The 2nd Symposium of International
Immunological Memory and Vaccine Forum (IIMVF)

Trends in Immunological Memory and Vaccine Development

August 25-26, 2014

La Jolla Institute of Allergy & Immunology
La Jolla, California, U.S.A.

Organized by

International Immunological Memory and Vaccine Forum
US-Japan Cooperative Medical Sciences Program on Immunology
La Jolla Institute for Allergy & Immunology
Leading Graduate School at Chiba University
Chiba Innovation Program for Therapeutics at Chiba University

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Welcome Message

We would like to welcome you to the 2nd symposium of “International Immunological Memory and Vaccine Forum (IIMVF)”. The 1st symposium was held in Tokyo in January 2013, in which Drs. Rafi Ahmed, Andreas Radbruch and Stephen Schoenberger, who are international advisors for our IIMVF gave keynote and special lectures. We heard about the cutting edge research being conducted in all of our core members’ laboratories. During the meeting, we discussed the outline and the venue of the 2nd symposium, and Drs. Stephen Schoenberger and Mitchell Kronenberg kindly accepted to host the 2nd symposium at LIAI, CA, USA. The theme of this symposium is “Trends in Immunological Memory and Vaccine Development”. About 20 Japanese senior and junior speakers and 8 US invited and 5 young local speakers will present their recent work in the symposium. We have also many poster presentations. More than forty Japanese scientists and students attend this meeting. We appreciate all of the advisory and core members, and the meeting participants of this forum for their active contribution to the symposium. We do hope all attendants enjoy presentations, questions and active scientific discussion on immunological memory and vaccine research. The aim of the IIMVF is shown below for the purpose of sharing the mission and value of the memory and vaccine platform with all of the meeting participants. Lastly, we are grateful for the co-organization by the US-JAPAN Cooperative Medical Sciences Program on Immunology, Leading Graduate School at Chiba University for help organizing the 2nd IIMVF symposium.

Toshinori Nakayama
Hiroshi Kiyono
Stephen Schoenberger
Mitchell Kronenberg
The organizers of the 2nd IIMVF symposium

The Aim of the IIMVF

Currently, more research studies are being undertaken to unlock the mechanisms underlying antigen-specific immune responses, a characteristic feature of the acquired immune system that is initiated by the innate immune system. Although "Immunological Memory" is a central theme in acquired immunity, it is not fully understood at the molecular, cellular and in vivo levels, and thus this research area needs to be given further stimulation particularly for the next generation of junior immunologists in Japan together with international leading scientists. New idea and findings from modern research on "Immunological Memory" will contribute to the development of more effective and safer vaccines against infectious diseases, therapeutic vaccines for controlling so called pathogenic immune memory cells that trigger allergy or autoimmune diseases, and vaccines for cancer. This forum covers the issues of "immunological memory and vaccine research" in a broad sense, including basic to clinical immunological studies and translational research for vaccine development, aiming to provide an interactive and simulative environment for promoting exchange of new dogma, idea and data among young and established immunologists interesting on the area of immunological memory and vaccine studies with an international perspective. Hence we launch the International Immunological Memory and Vaccine Forum (IIMVF) holding a meeting once a year (or as needed) as a global platform, in order to promote; 1) information exchange and mutual professional and social friendship among junior researchers, and between junior and senior researchers, 2) dissemination of junior researchers' studies to the world community by making presentations and discussions in the presence of world-class research leaders, and 3) industry-academia joint research at the international level, and securement of research funds.

From IIMVF web site "http://www.iimvf.jp/english/history.html".

~Program~

Monday, August 25

9:00	Arrival
9:00-9:10	Welcoming Remarks
Session I	**Mucosal immunology (Chair H. Kiyono/K. Takeda)**
9:10-9:40	Toshinori Nakayama “Pathogenic Memory Th2 Cells in the airway”
9:40-9:55	Masaki Miyazaki “Id2 and Id3 maintain the regulatory T cell pool to suppress Th2 inflammatory disease”
9:55-10:25	Mitchell Kronenberg “Mechanisms for the regulation of mucosal immunity by the herpes virus entry mediator"
10:25-11:00	**Coffee Break**
11:00-11:30	Kiyoshi Takeda “Regulatory mechanisms of gut homeostasis”
11:30-11:45	Yujun Huang "Driving pre-existing immunity at the mucosal borders of the intestine”
11:45-12:00	Satoshi Uematsu “Blockade of TLR3 protects mice from lethal radiation-induced gastrointestinal syndrome”
12:00-1:30	**Lunch and Poster Session**
Session II	**CD4 T cells (Chair: Shane Crotty)**
1:30-2:00	Sonoko Habu “The in vitro induction of serial transfer of anergy status: a model for long lasting immune tolerance”
2:00-2:15	Takanori So “A novel molecular function of TRAF5 in CD4 T cell lineage commitment ”
2:15-2:30	Koji Tokoyoda “Resting T helper cell memory in bone marrow”
2:30-3:00	Shane Crotty “Genetics of Tfh differentiation”
3:00-3:15	Norifumi Iijima “A local chemokine network sustains tissue-resident memory T cells crucial for antiviral protection”
3:15-3:45	**Coffee Break**

Session III	**CD8 T cells (Chair: Alex Sette)**
3:45-4:15	Alex Sette “Comparative analysis of human T cell responses to DENV and TB”
4:15-4:30	Makoto Kurachi “The transcription factor BATF operates as an essential differentiation checkpoint in early effector $CD8^+$ T cells
4:30-4:45	Shiki Takamura “CD69 controls a balance between S1P- and CXCL16-mediated chemotaxis in the establishment of $CD8^+$ T cell memory in the lung airways”
4:45-5:00	Patrick Metz “Regulation of asymmetric division and CD8+ T lymphocyte fate specification by PKCζ and PKCλ/ι”
5:00-5:30	Louis Picker “The Unique Immunobiology of Cytomegalovirus Vectors; New Immunology from a Very Old Virus”
5:30	Adjourn for the day

Tuesday, August 26

Session IV	**Vaccines 1 (Chair: Ken Ishii)**
9:00-9:30	Hiroshi Kiyono “Mucosal immunity and vaccine development”
9:30-10:00	Bali Pulendran “Systems Vaccinology: enabling rational vaccine design with systems biology**”**
10:00-10:30	Ken J Ishii “Innovation and renovation of vaccine adjuvant”
10:30-11:00	**Coffee Break**
Session V	**Novel regulatory pathways (Chair: Erika Pearce)**
11:00-11:30	Erika Pearce “Metabolic interactions in the tumor microenvironment”
11:30-11:45	Wataru Ise “Regulation of germinal center response by the transcription factor BATF”
11:45 -12:15	Masato Kubo “Notch regulates reciprocal expression of CCR7 versus CXCR5 to control central memory T cell generation”

12:15 -12:30	Kiyoshi Hirahara “Asymmetry of STAT action to define specificity and redundancy of IL-27 and IL-6 signals”
12:30-1:30	**Lunch**
Session VI	**B cells (Chair: Tomohiro Kurosaki)**
1:30-2:00	Tomohiro Kurosaki “Mechanisms underlying rapid memory IgG responses”
2:00-2:30	Daisuke Kitamura “Molecular mechanisms for the development of B-cell memory”
2:30-3:00	Nobuo Sakaguchi “Role of GANP in the DNA repair for AID-induced somatic hypermutation generating high affinity antibodies”
3:00-3:30	**Coffee Break**
Session VII	**Vaccines 2 (Chair: Hiroshi Kiyono)**
3:30-3:45	Klaus Ley “Protective Autoimmunity in Atherosclerosis”
3:45-4:15	Uli von Andrian “A mucosal vaccine against *Chlamydia trachomatis* generates two synergistic waves of protective memory T cells”
4:15-4:30	Raphael Zellweger “Importance of CD8 T cells for protection against heterotypic Dengue virus infection and antibody-mediated severe disease”
4:30-4:45	Sunnie Yoh “PQBP1 is an innate immune receptor for HIV-1”
4:45-5:15	Dennis Burton “HIV vaccine targets identified by broadly neutralizing antibodies”
5:15-5:30	Closing remarks
5:30-6:45	Refreshments

Monday, August 25

Session I

Mucosal Immunology

SESSION I

Pathogenic memory Th2 cells in the airway

Toshinori Nakayama, Yusuke Endo, Damon J. Tumes and Kiyoshi Hirahara

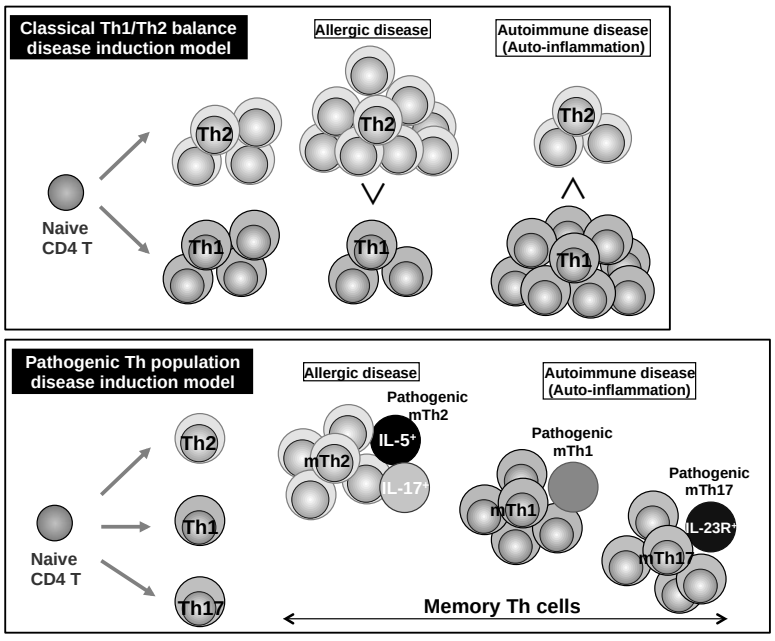
Department of Immunology, Graduate School of Medicine, Chiba University, Japan
CREST, Japan Science and Technology Agency, Japan

Immunological memory is a hallmark of adaptive immunity. Memory CD4 T helper (Th) cells are central to acquired immunity, and vaccines for infectious diseases are developed based on this concept. However, memory Th cells also play a critical role in the pathogenesis of various chronic inflammatory diseases, including asthma. We refer to these populations as "**pathogenic memory Th cells**". I would like to introduce our experimental results that address the generation of pathogenic memory Th2 cells in allergic airway inflammation.

We have found that memory Th2 cells are divided into four subpopulations by CD62L/CXCR3 expression. All four subpopulations produced IL-4 and IL-13, while only the $CD62L^{low}CXCR3^{low}$ population produced IL-5. Increased levels of H3-K4 methylation at the *IL-5* gene locus were specifically observed in this population. Memory Th2-dependent airway inflammation was attenuated in the absence of the $CD62L^{low}CXCR3^{low}$ population. We identified a pathogenic IL-5-producing memory Th2 cell subset in allergic airway inflammation (Endo et al. Immunity 35: 733, 2011). Moreover, we have addressed the mechanisms regulating the pathogenicity of memory Th cells. Since the expression of the IL-33R;ST2 is high on the pathogenic Th2 cells, we analyzed whether the IL-33/ST2 axis license memory Th2 cells to induce allergic airway inflammation via production of IL-5, and an important role for IL-33/ST2 axis was revealed.

Based on these findings we propose a model named "**Pathogenic Th population and disease induction model**" (Fig. 1) wherein a pathogenic subpopulation of Th cells possessing a distinct feature of effector function (e.g. unique expression of cytokines and chemokine receptors) is generated *in vivo* and is crucial for the pathogenesis of Th1, Th2 or Th17 diseases, regardless of the balance of these Th subsets (Endo et al. Trends in Immunology 35:69, 2014).

Fig.1



Endo et al. Trends in Immunol. 2014

SESSION I

Id2 and Id3 maintain the regulatory T cell pool to suppress Th2 inflammatory disease

Masaki Miyazaki, Cornelis Murre

University of California, San Diego, USA

Regulatory T (Treg) cells suppress the development of inflammatory disease, but our knowledge of transcriptional regulators that control this function remains incomplete. Here we show that expression of Id2 and Id3 in Treg cells was required to suppress development of fatal Th2 inflammatory disease. We found that T cell receptor (TCR)-driven signaling initially decreased the abundance of Id3, which led to the expression of CXCR5. Depletion of Id2 and Id3 expression in Treg cells resulted in compromised maintenance and localization of the Treg cell population. Transcriptome analysis shows that Id2 and Id3 regulate effector Treg cell transcriptional signitures. Thus, Id2 and Id3 enforce effector Treg cell checkpoints and control the maintenance and homing of Treg cells. (Nature Immunology Aug. 2014)

SESSION I

Mechanisms for the regulation of mucosal immunity by the herpes virus entry mediator

Mitchell Kronenberg

La Jolla Institute for Allergy & Immunology, USA

The herpes virus entry mediator (HVEM) is a TNF super family receptor that binds multiple ligands, including TNF and Ig super family members. We have explored the role of HVEM in epithelial cells, a radiation resistant cell type critical for preventing chronic inflammation and enhancing host defense. During *Citrobacter rodentium* infection by oral gavage, *Hvem*$^{-/-}$ mice had impaired colonic epithelial responses, resulting in higher bacterial burdens, inflammation and increased mortality. HVEM stimulation induced epithelial responses by a novel signaling pathway, with NIK-dependent Stat3 activation acting downstream of HVEM, resulting in the expression of genes important for mucosal immunity. A type of innate lymphoid cells (ILC) called ILC3, also expresses HVEM. ILC3 are characterized by the production of IL-22, and HVEM expression by these cells is required for their optimal production of this cytokine. In the absence of HVEM expression by ILC3 in conditional HVEM knockout mice, constitutive IL-22 production is reduced with corresponding changes in the microbiota and a resulting increase in mucosal Th17 cells. Therefore, HVEM expression by different cell types in the intestine has important effects on both innate and adaptive immunity.

SESSION I

Regulatory mechanisms of gut homeostasis

Kiyoshi Takeda

Department Microbiology and Immunology, Graduate School of Medicine, WPI Immunology Frontier Research Center, Osaka University, Japan

Intestine is a unique tissue, where many commensal microbes inhabit. Therefore, intestinal mucosa is protected from these commensal bacteria as well as pathogenic bacteria by several types of barriers. One of these barriers is constructed by mucus layers, composed of the inner firm mucus layer and outer loose mucus layer in the large intestine. Commensal bacteria are present in the outer mucus layer, whereas there is no commensal bacterium in the inner mucus layer. Separation of commensal bacteria from the intestinal epithelial cells contributes to prevention of intestinal inflammation. Indeed, the presence of bacteria on the epithelial surface of the large intestine was reported in several mouse models of intestinal inflammation. However, the precise mechanisms by which the inner mucus layer is free of bacteria in the large intestine remain unknown.

We found a molecule, which was selectively expressed on the apical surface of the epithelial cells at the top of colonic glands. This molecule was a highly glycosylated GPI-anchored protein, and cleaved and secreted into the colonic lumen, particularly the inner mucus layer. In mice lacking this molecule, bacterial free space in the inner mucus layer disappeared and they were highly susceptible to intestinal inflammation. On the intestinal epithelial cell layer and crypts of the large intestine of the mutant mice, flagellated bacteria such as *Helicobacter* spp. and *Proteus* spp. including *P. mirabilis* were present. Depletion of these bacteria by antibiotics restored the bacterial free space in the inner mucus layer and ameliorated the intestinal inflammation of the mutant mice. This molecule bound to *P. mirabilis* and inhibited their attachment to the intestinal epithelial cells. These findings demonstrated that this molecule mediates segregation of commensal bacteria from the intestinal epithelial layer in the large intestine, and thereby contributes to the maintenance of gut homeostasis.

SESSION I

Driving Pre-existing Immunity at the Mucosal Borders of the Intestine

Yujun Huang, Yiran Wang-Zhu, Alexandre Larange, Ryo Shinnakasu, Christopher J. Lena, Mitchell Kronenberg, and Hilde Cheroutre

La Jolla Institute for Allergy & Immunology, USA

CD8 effector memory T cells (TEM) residing at mucosal epithelium have a heightened and immediate effector function to prevent invasion of pathogens and to control the systemic immune sensing. The mechanisms that drive the differentiation of superior tissue resident TEM are still poorly understood. Previously we have shown that high affinity/avidity TCR stimulation induces CD8aa expression on the most effective CD8 Teff and that those CD8aahi CD8ab T effector cells (Teff) are selected to become mucosal TEM. We generated new data showing that the “fittest” CD8aahi CD8+ Teff are favored by strong TCR stimulation and pro-inflammatory conditions. CD8aa serves as a co-repressor for TCR signaling and tunes down TCR proximal signaling in order to rescue CD8 Teff from strong activation-induced cell death (AICD). Strong TCR stimulation and pro-inflammatory conditions induce more CD8aahi precursor cells and enhance TEM generation in small intestine. These fully differentiated TEM at the mucosal border have potent protective immunity against pathogens. Our findings emphasize the importance of tissue resident TEM for vaccinations and provide important and highly novel insights to design successful alternative vaccine approaches.

SESSION I

Blockade of TLR3 protects mice from lethal radiation-induced gastrointestinal syndrome

Satoshi Uematsu

Department of Mucosal Immunology, School of Medicine, Chiba University, Japan/ Division of Innate immune regulation, International Research and Development Center for Mucosal Vaccine, Institute of Medical Science, The University of Tokyo, Japan

High-dose ionizing radiation induces severe DNA damage in the epithelial stem cells in small intestinal crypts and causes gastrointestinal syndrome (GIS). Although the tumor suppressor p53 is a primary factor inducing death of crypt cells with DNA damage, its essential role in maintaining genome stability means inhibiting p53 to prevent GIS is not a viable strategy. Here, we show that the innate immune receptor Toll-like receptor 3 (TLR3) is critical for the pathogenesis of GIS. *Tlr3*$^{-/-}$ mice show substantial resistance to GIS owing to significantly reduced radiation-induced crypt cell death. Despite showing reduced crypt cell death, p53-dependent crypt cell death is not impaired in *Tlr3*$^{-/-}$ mice. p53-dependent crypt cell death causes leakage of cellular RNA, which induces extensive cell death via TLR3. An inhibitor of TLR3–RNA binding ameliorates GIS by reducing crypt cell death. Thus, we propose blocking TLR3 activation as a novel and preferable approach to treat GIS.

NOTE

Monday, August 25

Session II

CD4 T Cells

SESSION II

The in vitro induction of serial transfer of anergy status: a model for long lasting immune tolerance.

Naoko Negishi and Sonoko Habu

Department of Immunology, Juntendo University Scholl of Medicine, Japan

Start text here. Immunological memory is greatly important for protecting the individual against recurring diseases due to similar pathogens, but its cellular and molecular basis is still not fully understood, particularly the process of generating a particular long-lasting immunity against pathogens. For challenging this problem, any suitable experimental system is required. Immune cells responding to recurrent challenge are varied. Among them, we are focusing T cells that regulate immune tolerance, particularly transplantation tolerance where anergy status is maintained for a long duration. To address this issue, we have recently established the in vitro induction system in which anergy status is serially transferred into naïve T cells in turn, using TCR Tg mice in which the TCR recognizes OVA peptide and also reacts to superantigen TSST-1 through TCR Vb18 chain. In the present talk, we will introduce the in vitro experimental system we generated and show the features of CD4+T cells responsible for serial anergy status. We will also discuss about a possibility that our established experimental system may be a memory model in which anergy status of immune system can be generated by serial transfer of certain cell characters.

SESSION II

A novel molecular function of TRAF5 in CD4 T cell lineage commitment

Takanori So

Tohoku University Graduate School of Medicine,
Department of Microbiology and Immunology, Japan

The family of 6 TNF receptor-associated factors (TRAFs) function as adaptor proteins for the TNF receptor superfamily, the Toll-like receptor family, and the RIG-I-like receptor family by associating with the intracellular domains of these receptors to mediate downstream signaling events. Our previous data shows that allergic lung inflammation mediated by effector CD4 T cells is more exaggerated in *Traf5*$^{-/-}$ mice than in wild-type mice and suggests that TRAF5 has a function that has not been delineated in detail in CD4 T cells. In this study, we have identified a novel attribute of TRAF5 in IL-6 signaling pathway. In the presence of IL-6, differentiating naïve *Traf5*$^{-/-}$ CD4 T cells produced more IL-17 than did wild-type CD4 T cells and developed a pronounced Th17 phenotype both *in vitro* and *in vivo*. Accordingly, Th17-associated experimental autoimmune encephalomyelitis (EAE) was greatly exaggerated in *Traf5*$^{-/-}$ mice than in wild-type mice, and *Traf5*$^{-/-}$ CD4 T cells induced exaggerated EAE in TRAF-sufficient recipient mice. Surprisingly, TRAF5 constitutively associated with gp130 and suppressed the recruitment and activation of STAT3 in response to IL-6. Amino acid residues 774-798 in the cytoplasmic of gp130 were critical for the binding, and this region contains an SXXE motif and two di-acidic amino acids, which are recognition elements for the TRAF-C domain. The finding therefore provides a new perspective on the lineage commitment of CD4 T cell differentiation and an explanation why TRAF5 exhibits an anti-inflammatory function in CD4 T cells.

SESSION II

Resting T helper cell memory in bone marrow

Koji Tokoyoda

German Rheumatism Research Center Berlin (DRFZ), Germany

Immunological memory provides long-term protective immunity against pathogens which have encountered before. Memory is maintained by long-lived memory cells, generated in the primary challenge. Long-lived memory T helper (Th) cells play a crucial role in the generation, maintenance and reactivation of other memory cells. We have demonstrated that most of long-lived memory Th cells reside and rest in the bone marrow (BM). In the course of a systemic immune response, CD4 T cells relocate to the BM within 2 months after their generation. Antigen-specific memory Th cells of the BM express Ly-6C at high level and, upon challenge with antigen, they rapidly express cytokines and CD154 and efficiently induce the production of high-affinity antibodies by B cells. In the BM, memory Th cells are maintained on IL-7-expressing stromal niches which control the abundance of memory Th cells. However, the molecular mechanisms of their establishment and maintenance in the BM remained unclear. We have also shown that resting memory Th cells in the BM express high levels of two activation markers, CD49b and CD69, and that the loss of their markers reduces the number of memory Th cells only in the BM. Both markers can work as adhesion molecules for the transmigration of antigen-experienced CD4 T cells via sinusoids of the bone for the establishment of memory cells. These studies on these adhesion molecules clarified that BM memory Th cells are generated at a multi-step process. Here we show the kinetics on localization of memory Th cell precursors and memory cells during an immune response; memory Th cell precursors transmigrate into the marrow of bones via Collagen-$I^+$$CD144^+$ sinusoidal endothelial cells, are temporarily anchored on Collagen-II^+ and also $CXCL12^+$ stromal cells, and finally reach on Collagen-II^+ and also Collagen-$XI^+$$IL\text{-}7^+$ stromal cells. These results clarify how memory Th cell precursors relocate to the BM and also help an understanding of how memory cells are generated temporally and spatially.

SESSION II

Genetics of Tfh differentiation

Shane Crotty

La Jolla Institute for Allergy and Immunology, USA

Follicular helper T cells (Tfh) are the specialized providers of T cell help to B cells, and are essential for germinal centers, affinity maturation, and the development of most high affinity antibodies and memory B cells. Tfh differentiation is a multi-stage, multifactorial process involving Bcl6 and other transcription factors. Because of their important role regulating B cells and antibody responses, Tfh appear to be critical components of many protective immune responses against pathogens, as well as being positively associated with protective responses against multiple cancers. As such, there is strong interest in harnessing Tfh cell biology to enhance new vaccines. Tfh cell responses also are major components of a number of autoimmune diseases associated with autoantibody responses. Overall, there have been dramatic advances in this young field, but there are still major gaps in our understanding of Tfh differentiation and functions and there is much to be learned about the biology of these cells in the interest of applying that knowledge to biomedical needs. Our recent work has focused on understanding the genetics of Tfh cells. We have used RNAi based genetic screens and BCL6 Chip-seq to understand Tfh biology. The BCL6 cistrome in primary human GC TFH cells is markedly distinct from the BCL6 cistrome in primary human GC B cells. In Tfh, BCL6 directly represses genes regulating T cell migration and TCR signaling, as well as gene sets defining alternative T cell fates. Although some BCL6 binding sites contained BCL6 binding motifs, BCL6 binding sites were also highly enriched in AP-1 or STAT motifs. AP-1 complexes are key positive downstream mediators of TCR signaling and external stimuli. We show that AP-1 interacts with BCL6 and co-occupies BCL6 binding sites with AP-1 motifs suggesting that BCL6 prevents or subverts AP-1 activity. The BCL6 cistrome exhibits unique T cell specific features that may explain how this master regulatory mediates distinct cell-context dependent phenotypes.

SESSION II

A local chemokine network sustains tissue-resident memory T cells crucial for antiviral protection

Norifumi Iijima and Akiko Iwasaki

Department of Immunobiology, Yale University School of Medicine,USA

Tissue resident memory T cells (T_{RM}) are critical in protecting the host from a variety of pathogens. Some tissues are permissive for T cell entry, while others including the brain, skin and vagina require inducible factors. Skin infection generates CD8 T_{RM} that are sequestered in the epidermis, and CD4 T cells that traffic rapidly through the dermis into circulation However, whether CD4 T_{RM} cells can be established in an otherwise restrictive tissue, and if so, how such cells are maintained remains unclear. Here, by using parabiotic mice, we show that a pre-existing pool of memory CD4 T cells in the genital mucosa is required for full protection from a lethal herpes simplex virus 2 (HSV-2) infection. The vaginal memory T cell pool established through local immunization underwent minimal exchange with the systemic circulation, and was sufficient for protection. In contrast, circulating memory T cells entered the vagina only after the viral challenge, and provided only a partial protection against viral replication and disease. Vaginal CD4 T_{RM} cells were maintained in distinct memory lymphocyte clusters (MLCs) beneath the epithelial layer through inducible chemokines secreted by local network of resident cells. Our results highlight a critical need for vaccine strategies that enable establishment of local memory T cells for protection from sexually transmitted viruses, and provide insight about how such pool might be established through a local chemokine network.

Monday, August 25

Session III

CD8 T Cells

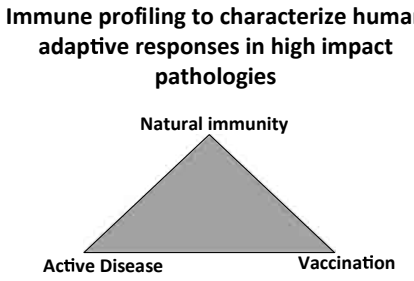
SESSION III

Comparative analysis of human T cell responses to DENV and TB

Alessandro Sette, Cecilia Lindestam, Daniela Weiskopf, Vijay Pandurangan and Bjoern Peters

La Jolla Institute for Allergy and Immunology, USA

The long-term goal of our research is to characterize immune response profiles that correlate with the outcome of infection or vaccination. More specifically, our current focus is on CD4 and CD8 T cell responses associated with two pathogens of widespread significance to human health and disease, namely mycobacteria tuberculosis and dengue virus. Our approach relies on the evaluation of responses observed 1) in populations previously exposed and naturally associated with immunity and/or control of infection 2) individuals associated with active and severe disease 3) individuals to which licensed or experimental vaccines have been administered. We will review experiments that led to the identification and characterization of hundreds of different CD4 and CD8 T cell epitopes derived from TB and DENV, and started to define phenotypes associated with protection, disease and vaccination. In the case of DENV we will describe phenotypes of *protective T cell responses*, as defined by phenotypes in populations from endemic areas, associated with multiple infections and protection form severe disease. We will present data relating to T cells restricted by HLA class I and class II molecules associated with decreased or increased susceptibility to severe disease. These data can be contrasted with those derived from *pathogenic T cell responses*, defined by T cell phenotypes observed in longitudinal sampling of PBMCs from DENV infected individuals associated with differential disease severity, from early infection time points into convalescence. Finally, these phenotypes will be compared with those observed following vaccination with experimental DENV live-attenuated vaccine (DLAV). Likewise, we are also analyzing T cells in the context of tuberculosis infection and vaccination. In this case, we are analyzing T cell responses associated with a contained TB infection as observed in individuals with latent MTB that are otherwise healthy. These T cell responses are compared to those observed as a result of exposure to nonpathogenic mycobacteria, and to those induced by BCG vaccination.



SESSION III

The transcription factor BATF operates as an essential differentiation checkpoint in early effector CD8$^+$ T cells

Makoto Kurachi[1,2,5], R Anthony Barnitz[3,5], W Nicholas Haining[3,4], E John Wherry[1,2]

[1]*Department of Microbiology, University of Pennsylvania Perelman School Medicine, USA* [2]*Institute for Immunology, University of Pennsylvania Perelman School Medicine, USA* [3]*Department of Pediatric Oncology, Dana-Farber Cancer Institute, Harvard Medical School, USA* [4]*Division of Hematology/Oncology, Children's Hospital, Harvard Medical School, Boston, USA* [5]*These authors contributed equally to this work.*

The transcription factor BATF is required for the differentiation of interleukin 17 (IL-17)-producing helper T cells (T_H17 cells) and follicular helper T cells (T_{FH} cells). Herein we identify a fundamental role for BATF in regulating the differentiation of effector of CD8$^+$ T cells. BATF-deficient CD8$^+$ T cells showed profound defects in effector population expansion and underwent proliferative and metabolic catastrophe early after encountering antigen. BATF, together with the transcription factors IRF4 and Jun proteins, bound to and promoted early expression of genes encoding lineage-specific transcription-factors (T-bet and Blimp-1) and cytokine receptors while paradoxically repressing genes encoding effector molecules (IFN-g and granzyme B). Thus, BATF amplifies T cell antigen receptor (TCR)-dependent expression of transcription factors and augments the propagation of inflammatory signals but restrains the expression of genes encoding effector molecules. This checkpoint prevents irreversible commitment to an effector fate until a critical threshold of downstream transcriptional activity has been achieved.

SESSION III

CD69 controls a balance between S1P- and CXCL16-mediated chemotaxis in the establishment of $CD8^+$ T cell memory in the lung airways

Shiki Takamura

Department of Immunology, Kindai University Faculty of Medicine, Japan

Memory $CD8^+$ T cells persist in the lung airways of both humans and animals following resolution of a respiratory virus infection and play a key role in mediating the initial phase of the recall response to secondary infection. These cells are separated from the circulation and thus categorized as a tissue-resident memory (T_{RM}) population. Unlike $CD8^+$ T_{RM} cells that reside in the other sites, the number of $CD8^+$ T_{RM} cells in lung airway is maintained by a process of continual recruitment. However, the mechanisms by which these cells are recruited to the lung airways during steady-state conditions are poorly understood. We show that reactivation of memory $CD8^+$ T cells in the lung parenchyma is vital for the conversion form T_{EM} to T_{RM} and subsequent recruitment to the lung airways. Upon reactivation, memory $CD8^+$ T cells upregulate two key molecules, CD69 and chemokine receptor CXCR6. CD69 inhibits sphingosine-1-phosphate receptor 1($S1P_1$)-mediated egression of memory $CD8^+$ T cells from the lung parenchyma, while CXCR6 promotes migration toward CXCL16 expressed in the lung airways. Thus, CD69 and CXCR6 cooperatively promote the recruitment of memory $CD8^+$ T cells to the lung airways. Our findings that reactivation in the lung parenchyma is required for continual recruitment of memory $CD8^+$ T cells to the lung airways has important implications for the development of vaccines that promote durable and effective immunity against respiratory pathogens, such as influenza virus.

SESSION III

Regulation of Asymmetric Division and CD8+ T Lymphocyte Fate Specification by PKCζ and PKCλ/ι”

Patrick Metz, Janilyn Arsenio, Boyko Kakaradov, Stephanie H. Kim, Kelly Remedios, Katherine Oakley, Gene W. Yeo, John T. Chang

UCSD, USA

During an immune response against a microbial pathogen, naïve T lymphocytes give rise to effector cells that provide acute host defense and memory cells that provide long-lived immunity. It has been shown that activated naïve T lymphocytes can undergo asymmetric division, enabling the daughter cells to inherit unequal amounts of fate-determining proteins and thereby acquire distinct fates from their inception. Here, we show that the absence of the atypical protein kinase C (aPKC) isoforms, PKCζ and PKCλ/ι, disrupts asymmetric CD8+ T lymphocyte division, resulting in aberrant acquisition of a ‘pre-effector’ transcriptional program, detected by single-cell gene expression analyses, in lymphocytes that had undergone their first division in vivo. These alterations were associated with enhanced differentiation toward effector fates at the expense of memory fates. Together, these results demonstrate a role for aPKC in regulating asymmetric division and the specification of divergent CD8+ T lymphocyte fates early during an immune response.

NOTE

Tuesday, August 26

Session IV

Vaccines 1

SESSION IV

Mucosal Immunity and Vaccine Development

Hiroshi Kiyono

Division of Mucosal Immunology, Department of Microbiology and Immunology,
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The epithelium of digestive tract is continuously exposed to infinite beneficial and harmful antigens including commensal and pathogenic microbe, in handling its day-to-day duties. The digestive tract is thus equipped with the mucosal immune system (MIS) offering the first line of defense against invasion of pathogens at same time, the MIS also creating a homeostatic condition to commensal microorganisms. Fucosylation of intestinal epithelial cells, catalyzed by fucosyltransferase 2 (Fut2), is a major glycosylation mechanism of host–microbiota symbiosis. Our most recent study has shown that innate lymphoid cells (ILCs) induced intestinal epithelial *Fut2* expression and fucosylation. Deficiency of intestinal ILC-mediated fucosylation led to increased susceptibility to infection by *Salmonella typhimurium*. Further, these ILCs play a critical role in the creation of intra-tissue co-habitation of commensal flora, *Alcaligenes* in Peyer's patches. Our data reveal critical roles of ILCs in shaping the gut microenvironment through the regulation of epithelial glycosylation and containment of intra-tissue commensal flora.

Gut pathogen-induced diarrhea is a life-threatening disease in developing countries. Based on our knowledge in mucosal immunology and cross fertilization with agriculture science, our effort has been aiming to develop rice transgenic (Tg) vaccine system, "MucoRice". Tg rice expressing B subunit of cholera toxin (MucoRice-CTB) has been shown to induce antigen-specific SIgA-mediated protective immunity against *Vibrio cholera*-induced diarrhea. Our study has been extended to construct Tg rice expressing the neutralizing variable domain of a rotavirus-specific nanoantibody. Orally administered MucoRice-ARP1 markedly decreased the viral load in rotavirus-challenged mice. The MucoRice system is thus an attractive vaccine antigen and nanoantibody expression, preservation and delivery platform for the development of next generation of mucosal vaccines.

1. Goto, Y., et al. 2014. Innate lymphoid cells regulate intestinal epithelial cell glycosylation. Science (in press)
2. Sonnenberg, G.F., et al. 2012. Innate lymphoid cells promote anatomical containment of lymphoid-resident commensal bacteria. Science 336:1321-1325.
3. Goto, Y., et al. 2012. Epithelial barrier: An interface for the cross-communication between gut flora and immune system. Immunol Rev. 245:147-163.
4. Tokuhara, D., et al. 2013. Rice-based oral antibody fragment prophylaxis and therapy against rotavirus infection. J. Clin. Invest. 123:3829-3838.
5. Tokuhara, D., et al. 2010. Secretory IgA-mediated protection against *V. cholerae* and heat-labile enterotoxin-producing enterotoxigenic *Escherichia coli* by rice-based vaccine. Proc. Natl. Acad. Sci. USA. 107: 8794-8799.

SESSION IV

Systems Vaccinology: enabling rational vaccine design with systems biology

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Despite their great success, we understand little about how effective vaccines stimulate protective immune responses. Two recent developments promise to yield such understanding: the appreciation of the crucial role of the innate immune system in sensing microorganisms and tuning immune responses, and advances in systems biology. In this presentation, I will discuss how these developments are yielding insights into the mechanism of some of the most successful vaccines ever developed. Furthermore, such developments promise to address a major challenge in vaccinology: that the efficacy of a vaccine can only be ascertained retrospectively, upon infection. The identification of molecular signatures induced rapidly after vaccination, which correlate with and predict the later development of protective immune responses, would represent a strategy to prospectively determine vaccine efficacy. Such a strategy would be particularly useful when evaluating the efficacy or immunogenicity of untested vaccines, or in identifying individuals with sub-optimal responses amongst high risk populations, such as infants or the elderly. We have recently used a systems biology approach to identify early gene signatures that correlate with, and predict the later immune responses in humans vaccinated with the live attenuated yellow fever vaccine YF-17D, or with the influenza vaccines. I will review these studies, and discuss their broader implications for vaccinology.

SESSION IV

Innovation and renovation of vaccine adjuvant

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The word adjuvant has its origin from the Latin "adjuvare", meaning "to help". It is a general term for substances (factors) which are co-administered with a vaccine with the aim of increasing the effect (immunogenicity) of the vaccine. The research and development of adjuvants has a history of more than 80 years, and their actual mechanism was not immunologically understood for a long time, with a famous sarcastic remark "Immunologist's dirty little secret". Recent advance in Immunolgy; however, allowed the development of adjuvants through an innovative scientific approach, and there is fierce competition worldwide for the development of next-generation adjuvants. I would like to introduce and discuss about several adjuvants with their novel mechanisms, including a small compound as a potent DAMPs inducer to target certain innate immune mechanisms.

On the other hand, however, adjuvants range widely in terms of origin and mode of action, and they may be the cause or underlying cause of vaccine toxicity, especially immunotoxicity. I will present our recent work that common particulate adjuvants can cause local but sustained inflammation and allergic responses via novel innate immune mechanisms and cell death.

1: Kobiyama K et al Nonagonistic Dectin-1 ligand transforms CpG into a multitask nanoparticulate TLR9 agonist. ***Proc Natl Acad Sci U S A.*** 2014 111(8):3086-91.

2: Kuroda E, Coban C, Ishii KJ. Particulate adjuvant and innate immunity: past achievements, present findings, and future prospects. ***Int Rev Immunol***. 2013 32(2):209-20.

3: Jounai N, et al. Recognition of damage-associated molecular patterns related to nucleic acids during inflammation and vaccination. ***Front Cell Infect Microbiol***. 2012;2:168.

4: Desmet CJ, Ishii KJ. Nucleic acid sensing at the interface between innate and adaptive immunity in vaccination. ***Nat Rev Immunol.*** 2012 12(7):479-91.

5: Marichal T et al. DNA released from dying host cells mediates aluminum adjuvant activity. ***Nat Med.*** 2011 17(8):996-1002.

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Session V

Novel Regulatory Pathways

SESSION V

Metabolic interactions in the tumor microenvironment

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Metabolism is the set of biochemical reactions that occur within cells to sustain life. As such, metabolism, by definition, remains the single most fundamental force driving cell fate. Given the critical nature of T cells in clearing and controlling infections and cancer, as well as mediating protective immunity over the long-term, it is logical that a considerable effort is made to target these cells for therapeutic purposes. However, while metabolism regulates the fate and function of T cells, or of any immune cell for that matter, metabolic interventions for manipulating immunity are rare and can be considered a largely untapped opportunity. Our research is focused on establishing fundamental mechanisms of metabolic regulation in T cells, with a view toward identifying new ways to regulate immune cell function through the manipulation of metabolic pathways. Underlying mechanisms of how T cell metabolism and function is altered in the tumor microenvironment will be discussed.

SESSION V

Regulation of germinal center response by the transcription factor BATF

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In response to T cell-dependent antigens, antigen-specific B cells interact with follicular helper T cells and are driven into the germinal center (GC) reaction, which is critical for the generation and selection of memory B cells and plasma cells that express somatically mutated high-affinity antibodies. The transcription factor BATF is essential for the formation of follicular helper T cells and thus GC, although whether BATF plays a distinct role in GC B cell remains unclear. To elucidate B-cell intrinsic function of BATF in vivo, we generated *Batf* floxed mice, which allowed us tissue-specific or inducible deletion of *Batf* and examined the effect of *Batf* deficiency in B cells on the induction or maintenance of GC response.

We found that GC formation was completely absent in mice in which *Batf* was deleted in B cells, after immunization with protein Ags. This effect was evident as early as day 5-7 following immunization, suggesting that BATF regulates early GC programs. Furthermore, GC response was collapsed when *Batf* was inducibly deleted from mature GC B cells after day 10-14 following immunization, suggesting that BATF might control sustained GC B cell proliferation. Thus, our results demonstrate a B-cell intrinsic requirement for BATF for not only generation but also for maintenance of GC formation.

SESSION V

Notch regulates reciprocal expression of CCR7 versus CXCR5 to control central memory T cell generation

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[2] *Laboratory for Cytokine Regulation, RCAI, RIKEN Center for Integrative Medical Sciences (IMS), RIKEN Yokohama Institute , Japan*

Notch signaling is currently reported to control the T_{FH} cell program by modification of the balance between Bcl6 and Blimp1 expression (*J Immunol* 2013; 191:2344-2350), and these T_{FH} cells seems to have a greater propensity to be maintained as memory T (TM) cells. Here we found that T_{FH} cells lacking Notch signaling in *Rbpj*f/f cd4-cre mice failed to generate TM cells and recall response in three different antigen systems, influenza virus infection, OVA-specific OTII Tg system, and a pMHCII tetramer-based system. Adaptive transfer of OVA-specific T_{FH} cell revealed that TM cells is considered to be maintained in specialized niches as central memory (TCM) cells in the T cell zone near the T-B border area. T_{FH} lost the expression of CCR7, and at 7 days after transfer, T_{FH} cells re-expressed CCR7 in transcription and protein level, instead of down-regulation of CXCR5 expression, subsequently leading to localization of memory niches. However, the T_{FH} cells derived from *Rbpj*f/f cd4-cre mice exhibited abnormal migration as a consequence of defect of CCR7 up-regulation. Importance of the CCR7 up-regulation responsible for TCM re-localization was confirmed by the functional evidence that the CCR7 blockade by antibody administration in normal mice terminated memory response. Moreover, Tamoxifen based conditional deletion of *Rbpj* indicated that input of Notch signaling was required at initial priming, but the deletion in contraction phase did not affect memory function. The defect of CCR7 up-regulation observed in *Rbpj* deficiency was due to the enhanced expression of *prdm1* (Blimp1), suggesting that the Notch-regulated Blimp expression had a role in the negative regulation of CCR7 expression. These data demonstrate that the Notch signaling have a great impact on memory maintenance regulated by the CCR7 up-regulation as a result of unbalanced regulation of Blimp1 and Bcl6.

SESSION V

Asymmetry of STAT action to define specificity and redundancy of IL-27 and IL-6 signals

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Interleukin (IL-)6 and IL-27 are regarded as pro-inflammatory and anti-inflammatory cytokines, respectively. While both cytokine-signals go through gp130 and use STAT3 and STAT1 as signal transducing factors. In order to understand how specificity and redundancy of cytokine action is formulated, we conducted genome-wide transcriptome and chromatin mapping analysis in T helper cells. The loss of STAT3 significantly collapsed the size of transcriptional output regulated by IL-6 or IL-27, while the loss of STAT1 reduced diversity between IL-6 and IL-27 regulated genes. *STAT1* gain-of-function mutations in human led to enhanced diversity between IL-6 and IL-27. On key genes that define T helper phenotype, gene expression was fine-tuned by balancing act of STAT3 and STAT1 on chromatin. Thus, the results indicated that STAT3 is a fundamental driver for both IL-6 and IL-27 response whereas STAT1 is an important modulator to shape cytokine unique signature, particularly of IL-27.

NOTE

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Session VI

B Cells

SESSION VI

Mechanisms underlying rapid memory IgG responses

Tomohiro Kurosaki[1,2], Kohei Kometani[2], Wataru Ise[1]

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One striking feature of humoral memory response is quick generation of neutralizing antibodies (Abs) upon re-invasion of pathogenic micro-organisms and eliminating them from our body. However, it is still unclear about cellular and molecular mechanisms underlying such quick humoral responses.

By using model T-dependent antigens, we propose that the following three mechanisms synergistically operate. First, IgG type memory B cells are pre-localized in the place where they can gain easy access to the antigen. Second, Tfh type memory T cells, which reside near to the memory IgG B cells. These memory T cells are promptly activated by antigen-presentation on IgG memory B cells and re-express Bcl6, which in turn activates IgG memory B cells. Finally, IgG memory B cells undergo repression of the Bach2 transcription factor (one of the BCR-extrinsic changes) during primary immune responses, which in turn, contributes to rapid differentiation towards plasma cells.

SESSION VI

Molecular mechanisms for the development of B-cell memory

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During T-dependent (TD) immune response, antigen-bound B cells undergo class-switching from IgM to IgG, and proliferate extensively to form germinal centers (GC) in the secondary lymphoid organs. After accumulating mutations at Ig V region genes, GC B cells with high antigen-binding affinity are selected and differentiate into either long-lived plasma cells (LLPCs) or memory B (B_{mem}) cells. Although this phenomenon is well known, mechanisms for the development and maintenance of B_{mem} cells remain poorly understood. Recently, it has been shown that B cells also switch to IgE in the GC but the IgE^+ B cells immediately become short-lived plasma cells and not differentiate into LLPCs or B_{mem} cells in healthy individuals. Thus, unlike IgG classes, serum IgE emerges only transiently at the early phase of the immune response and normally maintained at very low level, and IgE^+ B_{mem} cells can hardly be detected even after TD immunization, thus preventing the onset of allergic disorders. To study how the IgE^+ B_{mem} cell development is regulated will help us to understand the mechanism of IgG^+ B_{mem} cell development.

To study the regulatory mechanisms of IgE^+ B cell development, we utilized in-vitro induced germinal center-like B (iGB) cells. The iGB cells cultured on a feeder layer expressing CD40L and BAFF with IL-4 proliferate extensively while switching either to IgG1 or IgE. When transferred into mice, IgG^+ iGB cells generate memory-like B cells that survive for more than 2 months in the recipient, whereas IgE^+ iGB cells transiently become plasma cells and shortly disappear. Using iGB cells from “floxed” V-gene knock-in mice carrying *Cγ1-cre* transgene, we swapped the endogenous Ig H chain with exogenous ones of a given class. With this system, we found that IgE-class B-cell receptor (BCR) constitutively recruited BLNK and CD19-PI3K, resulting in IRF4-Blimp1 induction and spontaneous plasma cell differentiation. Crosslinking of IgE-BCR induced more robust and sustained tyrosine phosphorylation of cellular proteins including BLNK than that of IgG1. BLNK inhibited endocytosis of antigen-bound IgE-BCR and sustained activation of downstream signaling, resulting in attenuation of antigen presentation by IgE^+ B cells. In the BLNK-deficient mice, IgE^+ GC B and B_{mem} cells remained high in number during the TD response, and more IgE^+ LLPCs were produced, rendering the mice hypersensitive to the secondary challenge. These results together suggest that BLNK regulates both antigen independent and dependent IgE signaling, facilitating IgE^+ B cells to differentiate into short-lived plasma cells, and prohibiting their clonal expansion in the GCs and differentiation into B_{mem} cells or LLPCs. Considering the IgG^+ B_{mem} cell development, yet unknown signaling properties of IgG-BCR, especially in terms of BLNK-involvement, may affect the memory fate determination in the immune response.

SESSION VI

Role of GANP in the DNA repair for AID-induced somatic hypermutation generating high affinity antibodies

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Antigen (Ag) stimulation induces affinity maturation of BCRs during proliferation of Ag-specific B-cells in germinal centers (GCs) of peripheral lymphoid organs. Activation-induced cytidine deaminase (AID) is inevitable for induction of somatic hypermutation (SHM) at immunoglobulin variable region (*IgV-region*) genes. The Neuberger's original and the later modified models demonstrate that AID-induced C deamination primarily converts to U generating transition mutations of C→T and G→A. Uracil DNA Glycosylase (UNG) mounts further diversification at the U position to generate the transversion of C→A and G→T mutations. The UNG bares the DNA positions to give rise abasic sites, rendering the DNA strands sensitive to endonucleases and the physical stresses causing the breaks to either single strand or double strands DNA. These are eventually thought to repair through various DNA repair pathways including the base excision repair or the short patch repair mechanisms, all of which are expected to increase the SHM frequency of the *IgV-region*. However, it is unclear if only the increase of *IgV-region* SHM is necessary for affinity maturation.

Our current approach studied the molecular mechanism how such DNA injuries are controlled during the generation of SHM at the *IgV-region* genes in B-cells. To generate *IgV-region* SHM, AID needs to be accompanied with a co-factor for recruitment from the cytoplasm to the nucleus and toward the IgV-region nucleosomes in B-cells. Germinal center-associated nuclear protein (GANP) helps and enhances the AID accessibility through direct interaction at the cytoplasm and loosening the *IgV-region* nucleosomes by the GANP's acetyl-transferase (HAT) activity.

Here, we demonstrate that GANP plays a critical role in the quality control of SHM profile at the *IgV-region* genes in B-cells through regulating the choice of DNA repair pathways favoring the less harmful and high integrity condition. Furthermore, GANP regulates the amino acid change profile to maintain the overall protein structure of high affinity antibodies against the immunized Ag. This notion would be important for generating high affinity BCRs of Ag-reactive B-cell clones and maintaining memory B-cell condition in the acquired immunity.

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Session VII

Vaccines 2

SESSION VII

Protective Autoimmunity in Atherosclerosis

Klaus Ley, Kevin Tse, Takayuki Kimura, John Sidney, Alex Sette

LIAI, USA

Immunization of atherosclerosis-prone mice with low density lipoprotein (LDL) or its oxidized derivative (ox-LDL) was previously shown to be atheroprotective, but neither the antigen specificity nor the mechanism of protection is known. T-helper 1 CD4 T cells (Th1) are pro-atherogenic and regulatory CD4 T cells (Tregs) are atheroprotective in adoptive transfer mouse models. In two immunization studies, we tested the ability of five peptides from mouse Apo B, the main apolipoprotein in LDL, in a 1xCFA + 4xIFA (complete and incomplete Freund's adjuvant) to prevent atherosclerosis in Apoe-/- mice, a widely used model of atherosclerosis. The five peptides were selected for binding to mouse MHC-II (I-Ab, apparent binding affinities between 4.3 and 95 nM). Vaccination with each of the five peptides, but not irrelevant control peptides, reduced aortic en face lesion size by 35-60%, accompanied by small but significant atheroprotection in the aortic root. We saw no change in the number of FoxP3+CD25+ Tregs in lymph nodes or spleens, but significantly increased expression of IL-10 mRNA induced in aortas of immunized mice. The number of CD11c+CD103+RALDH+ dendritic cells (DCs), a phenotype that is consistent with DCs inducing peripheral tolerance, was expanded in aortas of immunized mice. B1a cells, a cell type thought to secrete atheroprotective antibodies, were expanded in the peritoneal cavities of immunized mice. We conclude that vaccination with MHC-II restricted peptides can protect from atherosclerosis in a relevant mouse model. Efforts are under way to generate MHC-II-peptide tetramers to detect antigen-specific T cells and identify their phenotype, to identify the nature of the IL-10 producing cells, and to translate this mouse work to develop an atheroprotective vaccine for humans.

SESSION VII

A mucosal vaccine against *Chlamydia trachomatis* generates two synergistic waves of protective memory T cells

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Vaccines that are administered via non-mucosal routes are often poorly protective against mucosal pathogens, presumably because such vaccines do not generate memory cells that migrate to mucosal surfaces. Although mucosa-tropic memory cells are inducible by mucosal immunization, few mucosal vaccines are currently in clinical use because live vaccine vectors pose safety risks and killed pathogens or molecular antigens (Ags) are weak immunogens when applied to intact mucosa. This poor immunogenicity can potentially be overcome by adjuvants, however, most conventional mucosal adjuvants possess unfavorable safety profiles. Moreover, the immune mechanisms of protection against many mucosal infections are not well understood. One case in point is *Chlamydia trachomatis* (*Ct*), a sexually transmitted intracellular pathogen that can cause mucosal infections resulting in female infertility and ectopic pregnancies, as well as blindness in the developing world and pneumonia in infants. In mice, genital *Ct* infection induces protective immunity that is thought to depend on interferon-γ (IFN-γ) producing CD4 T cells. By contrast, we observed that mucosal exposure to UV-inactivated *Ct* (UV-*Ct*) generates tolerogenic *Ct*-specific regulatory T cells, resulting in exacerbated bacterial burden upon subsequent *Ct* infection. Here, we show that mucosal immunization with UV-*Ct* complexed with charge-switching synthetic adjuvant particles (cSAP) did not exert the tolerogenic effect of UV-*Ct* alone but elicited long-lived protection against genital *Ct* infection. This differential effect of UV-*Ct*-cSAP versus UV-*Ct* was because the former was preferentially presented by immunogenic $CD11b^{+}CD103^{-}$ dendritic cells (DCs), while the latter was primarily acquired by tolerogenic $CD11b^{-}CD103^{+}$ DCs. Notably, genital protection was achieved after either intrauterine (i.u.) or intranasal (i.n.), but not subcutaneous (s.c.) immunization with UV-*Ct*-cSAP and was inducible in conventional and humanized mice alike. Regardless of vaccination route, UV-*Ct*-cSAP induced robust circulating and splenic *Ct*-specific IFN-γ^{+} memory CD4 cells. However, only mucosal vaccination, like mucosal infection with live *Ct*, induced an early wave of *Ct*-specific CD4 memory cells that established long-term residence in the genital mucosa. Antibody inhibition experiments and studies in parabiotic mice showed that in the absence of early mucosal seeding by tissue-resident memory cells, mice were poorly protected against *Ct*, even when circulating memory cells were abundant. However, for optimal clearance of *Ct*, a second memory cell wave needed to be recruited to the infected tissue from the circulating pool. Thus, using a novel platform for mucosal immunization, we demonstrate that protection against *Ct* depends on the synergistic action of two discrete memory T cell subsets with distinct differentiation kinetics and migratory properties.

SESSION VII

Importance of CD8 T cells for protection against heterotypic dengue virus infection and antibody-mediated severe dengue disease

Raphael Zellweger, William Eddy, William Tang, Sujan Shresta

LJI, USA

The four serotypes of dengue virus cause pathologies ranging from the febrile illness dengue fever to the potentially lethal severe dengue disease. A major risk factor for developing severe disease is the presence of sub-neutralizing levels of antibody from a previous infection, or, in the case of infants, from a dengue-immune mother. Despite the risk of antibody-mediated severe dengue disease, the goal of dengue vaccine development efforts has been to induce a neutralizing antibody response against the four dengue serotypes. To date, no vaccine has been licensed, and Phase IIb and III results of the most advanced dengue vaccine candidate in development showed an overall efficacy of only 30% and 57%, respectively. We investigated the role of CD8 T cells in protection against dengue virus infection using mouse models. We demonstrate that CD8 T cells induced by one serotype are protective not only against that serotype (homotypic protection), but also mediate cross-protection against the other serotypes (heterotypic protection). Moreover, we show that an efficient CD8 T cell response can prevent antibody-induced severe dengue disease. Our data imply that a vaccine strategy that induces efficient CD8 T cell responses reduce the risk of antibody-mediated severe dengue disease and increase protection across all serotypes. Therefore, a vaccine that triggers both arms of the immune system may be both safer and more efficient than those that focus on humoral responses alone.

SESSION VII

PQBP1 is an Innate Immune Receptor for HIV-1

Sunnie Yoh, Monika Schneider, Stephen Soonthornvacharin, Rana E. Akleh, Kevin C. Olivieri, Paul De Jesus, Chunhai Ruan, Elisa de Castro, Pedro A. Ruiz, Adolfo Garcia-Sastre, Sumit K. Chanda

Innate immune responses that trigger type-I interferon (IFN) secretion have been implicated in HIV-1 transmission and pathogenesis. HIV-1 has evolved countermeasures to escape the activities of several IFN-stimulated genes (ISGs), and those mechanisms not disabled by the virus define both cell type and species tropisms. Recent data suggest that one or more intrinsic signaling pathways sense invariant features encoded by HIV-1 and initiate innate immune responses, including IFN secretion. This response is proximally mediated by recognition of specific viral components in infected cells by pattern recognition receptors (PRR), resulting in the activation of transcription factors that participate in ISG expression and IFN synthesis, such as IRF3. Dendritic cells (DCs) play a critical role in the immune response to viral infection through the facilitation of cell intrinsic antiviral activity and the activation of adaptive immunity. Productive infection of DCs by HIV-1 triggers an IRF3-dependent innate immune response, which has recently been shown to require the activity of cyclic GAMP synthase (cGAS). Here, we report the results of a targeted RNAi screen utilizing primary human monocyte-derived DCs (MDDCs) to identify cellular immune regulators that directly interface with HIV-1-encoded features to initiate this innate response. One of the strongest candidates that emerged from this screen was polyglutamine binding protein 1 (PQBP1). We found that PQBP1 is required for IRF3-dependent signaling in HIV-1-infected MDDCs and human cell lines. PQBP1 directly binds to reverse-transcribed HIV-1 DNA and interacts with cGAS to initiate an IRF3-dependent innate response in myeloid cells. Our results demonstrate that PQBP1 acts as a proximal sensor of a pathogen-associated molecular pattern (PAMP) encoded by HIV-1 DNA to activate cGAS/IRF3-dependent antiviral responses.

SESSION VII

HIV vaccine targets identified by broadly neutralizing antibodies

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Highly antigenically variable viruses such as HIV present huge problems for vaccine design. Broadly neutralizing antibodies to HIV generated during natural infection can identify weaknesses in the surface structures of the virus. These weaknesses can help guide vaccine and drug design and reveal fascinating aspects of the interplay between two highly mutable systems-the virus and antibody.

Poster Session

POSTER SESSION

Notch signaling promotes Tfh memory cell generation by facilitating migration into survival niche

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T follicular helper (Tfh) cells in germinal centers (GCs) facilitate antibody responses by secreted cytokines and direct interactions with B cells. It has been suggested that memory Tfh cells are generated and contribute to a secondary humoral response. Thus, to clarify the mechanisms for generation of memory Tfh cells is relevant for effective vaccine design. Here we demonstrate that Notch signaling regulates generation of memory Tfh cells. During contraction phase, up-regulation of CCR7 and down-regulation of CXCR5 brings Tfh cells into the T-B border area, where they are maintained as central memory (T_{CM}) cells. Lack of Notch signaling during initial antigen priming, but not at the contraction phase, led to a failure of memory T cell generation due to abnormal migration caused by defective CCR7 up-regulation. Notch signaling regulated CCR7 expression through suppression of *Prdm1* (Blimp-1) expression. Our findings indicate that Notch signaling is critical for the generation and function of T_{CM} cells and could be a future target for the vaccination strategy.

GANP-mediated APOBEC3G targeting in generation of non-infectious HIV-1viruses

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GANP was designated as a mammalian homologue of the yeast prototype component of transcription/export-2 complex that is essential for mRNA export, implicating its potentiality in transcription and RNA metabolism. GANP is associated with cytidine deaminase AID and involved in AID-targeting toward the rearranged IgV-region gene for Ab-affinity maturation in B cells. Here, we demonstrate that GANP is upregulated in activated $CD4^+$ T cells and interacted with APOBEC3G (A3G) protein, another member of AID/APOBEC family. Upon HIV-1 infection, GANP augments targeting of A3G toward HIV-1 genomic RNA in the cores of secreted virions. This enhances mutation at A:T pairs of the reverse transcribed HIV-1 genomic cDNA, producing non-infectious HIV-1 virus particles. These results clarified the regulatory role of GANP in targeting of cytidine deaminase AID/APOBEC family proteins against the exogenous genetic invasion and also in protection of the host genome from DNA damages. Efficient targeting with APOBEC cytidine deaminase would be essential for generation of non-infectious viruses *in vivo*, not only for innate immunity but also for eliciting acquired immunological memory.

POSTER SESSION

TNF receptor-associated factor 5 inhibits Th17 differentiation by antagonizing IL-6-receptor signaling

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TNF receptor-associated factors (TRAFs) were initially discovered as adaptor proteins that couple the TNF receptor superfamily to downstream signaling pathways, but TRAFs can regulate other pathways, such as from the Toll-like receptor family, the interleukin-1 receptor family, and the RIG-I-like receptor family. This suggests that TRAFs have a variety of functions in various contexts. We found that in the presence of IL-6 *Traf5*$^{-/-}$ naïve CD4^{+} T cells displayed an increased ability to differentiate into Th17 cells. Consistent with this, Th17-associated experimental autoimmune encephalomyelitis (EAE) was greatly exaggerated in *Traf5*$^{-/-}$ mice. TRAF5 constitutively associated with a cytoplasmic region in gp130 that overlaps with the STAT3 binding site and contains SXXE motif and two di-acidic amino acids, which are recognition elements for the TRAF-C domain. The binding between TRAF5 and gp130 suppressed the recruitment and activation of STAT3 in response to IL-6. Our results uncover an unexpected molecular function of TRAF5. We are currently examining if other TRAF proteins have a similar function in CD4^{+} T cell development.

POSTER SESSION

Role of Sox5 in the development of Th17 cells

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Stat3 signaling is essential for the induction of RORγt and subsequent Th17 cell differentiation. However, the downstream targets of Stat3 for RORγt expression remain largely unknown. We show here that Sox5, named Sox5t, is induced in Th17 cells in a Stat3-dependent manner. In vivo, T cell-specific Sox5-deficient mice exhibit impaired Th17 cell differentiation and are resistant to experimental autoimmune encephalomyelitis. Retrovirus-mediated induction of Sox5 together with c-Maf induces Th17 cell differentiation even in Stat3-deficient $CD4^+$ T cells but not in RORγt-deficient $CD4^+$ T cells, suggesting that Sox5t functions in downstream of IL-6-Stat3 signaling and upstream of RORγt expression during Th17 cell differentiation. Sox5 physically associates with c-Maf via the HMG domain of Sox5 and DNA-binding domain of c-Maf, and Sox5 together with c-Maf directly activates the promoter of RORγt in $CD4^+$ T cells. Taken together, our results suggest that Sox5 and c-Maf cooperatively induce Th17 cell differentiation via the induction of RORγt as downstream targets of Stat3.

POSTER SESSION

C-terminal fragment of *Clostridium perfringens* enterotoxin is an efficient vaccine antigen delivery system for nasal pneumococcal vaccine.

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Mucosal vaccine can induce both systemic and mucosal immune responses and thus is considered to be an ideal vaccine for infectious diseases. In general, the efficient antigen delivery to mucosal tissues, especially mucosa-associated lymphoid tissues (MALTs) is essential for the development of mucosal vaccine. We previously found that claudin-4 was highly expressed in nasopharynx-associated lymphoid tissue (NALT) and claudin-4-targeting using C-terminal fragment of *Clostridium perfringens* enterotoxin (C-CPE) was an efficient antigen delivery system to NALT. In this study, we applied C-CPE to mucosal vaccine against pneumonoccal infection. Pneumococcal surface protein of A (PspA) is commonly expressed in *S. pneumoniae* and considered to be a vaccine candidate. We genetically fused PspA and C-CPE (PspA-C-CPE) and confirmed that PspA-C-CPE bound to claudin-4. Nasal immunization with PspA-C-CPE can induce PspA-specific systemic and mucosal immune responses. Additionally, PspA-C-CPE can elicit protective immunity against pneumococcal infection. These findings indicate that C-CPE is an efficient antigen delivery system for pneumococcal infection.

POSTER SESSION

Secondary IgM Antibody Response is Elicited from Low-affinity IgM+ Memory B cells but not High-affinity Ones.

Yasuyuki Tashiro[1,2], Yasushi Hara[1], Ryo Goitsuka[2] and Takachika Azuma[1]

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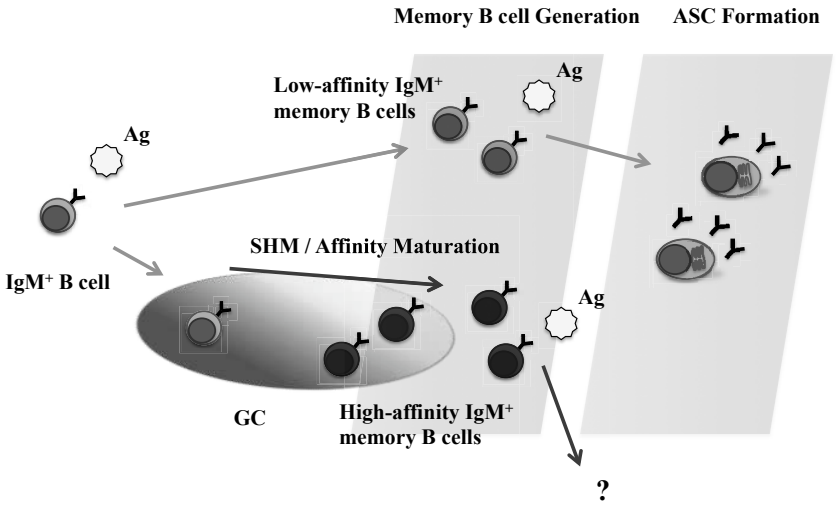
Germinal center (GC) is considered as the site for class switch recombination(CSR) and somatic hypermutation(SHM) in T-dependent (TD) response. Since IgG^+ memory B cells and Antibody Secreting Cells (ASCs) possess heavily mutated *Ig* genes, IgG^+ B cells develop mainly through GC-dependent pathway.

Recently it was reported that IgM^+ memory B cells were also generated in TD immune response. IgM^+ memory B cells were mainly composed of clones with few SHMs and, therefore, their development was independent on GC. Here we report that IgM^+ GC B cells possessing numerous *Ig* mutations resided in GC without maintaining IgM isotype usage. These IgM^+ GC B cells underwent affinity maturation, followed by memory B-cell development. On the other hand, low-affinity IgM^+ memory B cells were also generated abundantly as reported previously. Surprisingly, high-affinity IgM+ GC B cells had poor potential to develop into ASCs although high-affinity memory B cells were normally generated. In addition to GC B cells, high-affinity IgM+ memory B cells showed no contribution to secondary IgM antibody response in contrast to normal production of low-affinity IgM antibodies.

Secondary IgM response is elicited from low-affinity memory B cells but not high-affinity ones, suggesting that these two types of IgM+ memory B cells play distinct roles in secondary response. It remains elusive how high-affinity IgM^+ memory B cells function during the secondary response.



POSTER SESSION

C-type lectin Dectin-2 is a direct receptor for mycobacterial lipoarabinomannan that mediates its adjuvanticity.

Akiko Yonekawa[1,2], Sho Yamasaki[1]

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Mycobacteria possess various characteristic cell-wall components, which affect the host immune responses. Mannose-capped lipoarabinomannan (Man-LAM) is a lipoglycan derived from pathogenic mycobacteira and has long been known to have both inhibitory and stimulatory activities on host immunity. However, the direct Man-LAM receptor that explains its bidirectional effects have not been clearly identified.

Here, we show that a C-type lectin receptor Dectin-2 is a direct receptor for Man-LAM. Man-LAM activated bone marrow-derived dendritic cells (BMDCs) to produce TNF, whereas it was completely abrogated in Dectin-$2^{-/-}$ BMDCs. In addition, the IL-10 and IL-2 production was uniquely induced by Man-LAM in a Dectin-2-dependent manner. Furthermore, Man-LAM stimulation promoted antigen-presenting cell functions through Dectin-2 on BMDCs. In a murine model of autoimmune disease, Man-LAM actually induced experimental autoimmune encephalomyelitis (EAE) as an adjuvant through Dectin-2-FcRγ signaling axis.

These results demonstrate that Dectin-2 recognizes Man-LAM and play crucial roles in mediating the production of multiple cytokines and *in vivo* adjuvant activity.

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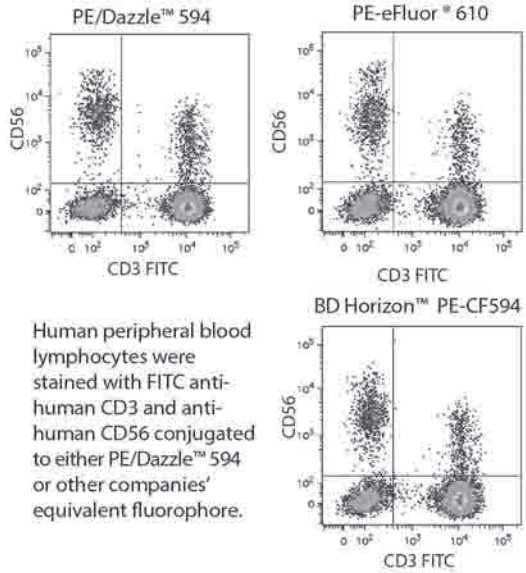

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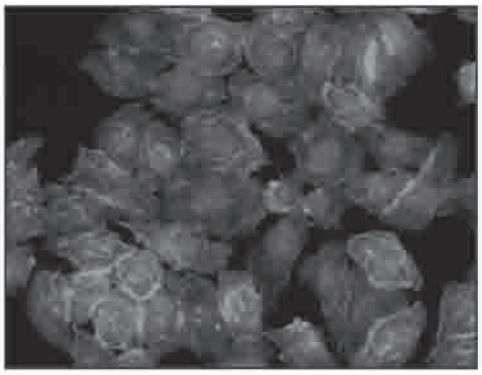

HeLa cells were fixed, permeabilized, and blocked, then intracellularly stained with Ki-67 (clone Ki-67) Alexa Fluor® 594 (*red*) and Alexa Fluor® 488 Phalloidin (*green*). Nuclei were counterstained with DAPI (*blue*). The image was captured with 40x objective.

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