Butyrophilin and Butyrophilin-like genes and their role in epithelial cell-intraepithelial T lymphocyte cross-talk

AKADEMISK AVHANDLING

som för avläggande av medicine doktorexamen vid Sahlgrenska akademin
vid Göteborgs universitet kommer att offentligen försvaras i hörsal
Ivan Ivarsson, Medicinaregatan 3, Göteborg

Torsdagen den 2 juni 2016, kl 09.00

av

Cristina Lebrero Fernández

Fakultetsopponent
Associate Professor **Bruno Silva-Santos**, PhD
Instituto de Medicina Molecular, Faculdade de Medicina
Univesidade de Lisboa, Lisboa, Portugal

Avhandlingen baseras på följande arbeten:

I. Cristina Lebrero-Fernández, Joakim H. Bergström*, Thaher Pelaseyed* and Anna Bas-Forsberg.
Murine Butyrophilin-like 1 and Btnl6 form heteromeric complexes in small intestinal epithelial cells and promote proliferation of local T lymphocytes.
Front Immunol. 2016 Jan 19; 7: 1. doi: 10.3389/fimmu.2016.00001

II. Cristina Lebrero-Fernández and Anna Bas-Forsberg.
The ontogeny of Butyrophilin-like (Btnl) 1 and Btnl6 in murine small intestine.
Submitted for publication

III. Cristina Lebrero-Fernández, Thaher Pelaseyed and Anna Bas-Forsberg.
Butyrophilin-like (Btnl) 4 forms heteromeric intra-family complexes and its expression is delayed in the intestine during ontogeny.
Manuscript

IV. Cristina Lebrero-Fernández, Ulf Alexander Wenzel, Paulina Akeus, Ying Wang, Hans Strid, Magnus Simrén, Bengt Gustavsson, Lars G. Börjesson, Susanna L. Cardell, Lena Öhman*, Marianne Quiding-Järbrink* and Anna Bas-Forsberg.
Altered expression of Butyrophilin (*BTN*) and BTN-like (*BTNL*) genes in intestinal inflammation and colon cancer.
Immun Inflamm Dis. 2016 April 1. doi: 10.1002/iid3.105

* The authors contributed equally to this study

UNIVERSITY OF GOTHENBURG

Gothenburg 2016

Butyrophilin and Butyrophilin-like genes and their role in epithelial cell-intraepithelial T lymphocyte cross-talk

Cristina Lebrero Fernández
Department of Microbiology and Immunology, Institute of Biomedicine
Sahlgrenska Academy at University of Gothenburg
Gothenburg, Sweden

ABSTRACT

More than 50% of our immune system is located in the gut. The intestinal epithelium, which forms an interface between the organism and the environment, harbors intraepithelial lymphocytes (IELs) that comprise a mixture of conventional αβ T cells and unconventional αβ- and γδ T cells. IELs play important roles in regulation of gut epithelial integrity and in recognition of stressed and infected epithelial cells, and thus, are critical effector components of mucosal immunity. However, the understanding of the IEL function and their interaction with the neighboring epithelial cells is still limited. The aim of this thesis was to investigate how the Butyrophilin (Btn) and Butyrophilin-like (Btnl) molecules are involved in the epithelial cell – IEL cross-talk and hence, to characterize their role in regulating local T cell mediated immune responses in the intestinal mucosa.

Btn and Btnl proteins have over the past decade emerged as novel regulators of T cell functions both in periphery and locally in the tissue, and have been shown to be genetically associated with various inflammatory and proliferative disorders. We have reported the ability of intestinal epithelial cell (iEC)-specific Btnl proteins to induce IEL activation and proliferation in conditions without exogenous stimulation, which may contribute to the upkeep of the intestinal IEL pool. We have furthermore identified novel intestinal epithelial cell expressed Btnl- heteromeric protein complexes, and demonstrated that one of them, the Btnl1-Btnl6 heteromeric complex, specifically enhances the expansion of intestinal IELs bearing the Vγ7Vδ4 receptor *in vitro*. We have additionally explored how iEC-specific Btnl proteins are regulated in the neonatal murine small intestine and found that Btnl- protein expression is delayed in the ontogeny and that the expression of the *Btnl* genes is regulated on post-transcriptional level. Our data demonstrate that the proteins are not detectable in the small intestinal epithelium of mice before 3 weeks of age, and that the appearance of Btnl1 and Btnl6 proteins correlates with the expansion of intestinal Vγ7Vδ4 IELs, further adding strength to our *in vitro* results. Since γδ IELs are essential for the maintenance of the homeostasis in the gut, our findings suggest that Btnl proteins have implications in the intestinal immune response. To increase the understanding of the Btn and Btnl molecules' role in intestinal disorders, we have characterized the expression of human and mouse Btn and Btnl genes in colonic inflammation and intestinal tumors. Our results show an altered expression of the *BTN* and *BTNL* genes in these diseases and indicate an association between Btn and Btnl genes and ulcerative colitis and colon cancer.

In summary, this thesis work has demonstrated that iEC-specific Btnl proteins can regulate the function of intestinal intraepithelial lymphocytes in the gut, and that Btn and Btnl genes are associated with bowel pathology. Nonetheless, further studies are necessary to identify the complete immunomodulatory implication of the Btn and Btnl family members in healthy and inflamed/infected gut mucosa.

Keywords: butyrophilin-like, butyrophilin, intraepithelial lymphocytes, mucosal immunity, intestinal epithelial cells, γδ T cells, intestinal inflammation, colon cancer, ulcerative colitis.

ISBN: 978-91-628-9756-7 (print) **ISBN**: 978-91-628-9757-4 (e-pub)