

Travel Report for IUIS VIC PhD/DVM Student Travel Award to attend the Keystone Symposium on "Immunity to Veterinary Pathogens: Informing Vaccine Development " to be held at Keystone Conference Center in Colorado, 20- 25 January, 2015

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5. Travel Report:

I am a PhD student in Graduate School of Veterinary Medicine at Hokkaido University in Japan, and study on bovine immunology of chronic infections. Thanks to the support from this travel award, I traveled to the U.S. and attended the Keystone Symposium on "Immunity to Veterinary Pathogens: Informing Vaccine Development " which was held at Keystone Resort in Colorado during 20-25 January, 2015. This report summaries my activities, interests and experiences during the Keystone Symposium.



Snap 1. Keystone Resort



Snap 2. Keystone Conference Center

In order to resolve intractable infectious diseases in the veterinary field, more various approaches from the both aspects of immunology and microbiology are required. This Keystone Symposium is the first meeting of its kind for veterinary immunology and the precious opportunity to share the novel information about the individual research projects with each other. In my country, Japan, researcher and conference in veterinary immunology are very limited, so attending the symposium was an invaluable opportunity for me to gain knowledge for my PhD research, get to know other researchers and facilitate research collaborations and career development.

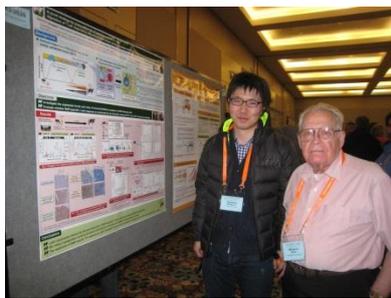
Fortunately, I had an opportunity to share my recent research entitled "A novel strategy for immunotherapy of paratuberculosis in cattle: regulation of T-cell

response to *Mycobacterium avium* subsp. *paratuberculosis* by the blockade of immunoinhibitory receptors” in both a short-talk session (at Workshop 1) and a poster session. I was so happy that many participants were interested in my talk and poster, gave me suggestions and comments, and most importantly, got to know each other.

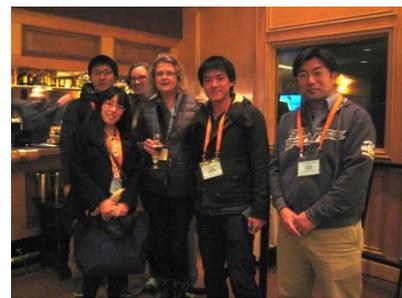
In my PhD research, I focus on the question: how and to what extent T-cell response is impaired in chronic infections of cattle. My previous studies have shown that the immunoinhibitory receptors, such as programmed death-1 (PD-1) and lymphocyte activation gene-3 (LAG-3), are upregulated on CD4⁺ and CD8⁺ T cells in cattle chronically infected with bovine leukemia virus (BLV), paratuberculosis and tuberculosis. As a result of the inhibitory signals through PD-1 and LAG-3, T cells are suppressed their effector functions and show the unresponsiveness to antigens of the pathogens, so-called “T-cell exhaustion”. More interestingly, these exhausted T cells are reactivated and restored the effector functions by interrupting the bindings between the inhibitory receptors and their ligands. Therefore, PD-1 and LAG-3 could be novel immunotherapeutic targets to enhance T-cell functions and resolve chronic infections in cattle.



Snap 3. Short Talk Session



*Snap 4. Poster Session
(with Dr. Davis)*



*Snap 5. In a bar
(with Dr. Brown and colleagues)*

During the 5 days of the meeting, there is so much scientific excitement for me. I was particularly very interested in 3 topics: 1) analysis of antigen-specific T cell responses using MHC tetramer, 2) antigen mining with new technologies for vaccine development, 3) active research and progress in the role of gamma delta ($\gamma\delta$) T cells.

1) Analysis of antigen-specific T cell responses using MHC tetramer

To understand kinetics of immune response deeply during infectious diseases, it is essential to analyze antigen-specific T cells. As everyone knows, MHC tetramer is the best way to detect antigen-specific T cells. This is not so new technology and many types of MHC-tetramer targeting various pathogens are commercially available in human and mouse immunology. In contrast, in veterinary field, MHC-tetramer technology is difficult to be applied to research because of genetic diversity of MHC genes of veterinary animals and lacking information on optimal antigens and epitopes. Although classical assays, such as T-cell proliferation, cytokine measurement and ELISPOT followed by antigen stimulation, are useful to investigate antigen-responding functional T cells, it is more difficult to track specific T cell responses using them when T cells are exhausted, in other words suppressed, during chronic infections. In my PhD research, I study T cell responses in BLV-

infection and paratuberculosis of cattle, and also have the same problem. So, in order to solve the problem, I would like to introduce MHC-tetramer to my experiment in the future.

In this meeting, 4 speakers introduced their researches on specific-T cells by MHC tetramer staining targeting *Theileria parva* and *Anaplasma marginale* in cattle and foot-and-mouth disease virus in pig. MHC-tetramer technology has been in the spotlight even in the veterinary field. It enables us to monitor T-cell responses to veterinary pathogens precisely and contributes to future vaccine developments.

2) Antigen mining with a new technology for vaccine development

The aggressive use of vaccines has been central for the control and eradication of several infectious diseases in the veterinary field. In spite of recent progress in research on immunology and vaccinology, there still remain many infectious diseases without effective vaccine. Therefore, a novel strategy for vaccine development is strongly required. Selecting ideal vaccine antigens is responsible for the development of effective vaccine. In this meeting, new technologies for antigen mining was one of big issues. In particular, I was interested in *in silico* identification of CTL epitope by estimating peptide-MHCI binding affinity. Recent progress in computer algorithm makes it possible to identify the most immunogenic epitope accurately without wet experiments. The much amount of time and labor are required for the epitope identification by classical methods, but *in silico* method can identify them with minimum steps. Therefore, I think this technology will accelerate the novel vaccine development.

3) Active research and progress in the role of gamma delta ($\gamma\delta$) T cells

My other interest is comparative immunology. Veterinary immunology is a very unique field because we pay attention to several animals with different immune systems. In terms of mammals, the high frequency of $\gamma\delta$ T cells in ruminants and pigs is one of the big questions. Although $\gamma\delta$ T cells are infrequent (0.5-10%) in human and mice, circulating $\gamma\delta$ T cells are abundant in cattle (20-60%) and pigs (30%). They should have important roles in the immune system of these animals, but the whole picture has not been clarified. I am working on bovine T cell immunology, so this issue has attracted my interest. This meeting had a session highlighting to the role of $\gamma\delta$ T cells. In this session, I was particularly interested in two presentations. Dr. J.C. Telfer introduced their unique research entitled "WC1 Is a Hybrid $\gamma\delta$ TCR Coreceptor and Pattern Recognition Receptor for Pathogenic Bacteria". It was a big surprise to me that WC1 directly recognizes bacteria, such as *Leptospira* spp., *Borrelia burgdorferi* and *Mycobacteria bovis*. Thus, they acts as not only TCR coreceptors but pattern recognition receptor and possibly antimicrobials. The other interesting research was presented by Dr. E. Guzman, which was entitled "Bovine gamma delta T Cells Are a Major Regulatory T Cell Subset". It is the common knowledge that regulatory T cells (Treg) is a critical population to balance immune response and inflammation. However, their previous findings support that $\gamma\delta$ T cells are also involved in immune suppression in cattle. In the present study, they showed that bovine $\gamma\delta$ T cells have the ability to produce much amount of interleukin-10 and suppress antigen-specific and nonspecific proliferation of T cells, which is licensed

by the contact of antigen-presenting cells. These studies look like quite original and cause a new sensation. Besides them, studies on $\gamma\delta$ T cells in cattle and pig can make outstanding findings from veterinary immunology.

I enjoyed the scientific program of the Keystone Meeting and was inspired so much. In addition, the program was well-prepared in the aspects of communication and recreation. The meeting had common breakfast, coffee break and social evening hour, so it is enough to get to know each other and talk about anything. I am sure that the friendship established in keystone will help me to develop my future collaboration and carrier. In addition, we had 4-hr afternoon breaks for recreation on day 3 and 5 and I joined snow-shoe trekking program. In this program, we went up to the mountain of Keystone Ski Resort by gondolas and walked around forests. I enjoyed this nice exercise and great view of Rocky Mountain. I hope I will go back to Keystone to join and enjoy the scientific program and the great nature.

Finally, I am deeply grateful for the financial support by IUIS-VIC bursary which enabled me to attend this wonderful meeting. I would like to express the deepest gratitude to Dr. Gary Entrican and Dr. Joan Lunney in IUIS-VIC comitee.



Snap 6. Snow-shoe Trekking



Snap 7. The Great Peaks