



Technology Outline

Kyoto University Technology Showcase New York 2009

[Technology Presentation]

KJ-1: Novel “Peptide A” with More Potent Anxiolytic Activity than Diazepam

We found for the first time that low molecular weight peptide A dose-dependently exhibits anxiolytic-like activity after intraperitoneal or oral administration at a dose of 0.1-1 or 0.3-3 mg/kg, respectively, in the elevated plus-maze test in mice. The anxiolytic activity was more potent than that of diazepam, a general anxiolytic drug. Mixture of amino acids composing peptide A was inactive, and the minimum effective dose of retro-sequence peptide, was approximately 300 times higher than that of peptide A, suggesting that the amino acid sequence of peptide A is necessary for its potent anxiolytic activity.

KJ-2: New Method for Producing Alkaloid

A novel method for producing reticuline and related alkaloids is provided. Reticuline is the key intermediate for producing the isoquinoline alkaloids. Main isoquinoline alkaloids are morphine, codeine, papaverine, berberine and so-on. These alkaloids are used for the medicine. In this technology, reticuline and the related alkaloids, scoulerine and magnoflorine, are produced with the combination of the *Micrococcus luteus* and plant enzymes. Technology feature is the effective synthesis of stereo-specific reticuline and related alkaloids from dopamine as a culture substrate.

KJ-3: Use of Germline Stem Cells for Developmental Engineering

In this technology, a novel method of growing spermatogonial stem cells *in vitro* is provided. When spermatogonial stem cells, which are isolated from the testis of a neonatal mouse, are cultured in the presence of glial cell-derived neurotrophic factor (GDNF) and leukemia inhibitory factor (LIF), the cells can be grown up to 10^{14} fold in 5 months. The cells is designated as germline stem (GS) cells and confirmed that GS cells are capable of producing actual sperm, normal offspring and transgenic offspring. In addition to the GS cells, a method of cultivating multipotent germline cells has also been established from neonatal mouse testes.

KJ-4: Knockout Mice for Inflammatory Disease Researches

The TRPM2 knockout mice have been developed. TRPM2 is one of the transient receptor potential protein (TRP) family members and its gene encodes a plasma-membrane calcium ion channel. Recent researches suggest that TRPM2 plays important roles in inflammatory responses, and it is/may be related to rheumatoid arthritis, asthma, Alzheimer's disease, or other diseases caused by inflammation. These TRPM2 knockout mice are valuable model animal for drug discovery, screening, developing and/or mechanism researches of various inflammatory diseases. TRPM2 knockout mice will be provided as research materials.

KJ-5: Method for Introducing Nucleic Acid into Cells

This invention facilitates the high-throughput analysis of gene functions. In this technology, gene expression constructs or siRNAs are arrayed on an electrode, and mammalian cells are cultured on the array. At desired moment, electric pulses are applied to the system to allow for parallel introduction of the nucleic acids into the cells. Specific phenotypes caused by overexpression or silencing of genes provide biological readouts from which we can infer the functions of particular genes. What is unique with our technology is that it performs surface-mediated electroporation, which enables us to transfect cells in highly efficient and temporally controlled manners.

KJ-6: Library of More than 20,000 Marine Microorganisms found in Nagasaki

A library of more than 20,000 marine microorganisms around Nagasaki of West Japan (bacteria: ca.17,000, streptomycetes: ca.3,000 and yeasts/molds: ca.200) have been established. Following are some examples of microorganisms:

- Restraint of fat cell splintering (15 cell strains)
- Suppressed telomerase (6 cell strains)
- Increased production yield of highly-unsaturated fatty acid such as EPA and DHA (3 cell strains)
- Suppressed melanin synthesis (2 cell strains)

Marine Microorganisms around Nagasaki will be provided as research materials.

KJ-7: Novel Contrast Agent for Cancer in Magnetic Resonance Imaging (MRI)

A newly designed gadolinium complex is effectively encapsulated (concentrated) into apoferritin cavity. The method of encapsulation of the gadolinium complex into apoferritin is improved significantly compared with the conventional methods of reconstitution. The relaxivity, a parameter for indicating the contrast, of gadolinium in apoferritin cavity is 3-10 times higher than

that of commercially available MRI contrast agent, Gd-DOTA . The relaxivity depends on the loading concentration. With chemical modification of apoferritin with hydrophilic polymers such as dextran and poly (ethylene glycol), a stealth ability from reticuloendothelial system is endowed. The novel contrast agent is effective for cancer imaging of tumor-bearing mouse.

KJ-8: Novel Iron-based Cross-Coupling Catalyst System for Synthesis of Aromatic Compounds

Novel Iron-based Cross-Coupling Catalyst System for Synthesis of Aromatic Compounds Recent advances in cross-coupling method have been making a significant contribution to the synthesis of the intermediates of medicines. The newly developed iron-based catalyst system is significantly more effective in the cross-coupling reactions than conventional process. In this method, a variety of alkyl halides and arylmetal reagents are cross-coupled with more than 90% yields. This catalyst system is more cost-effective, less toxic and easy-to-handle. As a result, this method for synthesis of aromatic compounds is expected to be used as a potent alternative to the existing Suzuki-Miyaura boron-based cross-coupling technology with an expensive rare metal catalyst such as palladium and nickel.

KJ-9: Method of Stabilizing Calcium Phosphate Fine Particles Applicable to Fabrication of Hydroxyapatite Microcapsule for Drug Delivery System (DDS)

This novel technology relates to a method for stabilizing calcium phosphate fine particles formed from Simulated Body Fluid (SBF) whose ionic concentrations are similar to those of human blood plasma. These fine particles have high activity for forming hydroxyapatite (HAp) from body fluid and are named Apatite Nuclei. The Apatite Nuclei are precipitated from SBF by raising pH or temperature and then the growth is halted by lowering the inorganic concentration surrounding them. The Apatite Nuclei can be preserved for long periods, while maintaining its high activity for forming HAp. Microspheres of metals, ceramics and polymers are easily encapsulated with HAp by using the Apatite Nuclei. The resultant microcapsules are biocompatible, show sustained release and are expected to be applied to one of ideal carriers for DDS.

[Poster Presentation]

KJ-10: Bioactive Apatite Nuclei Precipitated Titanium with Superior Osteo-Conductivity for Implants

This technology relates to a fabrication method of a new material of titanium (Ti) which

demonstrates extremely superior osteo-conductivity *in vivo*. Simulated Body Fluid (SBF) whose ionic concentrations are similar to those of human blood plasma, precipitates calcium phosphate fine particles having high activity for forming hydroxyapatite (HAp) from body fluid. On Ti plate surface, many micro pores are formed by H₂SO₄ treatment, in which the Apatite Nuclei are precipitated. The Apatite Neuclei functions as the nuclei of the crystal growth of HAp effectively from body fluid. In an animal test, this material indicated much faster and stronger bonding to a bone than either simple porous Ti plate or HAp-coated porous Ti fabricated by a conventional method. Also, this material has the same mechanical strength and machining performance as commercially available Ti. As a result, this material is expected to be significantly useful for implants, e.g. total hip replacement, dental implant, etc. This technology is able to be applied to not only Ti but any kind of metals, ceramics, and polymers.

KJ-11: Novel Device Restoring Lung Elastic Recoil

Chronic obstructive pulmonary disease (COPD) is a progressive disorder with poor prognosis and expected to be the third leading cause of death by 2020. There is no effective treatment to improve the prognosis of the patients for present. COPD is characterized by the airway flow limitation and loss of elastic recoil of the lung. A novel elastic net, named the Lung Net, which covers the lung to restore the elastic recoil, is designed.

KJ-12: Novel Visualization for Large Scale Multivariate in Bioactive Chemicals

This technology relates to the computer program which efficiently displays both primary characteristics and secondary characteristics of a plurality of compounds for association analysis, in which even four or more characteristics can be handled simultaneously as the secondary characteristics. Sample software is available.

KJ-13: A Novel Computational Approach for Drug Discovery: Chemical Genomics-based in Silico Ligand Screening

“Chemical-genomics-based drug design (CGBDD)” is a novel approach to predict bioactive compounds based on machine learning of protein-compound interaction patterns (chemical-genomics knowledge). The technique has the following advantages:

1. No need for three-dimensional protein structures
2. Achieve higher hit rate: more than 10% (Conventional methods: 1%)
3. Discover novel scaffolds
4. Reduce computational costs: 2 or 3 weeks calculation time (Conventional methods: 3 months)

CGBDD is a promising approach to accelerating drug discovery processes.

KJ-14: Novel Transporting Case for Regenerative Medicine

Regenerative medicine using cell transplantation is a hopeful and promising treatment for intractable diseases. To obtain a good clinical performance, transporting living cells in a good condition is important. We have developed a novel transporting case, which has temperature-conditioning and anti-shock structure. This case can transport not only living cells but also living organs. As a result, our new transporting case promotes the development of both cell transplantation and organ transplantation.

