Department of Anatomy (II)

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General Summary

We are engaged in teaching histology, embryology, endocrinology, and neuroanatomy to medical students. Our research projects have 5 focuses: histologic endocrinology, molecular biology of embryogenesis from embryonic stem (ES) cells, cell differentiation, the role of extracellular matrices in morphogenesis, and measurements with oxygen electrodes of respiration of cultured cells as a metabolic indicator for drug screening.

Research Activities

Establishment and characterization of a spontaneously cisplatin-resistant human gastric adenocarcinoma cell line

We established a cell line (IGSK-l) derived from a poorly differentiated adenocarcinoma of stomach. IGSK-1 cells grew as adhesive or floating cultures in the culture dishes. The population doubling time was about 34 hr. The cells were round shaped and had one nucleus. Electron micrographs of the cells showed many secretory granules and well-developed mitochondria. IGSK-l cells secreted gastrin and somatostatin. Although IGSK-1 cells showed immunoreactivity against serotonin, high performance liquid chromatography revealed the conditioned medium by IGSK-1 cells contained 3.7 ng/ml of serotonin, while cell-free medium contained 203.0 ng/ml of serotonin. These results suggested the IGSK-1 cells may reuptake serotonin in GM. The anti-cancer susceptibility test with an oxygen electrode-apparatus (DAIKIN, Dox-10, JPN) revealed the cells derived from the same adenocarcinoma were sensitive to TXL and not sensitive to CDDP, CPT-11 and 5-FU.

Establishment and characterization of a cisplatin-resistant cell line derived from ascitic fluid of recurrent mucinous and poorly differentiated adenocarcinoma of stomach

We successfully established a cisplatin-resistant adenocarcinoma cell line (IGSK-2) derived from ascitic fluid of a recurrent gastric cancer whose histopathological diagnosis was mucinous and poorly differentiated adenocarcinoma (mucinous>poorly) of stomach. IGSK-2 cells grew as adhesive cultures in the culture dishes. The population doubling time was about 83 hr. In xenotransplantation, no tumor mass was produced in host mice. Electron micrographs revealed the cells had one or two nuclei, well-developed Golgi apparati and numerous mitochondria. No mucous droplets were contained in the cytoplasm. The anti-cancer susceptibility test revealed the cells derived from the ascitic fluid were sensitive to TXL and not sensitive to CDDP, CPT-11 and 5-FU.

Establishment and characterization of human rhabdomyosarcoma cell line, HUMEMS, derived from primary embryonal rhabdomyosarcoma of the breast
The HUMEMS cell line was established from the pleural fluid of a 13-year-old girl who had embryonal rhabdomyosarcoma (alveolar type) of the breast. The HUMEMS grew as monolayer and consisted of two cytologic types of cells resembling those of the original tumor, spindle cells and large multinucleated cells. The multinucleated cells contained immunoreactive myoglobin. This cell line is very useful for studies on the susceptibility of anti-cancer drugs and analysis of the mechanism of metastasis.

Establishment and characterization of two human malignant mesothelioma cell lines (HMMME and HMMMF) from the same patient
Two cell lines designated HMMME (epithelial) and HMMMF (fibroblastic) were established from a pleural fluid from a Japanese male who suffered from cough, fever, dyspnea and fatigue. These cell lines were separated by the colonial techniques from the primary cultures, grew well without interruption for 5 years and characterized as producing hyaluronic acid. The HMMME cells were epithelial cell like and transplantable into the subcutis of nude mice, while the HMMMF line were fibroblast-like and transplantable into submucosa of Hamster's cheek pouches.

Establishment and biological characteristics of stem cell strains derived from human amniotic membrane
The amniotic membranes of full-term placentas obtained with the informed consent of patients were cultured and followed by cloning the small round cells that proliferated in multiple layers to obtain human amniotic membrane (HAM) stem cells. Forty amniotic membrane stem cell lines were established and these cell lines had common cellular biological properties. More specifically, the cells maintained a normal human karyotype, an undifferentiated state, and were alkaline phosphatase activity. They also expressed transcription factors Oct-4, Rex-1 and Ecat-4.

Induction of differentiation of human amniotic membrane stem cells in vitro
The multi-differentiation ability of human amniotic membrane stem (HAM-1) cells were assessed. The addition of 1) EGF, FGF2 and FGF9, 2) VEGF, or 3) FGF4 and HGF to the culture medium induced the differentiation of HAM-1 cells to 1) astrocytes and nerve cells, 2) endothelial cells and blood cells or 3) hepatocytes, respectively. Embryoid bodies formed by the hanging drop culture of EGFP gene-transfected HAM-1 cells mixed with ddY mouse EES-7 cells (ES cells) were, then, cultured stationary in a culture medium containing embryotrophic factors (ETFs). The bodies formed tridermal germ layer primordial organs (including skin, blood cells, neural tubes and digestive organs), and fluorescence of EGFP was confirmed in each of the organs. The transplanted HAM-1 cells formed immature teratomas in nude mice. These results suggested that HAM-1 cells were stem cells that have the ability to multi-differentiate into tridermal germ layer primordial organs, and had considerable expectations for application to regenerative medicine.

Publications

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