【Grant-in-Aid for Scientific Research (S)】

Science and Engineering (Interdisciplinary Science and Engineering)

Title of Project : Microfluidic approach to single cell transcriptome analysis and its applications

Teruo Fujii
(The University of Tokyo, Institute of Industrial Science, Professor)

Research Project Number : 16H06328 Researcher Number : 30251474
Research Area : Interdisciplinary Science and Engineering
Keyword : Genomic engineering, Single cell analysis, Transciptome analysis

【Purpose and Background of the Research】

A population of cells either forming a tissue or being cultured has been analyzed so far as a group of homogeneous elements. It is, however, being revealed that those cells in a population show heterogeneous behaviors, as new analytical methods are rapidly becoming able to achieve measurements at the single-cell level. Recently, several attempts were made to understand cell-to-cell variation in gene expression levels, that is, single-cell transcriptome analysis based on next-generation sequencing (NGS) technologies. However, the number of cells analyzed at one time is highly limited because isolation and handling of large numbers of single cells is difficult with conventional methods. Here, we propose a novel microfluidic approach to drastically improve the throughput for single-cell transcriptome analysis, up to 10,000 cells per run, to comprehensively understand the cellular heterogeneity on the gene expression level.

【Research Methods】

We propose a novel method combining the electroactive microwell array (EMA), which has been developed by our group for parallelized analysis of large numbers of single-cells, with an advanced picoCAGE protocol that allows identify 5' ends of transcripts based on cap-analysis gene expression. To identify each cell, a different pair of index identifiers for the picoCAGE reaction will be spotted on an inner surface of each microwell by using microfluidic approaches.

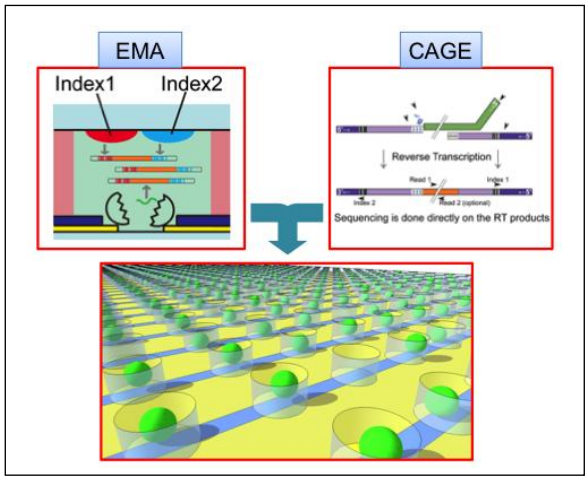

Figure 1 Single-cell transcriptome analysis

【Expected Research Achievements and Scientific Significance】

Large-scale single-cell analysis should be of interest for many biological studies and broadening the scope of biological understanding of cellular heterogeneity at the gene expression level. In this project, the present method is applied to the analysis of cervical cancer diagnosis and therapy. But it can be further extended to the analyses of biological and medical samples, such as rare cells, viable but not culturable microbes in various environments, drug resistant bacteria in early phase infection, etc., contributing to better understanding of environmental biodiversity as well as to the prevention of hospital-acquired infection.

【Publications Relevant to the Project】

Kim, S. H. and Fujii, T., "Efficient analysis of a small number of cancer cells at the single-cell level using electroactive double-well array," Lab on a Chip 16, pp. 2440 - 2449 (2016), selected as the outside front cover of the issue.
Plessy, C. et al., "Linking promoters to functional transcripts in small samples with nanoCAGE and CAGEscan", Nature Methods 7, pp. 528-534 (2010).

【Term of Project】 FY2016-2020

【Budget Allocation】 136,600 Thousand Yen

【Homepage Address and Other Contact Information】

http://www.microfluidics.iis.u-tokyo.ac.jp/
tfujii@iis.u-tokyo.ac.jp