SVII-1

GENETIC DIAGNOSIS: FROM BASICS TO CLINICAL MEDICINE

R. Yoshiyuki OASUMA

Department of Pathology, Tokai University School of Medicine, Bosei dai Isehara-city, Kanagawa, Japan 259-1193

Recent advances in genetic technologies have enabled us to make proper diagnosis of various diseased in order to entertain efficient treatment. Major chromosomal abnormalities include structural abnormalities (deletion, translocation) and numbers of autosomes (aneuploidy of autosomes and sex chromosomes). Single gene defects include autosomal dominant disorders, X-linked disorders, mutation in single gene defects (gene deletion, point mutation) and mitochondrial disorders. Genetic alterations for oncogenesis include (1) amplification, mutation and translocation of oncogenes, (2) deletion and mutation of suppressor genes, and (3) mutator genes. Recently, the Ministry of Health and Welfare of Japan has proposed clarification of etiologic gene abnormalities for major diseases such as cancers, diabetes mellitus, atherosclerosis, neurologic disease (such as Alzheimer disease). Especially, cancer therapy will focus on “tailor made therapy” based on gene analysis. In US, gene analysis has been of major commercial value for gene therapy. Histochemistry is expected to play an important role in genetic analyses at tissue and cell levels. The genetic technologies include (1) fluorescent in situ hybridization (FISH) for chromosomal abnormalities, (2) in situ hybridization (ISH), in situ PCR, and in situ RT-PCR for the changes of DNA and RNA, (3) enhanced immunohistochemistry and immunoelectron microscopy for the gene products (proteins). Instrumentation includes confocal laser scanning microscopy (CLSM), lader capture microdissection, DNA chip analyzer and staining automation. Among the gene based therapy for cancer, Herceptin is of special interests. It is immune based anti-cancer therapy by using humanized anti-HER2/neu monoclonal antibody. To select the effective cases with wide spread mammary carcinoma, IHC or FISH is used to analyze HER2/neu overexpression.

In summary, histochemical technologies are expected to play essential roles in gene analysis both at basic and clinical fields.

SVII-2

Laser Capture Microdissection

Kiyotetsu KOJIMA

Scientific Equipment Division, OLYMPUS JAPAN Co., Ltd.

Recently DNA/RNA analysis of specific tissue such as cancer tissue has been a popular technique. However, it is not so easy to obtain pure cell population without mixture of another cell type. Laser Capture Microdissection (LCM) provides research and pathology laboratories with the ideal microdissection technology. LCM was conceived and first developed as a prototype research tool at the National Institute of Child Health and Human Development (NICHD) and the National Cancer Institute (NCI) of the National Institutes of Health (NIH) (1). LCM is being used in the Cancer Genome Anatomy Project (CGAP) to catalog the development of cells from normal to diseased state. LCM is a technique that provides a rapid, reliable method to acquire pure populations of targeted cells from specific microscopic regions of tissue sections for subsequent analysis. In LCM, a transparent thermoplastic film affixed on the surface of a plastic cap, that has a function of a cap to microcentrifuge tube, is used to transfer the targeted cells. The film contacted on the sample tissue can be produced a thermoplastic bonding with cells of interest after activation by laser beam. We can choose three sizes of laser beam, 7.5, 15, 30 μm diameter respectively, depends on the targeted area. The 7.5 μm diameter of laser beam allows us to capture a specific single cell separately from another type of cells surrounding the target cells. Various analytical methods can be applied to the cells dissected by LCM (2-7), not only DNA/RNA but also proteins. LCM provides a useful method of the in vivo sample preparation for genomics and proteomics.

References

(3) Luo, Lin et al., Nature Medicine, Vol. 5, No. 1, January 1999
(4) Maitra, Anirban et al., Nature Medicine, Vol. 5, No. 4, April 1999
(5) Banks, Rosamonde E. et al., Electrophoresis, Vol. 20, 1999
(7) Banks, Rosamonde E. et al, Electrophoresis, Vol. 20, 1999