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Study on the development of a novel fluorescent linker molecule for both structural analysis and immobilization, as well as its application to the discovery of sugar-chain markers specific for adult T-cell leukemia (ATL) cell surface

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Chapter 1

In this thesis, I described the study on the development of a novel fluorescent linker molecule for both structural analysis and immobilization, as well as its application to the discovery of sugar-chain markers specific for adult T-cell leukemia (ATL) cell surface.

HTLV-I is an oncogenic retrovirus that is associated with adult T-cell leukemia (ATL) that is distributed in southern Kyushu. In Japan, new ATL patients are appeared more than 2000 people every year, and about 1000 people die by ATL. Although many researches of ATL have paid attention to tax gene that HTLV-1 has, effective therapy has not been established yet.

As a first step for an altenative ATL antibody therapy, we have tried to find antigenic sugar-chain makers on T cell surface infected by HTLV-1. A chip technology is one of promising methods for the screening of antibodies. To date, Suda et al. reported “sugar chip”, in which sugar-chains are immobilized on SPR sensor chip using their original linker molecules. Even though using the original linker molecules, they faced a difficulty since the purification of a conjugate with sugar-chain was impossible for less than 1 mg of sugar chain. To overcome this problem, I developed first a novel fluorescent linker molecule (named as f-mono) for both immobilization and structural analysis, characterized it by preparing the conjugate with simple disaccharides, and confirmed its application for the analysis of N-glycan of human immunoglobulin. Then, using this novel linker molecule, I have done the differential study for two leukemia cells to find the difference in the sugar-chains on their cell surfaces.

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Several sugar-chains were so far found for the possible candidates. Some of them are being immobilized on the chip and used for the screening.

Chapter 2

Syntheses of f-mono linker and two fluorescent ligand-conjugate (Galβ1-4Glc-fmono and Glcα1-4Glc-fmono) was described.

Chapter 3

The characterization of the fmono linker and FLCs using, Fluorescence spectrum, SPR, MS, MS/MS was described.

Chapter 4

The application of f-mono linker molecule to the analysis of N-glycans of human immunoglobulin G was described. The results were compared with a commercial conventional “blott-glyco” method.

Chapter 5

An differential study to find sugar-chain markers on the surface of leukemia cells was performed.

Chapter 6

This study were summarized.