity to their mammalian counterparts. It may be possible to use the technology of free flow electrophoresis to contribute to the DNA analysis of the eukaryote.

In this mission we attempted to make real-time communication between the space shuttle and on the ground during the electrophoresis processing. Because the separation tendency of the sample in space was not predicted so perfectly, the requests to the crews of the space shuttle was needed for selecting the fractions to be separated. According to the three dimensional electropherogram (3DEP) figured out the electrophoresis behavior, we were able to recover those fractions and kept in the deep freezer until landing. This real-time monitoring of the electrophoresis was the first evidence during space electrophoresis experiments which was started at Apollo 14, 1974.

Unfortunately, some troubles occurred by contamination of bubbles in space nevertheless the post flight analyses of the fractionated samples were succeeded. The DNAs estimated by the DNA probes were not isolated one but the migration tendencies were differ from each other. It was clear that certain parts of the process in this study are advanced; that is, the precise monitoring of the electrophoresis process in space was accomplished, in real-time, by using 3DEP. In addition, the detection of DNA of recovered samples indicates that all the processes during the study were kept to be free from biological contamination. These results were published in following paper and reviewed in the book.

Papers published on this subject

1) Kobayashi, H., Ishii, N. and Nagaoka S. (1996) Bioprocessing in Microgravity : Free Flow Electrophoresis of *C. elegans* DNA, Journal of Biotechnology 47, 367-376.

Abstract : During the processes of the experiment in space, the house keeping data of the FFEU and the electrophoretic migration profiles were monitored at POCC (Payload Operations Control Center) of MSFC (Marshall Space Flight Center, Alabama) according to the real-time down-link system. The three dimensional electropherogram (3DEP) on the basis of the down-linked data showed some trouble of the apparatus but three sequential experiments indicated this disturbance of apparatus is rather preferably stable. Comparing post-flight analyses of the DNA component fractionated, that is, amplification by PCR method it revealed that the DNAs were separated approximately into two peaks: one of them contained seven fold higher than content of a DNA estimated by a *sod-4* gene probe than an *unc-6* gene probe. These results suggested that this separation technique could still more effectively for the separation of biological macromolecules such as DNA, and the efficiency of separation of the free flow electrophoresis under microgravity environment was useful.

2) Bauer, J., Hymer, W., Kobayashi, H., Morrison, D., Seaman, GVF, Weber, G., "Electrophoresis in Space" in Advances in Space Biology and Medicine Vol.7, eds. by Bonting, S. L. and Cogoli, A., published by JAI Press Inc., Greenwich, CT 06836-1678, USA, in press.

Fig. 1 The 3D electropherograms of the DNA sample which were containing adenosine and NADP as markers. The electropherograms, a: in space, b: on the ground, show around the fractions of sample markers in every 10 seconds.