

Cytological Characteristics of Four Long-term Cultured Human Gastric Adenocarcinoma Cell Lines

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Examined were the properties of four CEA producing human gastric adenocarcinoma cell lines (HPE-GAC-T,-2,-3,-4) which were established by us and have been maintained for 7 to 11 years. Characteristic phenotypes of each of them were as follows: GAC-T; High alkaline phosphatase (ALP) activity. Inhibition profile and heat stability showed that it belonged to the isozyme of placenta type. Level of CEA production and immuno-stainability had been maintained. GAC-2; Formation of acinus-like aggregate and enzymocytochemical positive stainings of ATPase and ALP at apical portion of the structure. This means most differentiated structure had been phenotypically expressed in vitro. Low CEA production, but activation of its production by TPA showed the preservation of relevant genotype. GAC-3,-4; High beta-glucuronidase activity and lack of ALP activity.

The cytological varieties among cell lines seemed to be derived partly from the difference of differentiation status of original tumors and partly from change of phenotypes during long-term culture.

Analysis of cell kinetics based on multiparametric cytofluorometry and distribution patterns of tissue F-actin in the human colorectal cancers

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We analyzed cell kinetics and distribution patterns of tissue F-actin in the human colorectal cancers. First, single cells were separated and their DNA-RNA contents after acridine orange staining were measured using an epi-illumination multiparametric cytofluorometer (NIKON SPM-RFI-D). Non-cancerous colorectal mucosa examined as a control, was composed mostly of mononuclear diploid cells with a few cells in S-G₂ phases. Adenomas yielded almost the same results as the control, but the frequency of cells in S-G₂ phases was increased (10-20%). Intramucosal cancers and most of the mucinous carcinomas showed similar results to those of adenomas. In invasive cancers, various polyploid cells were observed in addition to a diploid cell population and the polyploidization progressed gradually in association with the tumor extension. Then, the distribution patterns of tissue F-actin were examined after NBD-phalloidin fluorescence staining. In controls and adenomas, F-actin was observed regularly at the cryptal surface, basement membranes and at cellular borders. But, in cancers, especially with less histological differentiation, F-actin was decreased markedly and irregularly.

These results demonstrate that, excluding mucinous carcinomas, cell kinetics was related to the tumor extension and the distribution patterns of tissue F-actin were related to histological differentiation of the tumors.

Intercellular Variations in the Cytochrome Oxidase Activities of Pulmonary Alveolar Epithelial Cells in Rats

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Significant variations were proven to exist among the cytochrome oxidase activities visualized cytochemically of Types I, II and III cells in the rat pulmonary alveolus. Lungs of male SPF Wistar rats were fixed for 10 min by tracheal instillations (under anesthesia) of a cold 2.5% glutaraldehyde-phosphate buffer solution. After washing for 60 min, 50 μ m-thick slices were incubated for 60 min at 37°C in a DAB medium containing 1.0 mg/ml 3,3'-diaminobenzidine-4HCl anhydride, 1 mg/ml cytochrome c, 0.1 mg/ml catalase, 7% sucrose and 0.1 M phosphate buffer at pH 7.4.

The mitochondrial intermembrane and cristalline spaces of Type II cells were filled with deposits. Reaction deposits had not fully accumulated within the spaces of Type I cells. The mitochondria of Type III cells were devoid of any significant product. The results suggest that Type III and Type I cells are rather deficient in cytochrome a.

Relationship Between Cell Kinetics and Production of HCG in Human Choriocarcinoma Cells in Vitro Treated with MTX

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MTX has a most effective value in the treatment of choriocarcinoma. In the case of follow-up and monitoring, levels of HCG is most important marker. We studied to clear the details of relationship between cell growth and HCG Production.

Human choriocarcinoma cells in vitro (ENAMI-1) were exposed to various concentrations of MTX for 48 hours, and then were harvested every 24 hours for 96 hours. Simultaneously number of viable cells were calculated and levels of HCG-B subunit in Medium were quantitated. Relative DNA content distribution for cell cycle analysis were measured from cell sample using flow cytometry.

The results were as follows, 1) In the ENAMI-1 cells, more than 10⁻⁷ M MTX inhibit cell growth and this effects were dose dependent. 2) In the contrast to cell growth, levels of HCG-B subunit raised remarkably at the administration of MTX. 3) In cell cycle analysis, more than 10⁻⁷ M MTX, the cells were accumulated in S-phase during administration of MTX. After the removal of 10⁻⁷ M MTX, the accumulated cells migrate toward the G₂+M phase. But in 10⁻⁶ M MTX the cells were not migrate toward the G₂+M phase.