【Grant-in-Aid for Scientific Research (S)】

Broad Section H

Title of Project : Analysis of regulation of intestinal homeostasis by glycan

TAKEDA Kiyoshi
(Osaka University, Graduate School of Medicine, Professor)

Research Project Number : 21H05043 Researcher Number : 20309446
Term of Project : FY2021-2025 Budget Allocation : 145,600 Thousand Yen
Keyword : intestine, glycosyltransferase, microbiota, mucosal immunity

【Purpose and Background of the Research】

Inflammatory bowel diseases (IBDs), such as Crohn's disease and ulcerative colitis, are intractable diseases. The mucosal immune responses against intestinal microbiota cause development of IBD. Therefore, it is necessary to clarify the mechanism by which the mucosal immune system does not recognize intestinal microbiota under healthy conditions.

There is a single layer of intestinal epithelial cells between the mucosal immune system and microbiota. We have shown that the intestinal epithelial cells have a barrier function that separates the mucosal immune system from the intestinal microbiota. It has been reported that the barrier function of the intestinal epithelium is mediated by a mechanical barrier constructed by the mucus produced by the epithelium, which is mainly composed of mucin. In addition, the glycoprotein Ly6/PLAUR domain containing 8 (Lypd8), which is specifically expressed in the epithelium, is secreted into the lumen. We have shown that Lypd8, a glycoprotein secreted into the lumen, associates with the flagella of highly motile bacteria and suppresses their motility, thereby inhibiting their entry into the intestinal tissues and maintaining intestinal homeostasis. Furthermore, we have shown that this expression is drastically reduced in patients with ulcerative colitis.

Lypd8 is an extremely glycosylated protein. Muc2 mucin, the main component of mucus in the colon, is also a highly glycosylated protein. Muc2-deficient mice are as susceptible to intestinal inflammation as Lypd8-deficient mice, suggesting the importance of both glycoproteins in maintaining intestinal homeostasis. However, the significance of glycosylation in maintaining intestinal homeostasis is not fully understood. It is well known that genetic abnormalities in glycosyltransferases are associated with a variety of diseases, coupled with the fact that glycans are crucial for higher-order functions of proteins. However, the function of glycosyltransferases in maintaining intestinal homeostasis is poorly understood. Furthermore, it is not known which glycan structures are involved in intestinal homeostasis.

In this study, we will focus on glycosylation, one of the post-translational modifications, and clarify the physiological functions of glycans themselves in terms of the mechanism of intestinal homeostasis.

【Research Methods】

In order to clarify the role of glycosylation in maintaining intestinal homeostasis, we will analyze the function of glycosyltransferases in the intestine as well as the function of the glycan structures modified by these enzymes, mainly targeting B3galt5, Chst4, St6galnac6, and B3gnt7, according to the following analyses.

·Susceptibility of KO mice to intestinal inflammation.
·Intestinal microbiota and mucosal immunity of KO mice.
·Glycan structures in intestinal tissues of KO mice.
·Function of target glycans in maintaining intestinal homeostasis.
·Significance of missense mutation of glycosyltransferase gene in IBD patients
·Glycan structures in intestinal tissues of IBD patients.

Through these analyses, we will clarify the roles of glycosyltransferases and glycan structures themselves in maintaining intestinal homeostasis, and clarify the mechanism of IBD pathogenesis caused by their disruption.

【Expected Research Achievements and Scientific Significance】

The analysis of the mechanism of intestinal homeostasis has so far focused on the function of a single molecule. In this study, we focused on glycosylation, one of the post-translational modifications, and analyzed the physiological functions of glycans themselves from the viewpoint of the intestinal homeostasis mechanism. In other words, we believe that this study will be the first in the world to clarify the mechanism of intestinal homeostasis by glycans. Furthermore, by clarifying the role of IBD susceptibility genes, which have been identified as rare variants, in the maintenance of intestinal homeostasis as a pathogenic mechanism of ulcerative colitis through these series of analyses, it will be possible to demonstrate for the first time in the world that ulcerative colitis is caused by abnormalities in intestinal bacteria based on abnormalities in epithelial barrier function involving many genes. In this way, we will be able to propose for the first time in the world that ulcerative colitis is caused by abnormalities in intestinal bacteria based on abnormalities in epithelial barrier function involving many genes.

【Publications Relevant to the Project】

· Kayama H, Okumura R, Takeda K: Interaction between the microbiota, epithelia, and immune cells in the intestine. *Annu. Rev. Immunol.* 38, 23-48 (2020).
· Okumura R, Takeda K et al: Lypd8 promotes the segregation of flagellated microbiota and colonic epithelia. *Nature* 532, 117-121 (2016).

【Homepage Address and Other Contact Information】

http://www.ifrec.osaka-u.ac.jp/en/laboratory/kiyoshi_takeda/