Research Center for Medical Sciences Radioisotope Research Facilities

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General Summary

The Radioisotope Research Facilities were established to support medical and biological research using radioisotopes. The Facilities also accept the research using nonradioactive isotopes. We have supported researchers by suggesting methods and practical techniques for experiments. Lectures and training courses are held for researchers and for medical students and graduate students. In 2018, the laboratory of this facility was used by 38 researchers from 12 departments and 12 students of 2 curriculums. Major nuclides used for experiments were ^{32}P, ^{51}Cr, ^{125}I, ^{14}C, and ^{3}H. Education related to radiation is also an interest.

Research Activities

Proteasome-resistant cells showed cancer stem cells by highly expressing CD44 and induced epithelial-mesenchymal transition

Down-regulation of E-cadherin plays an important role in epithelial-mesenchymal transition. Our previous study has demonstrated that suppression of E-cadherin induced expression of the transcriptional repressor zinc finger E-box-binding homeobox 1 (ZEB1) in endometrial carcinoma Ishikawa cells resistant to the proteasome inhibitor epoxomicin (EXM-resistant Ishikawa cells) established in our laboratory. Because we found that CD44, a cancer stem cell marker, was expressed in EXM-resistant Ishikawa cells but not in nonresistant Ishikawa cells, we studied the participation of CD44 in the suppression of E-cadherin expression in EXM-resistant Ishikawa cells. The CD44 expressed in EXM-resistant Ishikawa cells was identified as variant 4 (CD44v4). Overexpression of CD44v4 in Ishikawa cells by CD44v4/pcDNA3 transfection induced ZEB1 expression following E-cadherin suppression. On the other hand, CD44v4 suppression (knock-down) in EXM-resistant Ishikawa cells by antisense-CD44v4/pcDNA3 transfection induced E-cadherin expression and ZEB1 suppression. These results suggest that CD44v4 appears in EXM-resistant Ishikawa cells because ZEB1 is upregulated after E-cadherin is suppressed. Moreover, CD44v4 overexpression in Ishikawa cells acquired epoxomicin-resistance, and CD44v4 suppression in EXM-resistant Ishikawa cells repossessed epoxomicin-sensitivity. Epithelial-mesenchymal transition induction via extracellular signal-regulated kinase (ERK) 1/2 signal transduction has been observed previously. Therefore, we will further study CD44 involvement in epithelial-mesenchymal transition induction through ERK1/2 activation/phosphorylation.

Target chemotherapy against CD147-expressed carcinoma cells using polymeric micelles
Because a specific accumulation and cytotoxicity was observed in CD147-expressing cells treated with glutathione-doxorubicin conjugate-encapsulated anti-CD147 antibody-labeling micelles, we prepared tumor-bearing mice to investigate the *in vivo* chemotherapeutic effect.

Chemotherapy for drug-resistant cancer cells with curcumin
Curcumin, a component of turmeric, suppresses nuclear factor kappa B (NF-κB) activation by inhibiting the phosphorylation of inhibitor κB (IκB) bound to NF-κB and inhibiting IκB degradation by curcumin's proteasome inhibitory action. Furthermore, curcumin has been reported to exert an antitumor effect via NF-κB suppression against various types of cancer, including pancreatic cancer and lung cancer, in which KRAS proto-oncogene, GTPase (*KRAS*)-NF-κB is constitutively activated. In addition, colon cancer cell lines were shown to be sensitive to oxaliplatin, depending on the presence or absence of mutations of *KRAS* and *p53*, which do not affect the antitumor effect of curcumin. On the other hand, curcumin is considered to be a sensitive parent to EXM-resistant Ishikawa cells, an adriamycin-resistant, and a cisplatin-resistant ovarian cancer cell line A2780. Curcumin showed equal antitumor activity and the ability to be an effective therapeutic agent for cancers resistant to anticancer drugs.
In addition, curcumin has low bioavailability, and, free curcumin, when taken orally, is metabolized upon absorption from the intestinal tract to reduce its antitumor activity, and an sufficient antitumor effect cannot be obtained. Therefore, the newly developed water-soluble curcumin prodrug — curcumin monoglucuronide — has succeeded in achieving a blood concentration of free curcumin more than 1000 times that of conventional curcumin and is expected to be effective as a novel anticancer agent.

Analysis of resistance mechanisms in radiation-resistant organisms
Tardigrades, which are called water bears, can tolerate extreme environments, including ionizing radiation and dryness. The sludge water bear *Isohypsibius* was isolated from the activated sludge in the Morigasaki Water Reclamation Center, and the terrestrial water bear *Milnesium tardigradum* was isolated from moss collected in Tokyo's Minato Ward. To clarify the radiation-resistant mechanism, tardigrades were irradiated with X-ray at 500 Gy, and DNA damage was analyzed with the comet assay method.

Measuring and tracing of radioactive fallout in the environment
The distribution and behavior of radioactive fallout released into the environment by the accident of the Fukushima Daiichi Nuclear Power Plant in March 2011 have been investigated. Because contaminated water had been leaked into the ocean by accident, we recently examined a safe, simple, and rapid method of analyzing radioactive strontium in seawater. Radioactive strontium was separated with a column of cation exchange resin (Dowex 50WX8, Dow Chemical Company, Midland, MI, USA) and was measured with a newly developed plastic scintillator bottle and a liquid scintillation system (LSC-LB7, Hitachi Ltd.). With this method, the chemical separation of 10 hours (total, 2 days) could be evaluated and compared with 2 weeks via a conventional technique. The detection

limit in this procedure from 1 L of seawater was 0.02 Bq/L. This method might be able to be used to survey contaminated seawater.

Publications

Asakura T, Yokoyama M, Shiraishi K, Aoki K, Ohkawa K. Chemotherapeutic Effect of Anti-CD147 Antibody Labelled Micelles Encapsulating a Conjugate of Doxorubicin with Glutathione Targeting CD147-Expressing Carcinoma Cells. *Anticancer Res.* 2018; **38:** 1311-6.