Core Research Facilities

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General Summary

The Research Center for Medical Sciences was reorganized as the Core Research Facilities, which consist of the Divisions of Fine Morphology, Biochemistry, and Advanced-Research Laboratory. The mission of the facilities is the facilitation of research in the university. Two systems are constituted for use of the facilities.

Annual Registration System
This system is intended for researchers in the university to supply research space, benches, and other equipment to perform experiments. Once registered, researchers can freely use the various devices, such as fluorescent microscopes, optical microscopes, and equipment to prepare specimens for histological examinations, high-performance liquid chromatography is used, and nucleic acid amplification system (polymerase chain reaction). Because inspections and maintenance are regularly performed by the staff, equipment is in excellent condition and available at any time. In addition to providing space, devices, and equipment, this system can provide technical advice and guidance on specific fine-morphological or biochemical approaches to registrant's experiments.

Research Service-Providing System
Advances in research technologies and hardware enables us to perform more detailed and accurate observations of specimens in medical sciences. However, various technologically highly-advanced devices require specialized knowledge. These advances sometimes cost researchers both time and money. Also, all researchers are not necessarily familiar with all the equipment available for medical experimentation. For researchers who cannot perform experiments because of limitations of time and resources, our staff can prepare specimens for scanning electron microscopy and transmission electron microscopy, record images, or perform high-performance liquid chromatography and mass spectrometry for analysis. By the use of this system, researchers can proceed efficiently. The service fee is minimal because services are limited to the university.

Research Activities

Conjugation of fluorescent nanoparticles to monoclonal antibodies
Fluorescent nanoparticles have been used as tracers for the visualization, qualification, and quantification of biological molecules. We conjugated particles to the monoclonal antibody JT95 to investigate thyroid papillary carcinoma. The antibody recognizes antigens of gylcosylated fibronectin produced by thyroid carcinoma and can be used as a biomarker for detecting the tumor. The antibody-nanoparticle conjugate is also useful for

evaluating tumor progression or recurrence after surgery. Several methods for conjugation were tested, and we optimized the conditions.

Functional analysis of tight junctions

Tight junctions (TJs) among adjacent epithelial cells control paracellular permeability of solutes. Epidermal TJs are believed to restrict molecular movement to assist the stratum corneum as a secondary barrier in the skin. However, the role of TJs in molecular distribution in the epidermis has not been thoroughly studied. Calcium ions (Ca^{2+}), which induce keratinocyte differentiation, are distributed in a vertical gradient peaking in the stratum granulosum. In this study, we applied sodium caprate (C10), which elicits dilation of TJs on human reconstructed epidermis, and investigated Ca^{2+} distribution in the epidermis. The localization of Ca^{2+} in the epidermis was observed with ion-capture cytochemistry and electron energy-loss spectroscopy with a transmission electron microscope. After treatment with C10, the epidermal Ca^{2+} localization was altered compared with that in untreated epidermis. Precipitates containing Ca^{2+} appeared in the intracellular and extracellular spaces of the stratum corneum, and large clusters of these precipitates were occasionally observed in the stratum corneum and the stratum granulosum. Additionally, abnormal differentiation (e.g., parakeratosis) was observed in the stratum granulosum. To confirm that these changes were caused by TJ disruption, we observed the structure of TJ strands with the freeze-fracture replica method and measured transepidermal Ca^{2+} permeability by quantifying diffused Ca^{2+} through the epidermis. We found that the TJ strands were fragmented and that the Ca^{2+} permeability had increased. These findings suggest that epidermal TJs maintain Ca^{2+} under the stratum corneum and regulate epidermal differentiation.

Adhesion and structural properties of protein nanomaterials containing hydrophobic and charged amino acids

Protein polymers are being used or considered for biologically based adhesives and coating materials. Most adhesives derived from macroprotein molecules work through receptors or cross-links. Clarifying the adhesion mechanism of protein polymers would lead to a better understanding of adhesives and the discovery of new practical properties of protein polymers at both the nanoscopic and macroscopic levels. The objective of this research project was to study the adhesion properties of protein polymers at the nanoscopic level. Seven protein nanomaterial samples with different degrees of adhesive strength were designed and synthesized using solid phase chemistry. All protein nanomaterials contain a common hydrophobic core flanked by charged amino acid sequences. The adhesion properties of the protein nanomaterials were investigated at different pH values and curing temperatures. The protein nanomaterials self-aggregate and interact with a wood surface. The protein nanomaterial KKK-FLIVIGSII-KKK identified in this study had high adhesive strength toward wood. It had the highest shear strength at pH 12, with an amino acid sequence that was extremely hydrophobic and uncharged. This protein nanomaterial was subjected to structural analyses using circular dichroism, laser-Fourier transform infrared spectroscopy, and matrix-assisted laser desorption ionized mass spectrometry. At pH 12 this peptide adopted a pH-induced

beta-sheet-like conformation. Adhesive strength reflects contributions of both hydrogen bonding and van der Waals interactions. Our findings suggest that ionic and covalent bonds are not significant factors in adhesion.

Publications

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