14th Annual
Upstate
New York
Immunology
Conference
2011

Corporate Sponsor
BD Biosciences

Grant Funding
NIH
National Institute of
Allergy and Infectious
Diseases

The Sagamore Resort and Conference Center
Bolton Landing, NY
October 23-25, 2011
# Table of Contents

The Conference/The Venue ................................................................. 5

Schedule of Events ........................................................................... 6

Platinum Corporate Sponsor
   BD Biosciences ............................................................................. 15

Silver Corporate Sponsors
   BioLegend ..................................................................................... 18
   Cell Signaling Technologies ....................................................... 19
   eBioscience ................................................................................ 20

Bronze Corporate Sponsors
   Krackeler Scientific, Inc. ........................................................... 21
   Lonza Walkersville ....................................................................... 21
   New England BioLabs .................................................................. 21
   Stem Cell Technologies ............................................................... 21

NYIC Scientific Advisory Board ....................................................... 22

Institutional Financial Supporters ..................................................... 23

Grant Funding ................................................................................... 24

Keynote Speaker
   Ann Rothstein, Ph.D. ................................................................. 26

Session I
   Inflammation and Cancer ........................................................... 27

Session II
Featured Speaker and Selected Oral Poster Presentations ........31

Session III
Regulation of Immune Responses ..................................37

Session IV
Featured Speaker and Selected Oral Poster Presentations .......43

Session V
Virus-Host Interactions ...............................................49

Dinner Presentation
Darci Gorgone—BD Biosciences ....................................52

Session VI
Inflammation .............................................................53

Session VII
Immune Response to Pathogens ....................................57

Poster Abstracts ............................................................63

Attendee Contact Information ..........................................86

Authors Index ..............................................................91

Exit Survey .................................................................94

Upstate New York Immunology Conference
Center for Immunology & Microbial Disease
Albany Medical College
47 New Scotland Ave., MC-151, Albany, NY 12208
518-262-5365
Fax: 518-262-5748
www.amc.edu/NYIC/index.html
THE UPSTATE NEW YORK IMMUNOLOGY CONFERENCE (NYIC)

Do you remember Garnet Hill? Many of you do! This meeting started in 1997 as a small retreat to facilitate interactions among young scientists, institutions, and renowned experts in the field of Immunology. In just a few short years, the number of attendees grew and a larger venue was needed to meet the future needs of the Conference.

Although there have been many changes, one simple principle has remained: To provide an opportunity for young and senior scientists to gather in a setting that is diverse enough to meet the needs of all attendees while remaining small enough to allow for personal interactions. While always challenging, it is the goal of the NYIC Scientific Advisory Board and the NYIC Conference Organizers to give graduate students and postdoctoral fellows the opportunity to present their research and engage in conversations that will stimulate further discussions, collaborations and interest in pursuing a new or different way of looking at their research.

We hope you share our enthusiasm and enjoy your time with us!

THE SAGAMORE RESORT

The Sagamore Resort and Conference Center celebrated it’s 125 year anniversary in 2008. If this is your first visit to the Resort, take some time to enjoy the beauty that surrounds you. Although we may have missed the grandeur of the Fall colors, there are still breath-taking views to be seen.

The staff are friendly, courteous, and hard-working. If you require any information or have a special need, please see either Dawn Bellville, Administrative Conference Coordinator, or any of the Resort personnel. Many thanks to Lori Rehm (Assistant Director of Group Sales), Derrick Hammond (Conference Services Manager), Don Vilmar (Banquet Manager), Joel Clark (Function Set-up Manager) and his amazing crew, Dave Reynolds (CMI Communications-Audio/Visual Manager) and his crew, and all of the associates who attend to our every need. Thank you!
Schedule of Events

Sunday, October 23rd

2:00-3:30 p.m.  Conference Registration
               (Conference Center Lobby)

3:00-6:00 p.m.  Hotel Check-In
               (Main Hotel Lobby)

3:00-4:00 p.m.  Welcome Cocktail Reception
               (Conference Center Foyer)

4:00-5:00 p.m.  Keynote Speaker
               (Bellvue)

   Introduction by: Kate MacNamara, Ph.D.

   Ann Marshak-Rothstein, Ph.D.
   Professor, School of Medicine
   Rheumatology
   Graduate School of Biomedical Sciences
   Immunology & Virology Program
   University of Massachusetts

   “Distinct Features of BCR/TLR7 and BCR/TLR9 Activation of Autoreactive B-cells”

5:00-6:00 p.m.  Free Time

6:00-7:00 p.m.  Dinner
               (Wapanak)

7:00-8:00 p.m.  Session I
               (Bellvue)

   Inflammation and Cancer
   Chair: Edith Lord, Ph.D.
7:00-7:20  
**Steven Fiering, Ph.D.**  
Dartmouth Medical School  
“Host Cell Manipulation in the Tumor Microenvironment by Nonreplicating *Toxoplasma gondii* Abrogates Tumor-induced Immunosuppression and Rejects Established Aggressive Ovarian Cancer”

7:20-7:40  
**Shu Shien Chin, B.S.**  
University at Buffalo  
“If Mutational Analysis to Identify Amino Acids Necessary for Ets-1 Activity in Blocking Plasma Cell Differentiation”

7:40-8:00  
**Scott A. Gerber, Ph.D.**  
University of Rochester  
“If Radiation Therapy for Cancer: The Essential Role of IFN-γ and the Immune System”

Enjoy the rest of the evening with colleagues  
at Mr. Brown’s Pub, Caldwell’s Lounge, or The Veranda.

---

**Monday, October 24th**

7:00-8:15 a.m.  
**Breakfast at Leisure**  
(LA Bella Vita, Main Hotel)

8:30-10:00 a.m.  
**Session II**  
*Featured Speaker & Selected Oral Poster Presentations*  
Chair: Kelvin P. Lee, M.D.

8:30-8:50  
**Kelvin P. Lee, M.D.**  
Roswell Park Cancer Institute  
“If CD28 Plays an Intrinsic Role in the Survival of Normal and Malignant Plasma Cells”
8:50-9:05  **Katrina Simmons, M.S.**  
School of Public Health/University at Albany  
“Effects of Vitamin D on Mammary Epithelial Cell Synthesis and Secretion of CD14”

9:05-9:20  **Joanne Lim, M.S.**  
University of Rochester  
“Exploring the Role of Type I Interferons in the Induction of Antitumor Response Following Radiation Treatment”

9:20-9:35  **Jeffrey H. Mills, Ph.D.**  
Cornell University  
“Extracellular Adenosine as a Master Regulator of Immune Cell Migration into the Central Nervous System via Induction of CX3CL1”

9:35-9:50  **Debarati Banik, M.S.**  
Roswell Park Cancer Institute  
“Role of MMP3 in Tumor Progression and IRF8 Mediated Regulation of MMP3 Function”

10:00-10:20 a.m.  **Break**  
(Conference Center Foyer)

10:20-12:00 p.m.  **Session III**  
**Regulation of Immune Responses**  
Chair: Margaret S. Bynoe, Ph.D.

10:20-10:40  **Katelyn T. Byrne, B.S.**  
Dartmouth Medical School  
“The Role of Autoimmunity in Maintaining CD8 Memory T-cell Responses to B16 Melanoma ”

10:40-11:00  **Erik L. Brincks, Ph.D.**  
Trudeau Institute  
“Memory T_{reg} cells Regulate the Immune Response to Influenza Virus Infection”
11:00-11:20  Margaret S. Bynoe, Ph.D.
Cornell University
“Epicutaneous (Skin) Immunization with Auto-antigen Induces Neuropilin-1-mediated Suppressor in CD4 T-cells”

11:20-11:40  Vandana Kalia, Ph.D.
Pennsylvania State University
“Mechanisms Regulating CD8 T-cell Longevity and Function”

11:40-12:00  Jamie L. Harden, Ph.D.
University at Buffalo
“Induction and Activation of the Aryl-Hydrocarbon-Receptor Maintains an IDO-mediated Tolerogenic Tumor Microenvironment”

12:15-1:00  Lunch
(Wapanak)

1:00-2:00 p.m.  Display Posters and Free Time

2:00-3:30 p.m.  Session IV
(Bellvue)  Featured Speaker & Selected Oral Poster Presentations
Chair: Sharon S. Evans, Ph.D.

2:00-2:20  Sharon S. Evans, Ph.D.
Roswell Park Cancer Institute
“IL-6 Trans-signaling Licenses Tumor Microvascular Gateways for Trafficking of Cytotoxic T-cells”

2:20-2:35  Julie S. Lefebvre, Ph.D.
Trudeau Institute
“Impairment of CD4+ T-cell Cognate Helper Functions in Response to Influenza Immunization with Aging

2:35-2:50  Anastasiya Yermakova, B.S.
Wadsworth Center
“Mechanism of Neutralizing Antibodies Against the Binding Subunit (RTB) of Ricin”
2:50-3:05  
Elizabeth M. Samuelson, B.S.
SUNY Upstate Medical University
“Blk-Haploinsufficiency Impairs the Development, but Enhances the Functional Responses, of MZ B-cells”

3:05-3:20  
Jennifer Yates, B.S.
Wadsworth Center
“Identification of a Putative IgM Memory B-cell Population During Bacterial Infection”

3:30-3:50 p.m.  
Break
(Conference Center Foyer)

3:50-4:30 p.m.  
Session V
Virus-Host Interactions
Chair: Christopher Norbury, Ph.D.

3:50-4:10  
Gary C. Chan, Ph.D.
SUNY Upstate Medical University
“Human Cytomegalovirus Reprograms Monocytes to Function as Vehicles of Viral Dissemination”

4:10-4:30  
Christine A. King, Ph.D.
SUNY Upstate Medical University
“Kaposi Sarcoma Associated Herpesvirus Infection of Primary Human Endothelial Cells Activates the Proto-oncogene STAT3”

4:30-5:30 p.m.  
Cocktail Reception & Poster Preview
(Nirvana/Wapanak)

5:30-6:30 p.m.  
Vendor Fair & Odd Number Poster Presentations
(Nirvana/Wapanak)

6:30-7:30 p.m.  
Vendor Fair & Even Number Poster Presentations
(Nirvana/Wapanak)
7:30-8:00 p.m.    Dinner
(Wapanak)

8:00-8:30 p.m.    Dessert & Corporate Presentation
Introduction by: James R. Drake, Ph.D.

Darci Gorgone, M.A.
Technical Applications Specialist
BD Biosciences

“Multicolor Flow Cytometry: Optimizing Instrument Settings”

Tuesday, October 25th

Please check-out of your room by 11:00 a.m. or you will be charged a late check-out fee of $75 on your personal credit card.

7:00-8:45 a.m.    Breakfast at Leisure

9:00-10:00 a.m.   Session VI
Inflammation
Chair: Ann Marshak-Rothstein, Ph.D.

9:00-9:20    Yoichi Furuya, Ph.D.
Albany Medical College
“Susceptibility of Asthmatic Mice to Influenza A Virus Challenge”

9:20-9:40    Eric Y. Denkers, Ph.D.
Cornell University
“Insights into Inflammatory Bowel Disease Using Toxoplasma gondii as an Infectious Trigger”
9:40-10:00  
**Michael R. Elliott, Ph.D.**  
University of Rochester  
“Apoptotic Cell ‘Find-me’ Signals in Cell Clearance and Inflammation”

10:00-10:30 a.m.  
**Break and Hotel Check out**

10:30-12:00 p.m.  
**Session VII**  
*Immune Response to Pathogens*  
Chair: Gary Winslow, Ph.D.

10:30-10:50  
**Keke C. Fairfax, Ph.D.**  
Trudeau Institute  
“IL-10 Maintains a Pool of Hepatic B-cells that Prevents Development of Severe Cardiopulmonary Disease in Schistosomiasis”

10:50-11:10  
**Bikash Sahay, Ph.D.**  
Albany Medical College  
“cAMP-dependent IL-10 Production Determines the Severity of Lyme Disease”

11:10-11:30  
**Derek D. Jones, B.S.**  
Wadsworth Center  
“Induction of a Polyreactive IgM Response by Bacterial Infection”

11:30-11:50  
**Girish S. Kirimanjeswara, D.V.M., Ph.D.**  
Pennsylvania State University  
“Autophagy-mediated Defense Against Francisella tularensis”

11:50-12:10  
**Jayaleka J. Amarasinghe, Ph.D.**  
Wadsworth Center  
“Interactions Between Salmonella enterica serovar Typhimurium and Secretory IgA at Epithelial Surfaces”
12:15-12:30 p.m.  Closing Remarks

12:30-2:00 p.m.  Lunch
    (Shelving Rock)

*Depart from Conference*

Hope to see you at the
15th Annual Upstate New York Immunology Conference

**October 21-24, 2012**
The Sagamore Resort & Conference Center
Platinum Corporate Sponsor

BD Biosciences
Introducing BD FACSVerse™

What’s really exciting is what’s behind it.

The new BD FACSVerse™ flow cytometer is the latest in the family of high-performance analyzers that incorporates the best of our thinking and the very latest advances in technology to simplify your workflow today and tomorrow.

The innovations built into the new BD FACSVerse are on everyone’s wish list, from ingenious automation to unparalleled flexibility, so you can start strong today and grow tomorrow. Take a good look, there’s quite a lot to see.

For more information, please visit bdbiosciences.com/verse.

Simply brilliant.

Class 1 Laser Product.
For Research Use Only. Not for use in diagnostic or therapeutic procedures.
BD, BD Logo and all other trademarks are property of Becton, Dickinson and Company. © 2011 BD
23-13498-00

BD Biosciences
2350 Quine Drive
San Jose, CA 95131
bdbiosciences.com
Silver Corporate Sponsors

BioLegend
Cell Signaling Technologies
eBioscience
Brilliant Violet 421™ and Brilliant Violet 570™
Two Brilliant Choices for Multi-Color Flow Cytometry

Introducing Brilliant Violet 570™
- Excellent signal-to-noise
- Intracellular staining compatible, non-nanocrystal
- Stable to fixation

Brilliant Violet 570™ is the latest member of the Brilliant Violet™ family of novel highly fluorescent polymers excitable by the 405 nm violet laser.

Joining Brilliant Violet 421™, our exceptionally bright alternative to Pacific Blue™, Brilliant Violet 570™ brings new choice and capability to the violet laser.

Stain Index of BV570™ and comparables

<table>
<thead>
<tr>
<th>Specificity/Clone</th>
<th>Fluorochrome</th>
<th>Laser Excitation</th>
<th>Filter</th>
<th>Stain Index</th>
</tr>
</thead>
<tbody>
<tr>
<td>CD8/RPA-T8</td>
<td>Brilliant Violet 570™</td>
<td>405 nm</td>
<td>585/42</td>
<td>145</td>
</tr>
<tr>
<td>CD8/RPA-T8</td>
<td>Pacific Orange™</td>
<td>405 nm</td>
<td>585/42</td>
<td>27</td>
</tr>
<tr>
<td>CD4/PanT Cell</td>
<td>BD Horizon™ V500</td>
<td>405 nm</td>
<td>520/50</td>
<td>23</td>
</tr>
<tr>
<td>CD45IUCHT1</td>
<td>Brilliant Violet 570™</td>
<td>405 nm</td>
<td>585/42</td>
<td>66</td>
</tr>
<tr>
<td>CD45IUCHT1</td>
<td>Pacific Orange™</td>
<td>405 nm</td>
<td>585/42</td>
<td>10</td>
</tr>
</tbody>
</table>

Human RBC-lyzed whole blood cells were stained with anti-CD8 or anti-CD3 conjugated to the above fluorochromes and run on the BD™ LSR II flow cytometer. The stain index values indicated are derived at the optimal concentration for each conjugate.
Antibodies and related reagents for

Immunology Research

...from Cell Signaling Technology

Unparalleled product quality, validation and technical support.

:: The highest quality antibodies provide you with the brightest signal, lowest background and greatest reproducibility over time.

:: Extensive in-house validation means that optimization is not left up to you, the user.

:: Technical support provided by the same scientists who produce and validate the antibodies — this translates into a thorough, fast and accurate response.

Flow cytometric analysis of: A. Jurkat cells, untreated (blue) or IFN-α treated (green), using Phospho-Stat3 (Tyr705) (D3A7) Rabbit mAb (Alexa Fluor® 488 Conjugate) #4323 compared to nonspecific control antibody (red). B. K-562 cells, imatinib-treated (blue) or untreated (green), using Phospho-Stat5 (Tyr694) (C71E5) Rabbit mAb #9314 compared to concentration matched Rabbit (DA1E) mAb IgG Isotype Control (red). C. whole blood using CD44 (156-3C11) Mouse mAb.

for quality products you can trust...

www.cellsignal.com
Capture Apoptosis

... from beginning to end

Make It Happen – Functional antibodies and recombinant protein ligands from multiple death receptor pathways; perfect for cell-based assays.

See It Early – Define first events with Annexin-V in over 30 formats, sizes and configurations.

See It All – Choose from hundreds of relevant antibodies and novel immunoassays including Instant ELISA®, Platinum ELISA, and FlowCytomix™ multiplexing kits.

Wrap Up Loose Ends – Capture ssDNA and DNA fragmentation events with kits and reagents suitable for both flow cytometry and immunohistochemistry.

Visit eBioscience.com/cell-apoptosis for a complete list of reagents for your workflow.
Bronze Corporate Sponsors

Krackeler Scientific, Inc.

Lonza Walkersville, Inc.

New England BioLabs, Inc.

Stem Cell Technologies, Inc.
NYIC Scientific Advisory Board
Institutional Representatives

Albany Medical College
Jim Drake, Kate MacNamara
(NYIC Conference Organizers)

Cornell University
Margaret Bynoe

Dartmouth College
Brent Berwin

Pennsylvania State University
Chris Norbury, Eric Harvill

Roswell Park Cancer Institute
Yasmin Thanavala

SUNY Upstate Medical University
Michael Princiotta

Trudeau Institute
Laura Haynes

University at Buffalo
Nejat Egilmez

University of Rochester Medical Center
Edith Lord

Wadsworth Center
Gary Winslow
Institutional Financial Supporters

Albany Medical College
Alumni Association

Cornell University
Department of Microbiology & Immunology

Dartmouth College
Department of Microbiology & Immunology

Pennsylvania State University
Department of Veterinary and Biomedical Sciences

Roswell Park Cancer Institute
Department of Immunology

SUNY Upstate Medical University
Microbiology & Immunology Program

Trudeau Institute

University at Buffalo
Department of Microbiology & Immunology

University of Rochester Medical Center
Department of Microbiology & Immunology
For grant support provided to Graduate Students and Postdoctoral Fellows

“Thank You”

National Institute of Allergy and Infectious Diseases

R13AI051522
Keynote Speaker
In vitro studies with B cells that express a prototypic autoreactive BCR, AM14, have shown that many of the canonical autoantigens targeted in patients or mice with systemic autoimmune disease are potent endogenous autoadjuvants. These mice express a low affinity BCR specific for autologous IgG2a and are potently activated by DNA- or RNA-associated immune complexes through a mechanism dependent on both the BCR and either TLR9 or TLR7. A number of groups have now tested this BCR/TLR paradigm in vivo. As expected, TLR9-/- autoimmune-prone mice fail to produce autoantibodies directed against chromatin, while TLR7-/- mice fail to produce autoantibodies directed against numerous RNA-associated proteins. However, the TLR9-/- mice develop accelerated clinical disease, while the TLR7-/- mice exhibit remarkably prolonged survival. To better understand this dichotomy, we have carefully compared the AM14 B cell response to DNA and RNA ICs. The AM14 B cells stimulated with DNA ICs enter cell cycle within 24 hrs, but shortly thereafter they coordinately succumb to a form of programmed cell. By contrast, the response to RNA ICs is more delayed, but the B cells then continue to divide for an extended period of time. B cells stimulated with a combination of PA4 and BWR4 are protected from programmed cell death. Together these data suggest that TLR9 may limit autoreactive B cell responses through a negative feedback loop, while TLR7 promotes a more unrestrained activation pathway.
Session I

Inflammation and Cancer
Host Cell Manipulation in the Tumor Microenvironment by Nonreplicating
*Toxoplasma gondii* Abrogates Tumor-induced Immunosuppression and Rejects
Established Aggressive Ovarian Cancer

Jay Baird, David Bzik, Jose Conejo-Garcia¹, Steven Fiering
Dept. of Microbiology/Immunology, Dartmouth Medical School, Hanover, NH
¹ Wistar Institute, Philadelphia, Pa

Solid tumors develop active mechanisms to impair antitumor immune responses. Breaking
tumor-mediated immunosuppression is therefore required for effective immunotherapies. A
nonreplicating form of the intracellular parasite *Toxoplasma gondii* (CPS) breaks immunosup-
pression at tumor locations and elicits antitumor immune responses that cure established, ag-
gressive, orthotopic ovarian cancer without toxicity. In the tumor microenvironment, CPS se-
lectively infects myeloid cells with phenotypic attributes of dendritic cells (DCs), transforming
them from an immunosuppressive to an immunostimulatory cell type. CPS-mediated DC re-
programming is Myd88-independent, and therefore different from antitumor mechanisms in-
duced by other intracellular microorganisms such as Listeria. However, tumor rejection requires
the generation of immunostimulatory cytokines by tumor DCs and is associated with enhanced
antigen presentation and increased co-stimulation. Correspondingly, tumor-reactive cytotoxic T
cells able to transfer anti-tumor protection are dramatically expanded at tumor and splenic loca-
tions. Our results provide a mechanistic rationale for using CPS to treat ovarian cancer.
Mutational Analysis to Identify Amino Acids Necessary for Ets-1 Activity in Blocking Plasma Cell Differentiation

Shu Shien Chin, Lisa Russell, Shinu John and Lee Ann Garrett-Sinha
Department of Biochemistry, State University of New York at Buffalo, Buffalo, NY

Differentiation of B cells into immunoglobulin-secreting plasma cells is a tightly and intricately regulated program under the control of various transcription factors, such as Ets-1 and Blimp-1. Ets-1 and Blimp-1 have opposing functions in regulating B cell differentiation. By suppressing key genes involved in the mature B cell program, Blimp-1 is promotes the transition of mature B cells into terminally-differentiated immunoglobulin-secreting plasma cells. On the other hand, Ets-1 plays a negative role in controlling the plasmacytic differentiation, as we have previously shown that Ets-1 knockout mice exhibit dramatic increase in plasma cells and forced expression of Ets-1 in B cells prevents their differentiation to plasma cells. We further demonstrated that the mechanism underlying the counterbalancing effects of Ets-1 and Blimp-1 is a result of Ets-1’s ability to engage two separate yet synchronizing actions against Blimp-1: (1) Ets-1 inhibits DNA binding ability of Blimp-1 by a direct physical interaction between the two transcription factors and (2) Ets-1 stimulates expression of critical genes that promote mature B cell differentiation, which some of them are repressed by Blimp-1. Interestingly Ets-1 executes both of these two distinct tasks through the Ets domain (amino acids 331-415), a highly conserved DNA binding domain of Ets family transcription factors. We sought to identify specific amino acids in the Ets domain that contribute to DNA binding without disrupting interaction with Blimp-1 and vice versa. We have so far identified two Ets-1 mutants (I401E and L342A) that appear to have the desired properties. Mutation of isoleucine to glutamic acid at position 401 in the Ets domain greatly diminishes Ets-1 DNA binding ability, but does not block Blimp-1 binding to DNA. Conversely, mutation of leucine to alanine at residue 342 in the Ets domain retains its ability to bind DNA and yet fails to inhibit Blimp-1 DNA binding function. We are now testing the effects to these two point mutations on Ets-1 inhibition of plasmacytic differentiation. This will allow us to test the relative roles of the two pathways described above in the overall ability of Ets-1 to block plasma cell differentiation. This information may prove useful in future studies to define compounds that can modulate Ets-1 activity to control B cell differentiation pathways.
Radiation Therapy for Cancer: The Essential Role of IFNγ and the Immune System

Scott A. Gerber
University of Rochester Medical Center

Radiation (IR) therapy has long been an effective cancer treatment acting primarily through the induction of tumor cell damage at a molecular level. However, a new concept is emerging suggesting that the immune system plays a pivotal role in determining the effectiveness of IR therapy. We have previously demonstrated that IR therapy promotes antitumor immunity through the release of tumor antigens and induction of a pro-inflammatory tumor microenvironment.

Our work here has identified IFNγ as an essential cytokine, which mediates the efficacy of IR therapy. Local IR (15Gy) to mice bearing Colon38, a colon adenocarcinoma, resulted in decreased tumor burden in wild type animals. Interestingly, IR therapy had no effect on tumor burden in IFNγKO mice. We further determined that intratumoral (i.t.) levels of IFNγ spiked two days following IR, which directly correlated with a decrease in tumor burden that was not a result of direct cytotoxic effects of IFNγ on tumor cells. CD8+ T cells were the predominant producers of IFNγ, as demonstrated by IFNγ intracellular staining. Elimination of CD8+ T cells by antibody treatment resulted in a reduction of over 90% of the intratumoral levels of IFNγ. More importantly, knock down of CD8+ T cells completely abrogated the effects of radiation therapy.

We speculate that radiotherapy promotes the production of IFNγ from CD8+ T cells, which then acts back, in an autocrine fashion, to enhance the cytotoxic function of these cells. This concept is further supported as CD8+ T cells from irradiated tumors, isolated by FACSort, expressed higher levels of genes thought to enhance killer function. Our data suggest that IFNγ may play a pivotal role in mediating the antitumor effects of IR therapy. Supported by NIH grant CA28332.
Session II

Featured Speaker and Selected Oral Poster Presentations
CD28 Plays an Intrinsic Role in the Survival of Normal and Malignant Plasma Cells

Cheryl H. Rozanski, Jayakumar Nair, Louise M. Carlson, Chandana Koorella, P. Leif Bergsagel, Asher A. Chanan-Khan, Stephen P. Schoenberger, Lawrence H. Boise, and Kelvin P. Lee

Interactions between malignant plasma cells of multiple myeloma (MM) and stromal cells within the bone marrow (BM) microenvironment are essential for myeloma cell survival, mirroring the same dependence of normal BM-resident long-lived plasma cells (PC, which are responsible for durable humoral immunity) on specific marrow niches. These interactions directly transduce pro-survival signals to the myeloma cells and also induce niche production of supportive soluble factors. However, despite their central importance the specific molecular and cellular components involved remain poorly characterized. We now report that the prototypic T cell costimulatory receptor CD28 is overexpressed on myeloma cells during disease progression and in the poor prognosis subgroups, and plays a previously unrecognized role as a two-way molecular bridge to support myeloid stromal cells in the microenvironment. Engagement by CD28 to its ligand CD80/CD86 on stromal dendritic cell (DC) directly transduces a pro-survival signal to myeloma cell, protecting it against chemotherapy and growth factor withdrawal-induced death. Simultaneously, CD28-mediated ligation of CD80/CD86 induces the stromal DC to produce the pro-survival cytokine IL-6 (involving novel crosstalk with the Notch pathway) and the immunosuppressive enzyme indoleamine 2, 3 dioxygenase (IDO). These findings identify CD28 and CD80/CD86 as important molecular components of the interaction between myeloma cells and the bone marrow microenvironment, and point to similar interaction for normal plasma cells as well as suggesting novel therapeutic strategies to target malignant and pathogenic (e.g. in allergy and autoimmunity) plasma cells. In addition, we have found in normal plasma cell biology that long-term humoral immunity is dependent on plasma cell-intrinsic function of CD28, which selectively supports the survival of BM long-lived PC (LLPC) but not splenic short-lived PC (SLPC). LLPC and SLPC both express CD28, but CD28 signaling with enhanced survival occurred only in the LLPC. In vivo, even with sufficient T cell help the loss of CD28 or its ligands CD80/CD86 caused significant loss of the LLPC population, reduction of LLPC survival t1/2 from 426 to 63 days, and inability to maintain antibody titers long-term - with no effect on SLPC populations. These findings establish the existence of the distinct BM LLPC subset that is necessary to sustain antibody titers, and uncover a central role for CD28 function in the longevity of plasma cells and humoral immunity.
Effects of Vitamin D on Mammary Epithelial Cell Synthesis and Secretion of CD14

Katrina Simmons and JoEllen Welsh
Department of Biomedical Sciences, School of Public Health, University at Albany, Rensselaer, NY

Vitamin D has been shown to decrease the risk for infectious diseases by regulating the immune responses at the molecular level. The majority of the work in this area has focused on the direct effects of vitamin D on immune cells such as macrophages and monocytes. Very few studies have investigated how vitamin D alters the immune environment of epithelial cells in barrier tissues, such as the mammary gland, that are exposed to infectious agents. Through microarray screening of human mammary epithelial cells (hTERT-HME cell line, ATCC #CRL-4010) treated for 24 hours with 100nM 1,25D (the active form of vitamin D), we found an enrichment of genes involved in innate immunity, including CD14 which was elevated more than 60-fold. Based on these findings, we are examining the effects of vitamin D on the synthesis, secretion, and function of CD14 in hTERT-HME cells. CD14 is a pattern recognition receptor that binds lipopolysaccharide (LPS), a microbial cell wall component of gram-negative bacteria. Through quantitative PCR (qPCR), we confirmed that CD14 mRNA was significantly increased in hTERT-HME cells treated with either 1,25D or the circulating metabolite 25 hydroxyvitamin D (25D), which reflects overall vitamin D status. In association with up-regulation of CD14, we found down regulation of the pro-inflammatory cytokines IL-6, TNFα (alpha), and IL-1β (beta) in hTERT-HME cells treated with 25D. We next used a CD14 ELISA to examine whether vitamin D metabolites promoted the secretion of the soluble form of CD14 (sCD14) from mammary cells. Both 1,25D and 25D increased the accumulation of sCD14 in the conditioned media of hTERT HME cells in a dose and time dependent manner. Surprisingly, whereas vitamin D metabolites promoted the secretion of sCD14, purified LPS, a potent inducer of CD14 in the mammary gland and immune and non-immune cells, had no effect. This data suggests that the regulation of CD14 in mammary epithelial cells is distinct from that in other cell types. In conclusion, this data is the first to demonstrate effects of vitamin D on secretion of CD14 in mammary cells and support the central hypothesis that vitamin D promotes an anti-inflammatory environment via secretion of sCD14 from mammary epithelial cells.

Poster No. 3
Exploring the Role of Type I Interferons in the Induction of Antitumor Response Following Radiation Treatment

Joanne Lim, Scott Gerber, and Edith Lord
University of Rochester

Although type I interferons (IFNs-I) play critical roles as factors that protect the host against viral infections, there has been increasing evidence that they are also essential for many immunological processes. Based on previous work by several investigators, there is clear indication that the responsiveness of hematopoietic cells to type I IFNs is important in generating antitumor immunity.

Our lab is interested in examining the importance of endogenous type I IFNs in the induction of antitumor immunity in response to radiation therapy. Using B16 melanoma mouse model, we have previously demonstrated that single high dose radiation therapy results in a dramatic delay in tumor progression, and this in turn is dependent on an increase in IFN-\(\gamma\) within the tumor microenvironment. We hypothesize that endogenous IFNs-I play a role in the induction of IFN-\(\gamma\) response following radiation therapy. We first compared the growth of B16 tumors between wild type (WT) mice and mice that lack functional IFN-\(\alpha/\beta\) receptor (KO). Interestingly, tumor growth was faster in KO mice than in WT mice. When the tumors were treated with 15Gy radiation, tumor progression in both KO and WT mice were slowed, but tumor burdens were still larger in the KO than WT mice. The induction of intratumoral IFN-\(\gamma\) mRNA levels by radiation therapy observed in WT mice, was drastically lower in KO mice. Importantly, the increase in CD8\(^+\) T cells as well as the increase in activation status of CD8\(^+\) T cells occurred only in mice that were able to signal through intact IFN-\(\alpha/\beta\) receptors. Since dendritic cells (DCs) within tumor play a major role in the recruitment and activation of CD8\(^+\) T cells, we examined the activation status of these cells as well. MHC class I molecules, required for presentation of tumor antigen to CD8\(^+\) T cells, were upregulated on the surface of intratumoral DCs of WT mice in response to radiation treatment, but not in mice that lacked IFN-\(\alpha/\beta\) receptors. Further, when intratumoral DCs were FACsorted and analyzed for chemokine mRNA levels, we observed that the relative levels of CXCL10 and CXCL11 were much higher in WT than KO cDCs, especially after radiation treatment. In conclusion, our data suggest that IFNs-I are required for complete activation of intratumoral cDCs and CD8\(^+\) T cells. However, whether the effects of type I IFNs on these cell types are direct or indirect is unclear, and we are interested in exploring these mechanisms in the future.
Extracellular Adenosine as a Master Regulator of Immune Cell Migration into the Central Nervous System via Induction of CX3CL1

Jeffrey H. Mills¹, Leah Alabanza¹, Deeqa Mahamed¹, and Margaret S. Bynoe¹
¹Department of Microbiology and Immunology, Cornell University College of Veterinary Medicine, Ithaca, NY 14853

Having the ability to modulate lymphocyte entry into the central nervous system (CNS) would benefit patients with neuroinflammatory diseases. We have previously shown that extracellular adenosine regulates CNS entry of lymphocytes during experimental autoimmune encephalomyelitis (EAE), the animal model for the CNS inflammatory disease multiple sclerosis. For instance, while extracellular adenosine levels are vastly increased following inflammatory cellular damage (from the hydrolysis of released cytoplasmic ATP by CD39 and CD73), mice lacking CD73 or given adenosine receptor (AR) antagonists have significantly reduced CNS lymphocyte entry during EAE. We now show through detailed genetic studies that AR signaling regulates lymphocyte migration into the CNS though induction of CX3CL1, a specialized chemokine that acts as both an adhesion molecule and chemoattractant for lymphocytes, monocytes, and NK cells. We show that AR signaling is necessary and sufficient to induce CNS expression of CX3CL1 (as compared to over 40 other chemokines and adhesion molecules). AR regulation of CX3CL1 is critical for EAE progression, as daily anti-CX3CL1 treatments prevent CNS lymphocyte infiltration during EAE. Importantly, AR signaling induces CX3CL1 expression on the choroid plexus, a known CNS entry point for lymphocytes. As AR signaling can promote lymphocyte transmigration across a choroid plexus transwell barrier, we conclude that adenosine is a master regulator of CNS lymphocyte entry.

F32 NS 066682 (J.H.M.) and R01 NS 063011 (M.S.B)
Matrix metalloproteinases (MMP) are involved in tissue remodeling under various physiological or pathological conditions. A Stromelysin family member MMP3 (or Str1) is significantly involved in several physiological processes mediating enzymatic modification on a broad range of substrates. In cancer, MMP3 has demonstrated a significant role in the initiation of tumor development. However, whether MMP3 plays important roles in the stages of tumor progression and metastasis, remain unexplored. Therefore our first aim is to test the hypothesis that MMP3 can be a determining factor in neoplastic progression, focusing on primary tumor growth and metastasis. We approached the problem using loss and gain of function methods. We used a sarcoma cell line (CMS4) that showed detectable levels of MMP3 expression at both mRNA and protein levels. Thus far, our results showed that: 1) silencing MMP3 with shRNA in the parental tumor population significantly reduced local tumor growth, and 2) over-expression of MMP3 significantly increased tumor growth, which suggests that MMP3 can affect the neoplastic process beyond initiation. Next we plan to extend our study to experimental or spontaneous metastasis models to determine if MMP3 is causally linked to metastatic events. Our next aim is to identify a novel mechanism of regulation of MMP3. Ongoing studies in our lab suggest that the transcription factor, Interferon regulatory factor-8 (IRF-8), serves as a negative regulator of MMP3. IRF8 previously has been shown to mediate antitumor responses in both hematopoietic and non-hematopoietic models. Disruption of IRF8 in CMS4 cells significantly augments its aggressiveness in vivo. A number of experimental data (differential microarray, RT-PCR, qPCR and Western blot analyses) revealed up-regulation of MMP3 as a result of alterations in IRF8 function in CMS4 cells. MMP3 reporter assays reveal that IRF8 is a negative regulation of MMP3 transcription; CMS4 cells experimentally lacking IRF8 were more efficient in driving MMP3 promoter activity than CMS4 expressing IRF8. Thus we hypothesize that IRF-8 behaves as a negative regulator of MMP3 by either directly binding to MMP3 promoter or indirectly through interactions with other IRF-8 binding partners. Ongoing studies are testing the hypothesis that altered MMP3 activity, at least in part, accounts for differences in tumor growth between IRF-8-deficient and IRF-8-expressing tumor cells. Understanding the importance of MMP3 and its regulation may have important implications for cancer therapy.
Session III

Regulation of Immune Responses
The Role of Autoimmunity in Maintaining CD8 Memory T-cell Responses to B16 Melanoma

Katelyn T. Byrne and Mary Jo Turk
Dartmouth Medical School and Norris Cotton Cancer Center, Lebanon NH

The autoimmune destruction of melanocytes (i.e. vitiligo) has long been correlated with improved survival in metastatic melanoma patients. In spite of this, vitiligo is often disregarded as a side effect of anti-tumor immunity. We recently demonstrated that melanocyte antigen liberated during vitiligo is required to maintain melanoma/melanocyte specific memory CD8 T cell responses that protect against melanoma re-challenge. However, the possibility that melanocyte killing itself is immunogenic enough to prime naïve CD8 T cell responses against melanoma/melanocyte antigens has not been investigated. We have previously reported that CD8 T cell mediated vitiligo develops in B16 tumor bearing mice that are treated by depletion of CD4 regulatory T cells followed by surgical excision of tumor. To directly probe the immunogenicity of vitiligo, we adoptively transferred naïve melanoma/melanocyte antigen (gp100) specific pmel CD8 T cells in to actively depigmenting hosts following tumor removal. We found that naïve pmel CD8 T cells upregulated CD44 and underwent several rounds of division upon exposure to vitiligo. These vitiligo-primed CD8 T cells also began to express the skin-homing marker, P-selectin ligand. Pmel CD8 T cells primed by vitiligo produced Granzyme B and expressed LAMP-1, but did not produce IFN-γ upon peptide restimulation. Furthermore, vitiligo-primed CD8 T cells were unable to form long-lived memory populations. These studies demonstrate that vitiligo is immunogenic enough to prime melanocyte-antigen specific CD8 T cells that acquire certain functional capabilities, and therefore may play a role in immune responses to melanoma.
Memory T_{reg} cells Regulate the Immune Response to Influenza Virus Infection

Erik L. Brincks, Alan D. Roberts, Tres Cookenham, Jacob E. Kohlmeier, and David L. Woodland
Trudeau Institute, Saranac Lake, NY 12983

Regulatory CD4^{+}FoxP3^{+} T cells (T_{reg}) are key regulators of inflammatory responses and control the magnitude of cellular immune responses to viral infections. However, little is known about how T_{reg} contribute to immune regulation during memory responses to previously-encountered pathogens. Here we investigated the extent to which T_{reg} influence memory responses to influenza virus infection. Utilizing influenza NP_{311}-Class II tetramers, we tracked the antigen-specific T_{reg} response to primary and secondary influenza virus infections. Antigen-specific memory T_{reg} showed accelerated accumulation in the lung-draining lymph node and lung parenchyma relative to a primary influenza virus infection. These antigen-specific memory T_{reg} arose from the thymically-derived pool of CD4^{+} T cells, and they did not arise from the conversion of FoxP3^{neg}CD4^{+} T cells to a FoxP3^{+} phenotype. In vitro studies showed that memory T_{reg} cells effectively controlled the proliferation of memory CD8^{+} cells in an antigen-specific fashion, and that this regulation was MHC class II dependent. Additionally, memory T_{reg} controlled in vivo responses to secondary influenza infection by limiting the magnitude of recall responses by antigen-specific memory CD8^{+} T cells as well as by regulating airway chemokine and cytokine expression. Together, these data demonstrate a previously unappreciated role for T_{reg} in shaping the cellular immune response to secondary influenza virus challenges and offer an additional parameter to consider when determining the efficacy of vaccinations.
Epicutaneous (Skin) Immunization with Auto-antigen Induces Neuropilin-1-mediated Suppressor CD4 T-cells

Margaret S. Bynoe, Benjamin D. Solomon, Cynthia Mueller, Wook-Jin Chae, Leah Alabanza

1Department of Microbiology and Immunology, College of Veterinary Medicine
Cornell University, Ithaca, NY
2Department of Immunobiology, Yale University School of Medicine, New Haven, CT

Neuropilin-1 (Nrp1) is a type-1 transmembrane cell surface molecule originally identified for its role in neuronal development. We have found that Nrp1 is highly expressed on CD4 T cells from mice that had been epicutaneously immunized with a myelin antigen, myelinoligodendrocyte glycoprotein (MOG), and confer dominant tolerance against experimental autoimmune encephalitis (EAE, the animal model for multiple sclerosis). Recently, Nrp1 has been implicated in several aspects of immune function including maintenance of the immune synapse and development of regulatory T cells (Treg). Here, we identify Nrp1 as a critical negative regulator of CD4 T cell effector function. We demonstrate that CD4 T cells lacking Nrp1 (via conditional deletion) exhibit preferential Th17 lineage commitment, are hyperproliferative and produce high levels of proinflammatory cytokines. Mice with conditional deletion of Nrp1 in CD4 T cells develop fulminant EAE that can be inhibited by blocking Th17 cell development. Conversely, mice receiving CD4 T cells that are retrovirally transduced with Nrp1 do not develop EAE. Further, Nrp1-expressing CD4 T cells are potent suppressors of CD4 T cell effector function. This suppression is independent of FoxP3 and can be inhibited by blockade of TGF-beta but not by IL-10 blockade. These findings are the first to demonstrate a specific regulatory role for Nrp1 in immune regulation and provide the impetus for the use of Nrp1 as a therapeutic immune modulator.

Funding: NIH: AI 57854 and A1 072434 (to M.S.B.).
Generation of long-lived protective memory CD8 T cells is an important goal of all vaccines against intracellular pathogens. Thus, an understanding of the mechanisms regulating development of memory CD8 T cells with robust recall proliferation upon secondary challenge, and antigen-independent longevity is crucial for rational vaccine design. Previous studies from our lab and others’ have delineated cell surface markers capable of distinguishing memory-fated effector cells (memory precursors, MPs) from effector cells that largely die after antigen clearance (terminal effectors, TEs). These include Killer cell lectin-like receptor subfamily G member 1 (KLRG-1) and IL-2Ra (CD25). Identification of MPs within the effector cell pool during priming and CD8 T cell expansion supports the notion that commitment to the memory lineage occurs early during primary CD8 T cell responses. Moreover, it is proposed that hallmark memory properties of self-renewal via homeostatic proliferation and recall proliferative potential are “programmed” in the MPs during priming. However, the factors that program optimal memory function and longevity are largely undefined. Recent data delineating key cell-intrinsic factors governing memory cell longevity and secondary expansion will be discussed.
Induction and Activation of the Aryl-Hydrocarbon-Receptor Maintains an 
IDO-mediated Tolerogenic Tumor Microenvironment

Jamie L. Harden and Nejat K. Egilmez
SUNY Buffalo, Department of Microbiology and Immunology

A single intra-tumoral injection of IL-12 and GM-CSF microspheres results in tumor regression and initiation of an anti-tumor immune response. Previous studies in our lab have shown that treatment results in immunogenic DC 2 days post-treatment, followed by induction of the tolerogenic enzyme, indoleamine 2, 3-dioxygenase (IDO). Studies in IFN-gamma-/- mice demonstrated that this cytokine was responsible not only for the immunogenic DC phenotype, but also for the subsequent tolerogenic, IDO-positive DC. How IFN-gamma induced only transient DC activation, but prolonged IDO-mediated tolerogenicity however, was not understood. Recently, kynurenine (the IDO produced degradation product from tryptophan) was determined to be a natural ligand for the aryl-hydrocarbon receptor (AhR). Kynurenine activated AhR was found to polarize CD4+ T-cells to a T-regulatory cell phenotype, and induce IDO expression in bone marrow derived DC. Therefore, we hypothesized that if AhR expression was induced by IFN-gamma after GM-CSF and IL-12 treatment, this transcription factor may serve as the conduit to maintain a tolerogenic environment. We found that AhR expression increased in the tumor after GM-CSF and IL-12 microsphere treatment. Additionally, AhR was found to be bound by ligand and active in gene transcription one week after GM-CSF and IL-12 microsphere treatment. No induction of IDO or AhR was found in IFN-gamma-/- mice after GM-CSF and IL-12 microsphere treatment. Additionally, AhR transcriptional activity was not observed one week after treatment in IFN-gamma-/- mice, presumably due to loss of induction of IDO, and therefore lack of the production of the AhR ligand, kynurenine. These data suggest that IDO expression is maintained post-immunotherapy via binding of kynurenine to the AhR.
Session IV

Featured Speaker and Selected Oral Poster Presentations
IL-6 Trans-signaling Licenses Tumor Microvascular Gateways for Trafficking of Cytotoxic T-cells

Daniel T. Fisher, Qing Chen, Joseph J. Skitzki, Jason B. Muhitch, Lei Zhou, Michelle M. Appenheimer, Trupti D. Vardam, Stefan Rose-John, Elizabeth A. Repasky, Heinz Baumann, and Sharon S. Evans

Department of Immunology and Department of Molecular and Cellular Biology, Roswell Park Cancer Institute, Buffalo, NY, USA. Cancer Biology and Genetics Program, Memorial Sloan-Kettering Cancer Center, New York, NY, USA Department of Biochemistry, Christian-Albrechts-University, Kiel, Germany.

Inflammatory cytokines exert tumor-promoting activities by driving growth and survival of neoplastic cells. However, the opposing role of cytokines in recruiting mediators of adaptive anticancer immunity has not been investigated. Here we report that homeostatic trafficking of tumor-reactive CD8 T lymphocytes across microvascular checkpoints is limited in tumors despite the presence of inflammatory cytokines. Intravital imaging revealed a switch to an adhesive vasculature supporting trafficking of CD8 effector/memory T cells (T_{EM}) that was triggered in murine tumor vessels by an IL-6 trans-signaling program during systemic thermal therapy. Conversely, LPS-induced inflammation was ineffective in improving intratumoral T_{EM} homing. A concomitant decrease in infiltration of T_{Reg} during thermal therapy resulted in substantial enhancement of T_{EM}:T_{Reg} ratios. Mechanistically, IL-6 produced by non-hematopoietic stromal cells acted cooperatively with soluble IL-6 receptor and thermally-inducible gp130 to promote E/P-selectin and ICAM-1–dependent extravasation of cytotoxic T cells in tumors. Parallel increases in vascular adhesion were induced by IL-6/soluble IL-6 receptor fusion protein in murine tumors and patient tumor explants. Finally, a causal link was established between IL-6–dependent licensing of tumor vessels for T_{EM} trafficking and apoptosis of tumor targets. These findings suggest that the unique IL-6–rich tumor microenvironment can be exploited to create a therapeutic window to boost T cell-mediated antitumor immunity and immunotherapy.

Supported by the NIH (CA79765, CA094045, and AI082039)
Impairment of CD4+ T-cell Cognate Helper Functions in Response to Influenza Immunization with Aging

Julie S. Lefebvre, Ashlee H. Petell, Paula A. Lanthier, Sheri M. Eaton, and Laura Haynes
Trudeau Institute, Saranac Lake NY

Influenza infection is a major cause of hospitalization and mortality in the elderly population. This population is therefore targeted for yearly vaccination against influenza, which is the principal method of disease prevention. The influenza vaccine, however, only provides 40-60% efficacy in people over 65 years old. This low efficacy results from low antibody titers with poor neutralizing activity. CD4+ T cells have been shown to be critical for germinal center formation, which is required for the generation of high affinity antibodies by B cells. Using model antigens such as ovalbumin and pigeon cytochrome c, our previous work showed that CD4+ T cells from old mice (>20 months) acquire age-associated defects that impair their cognate helper functions.

In the present studies, we aimed to determine whether defects in the CD4+ T cell response to influenza immunization in aged mice is responsible for the poor vaccine efficacy. To do so, we vaccinated young (2-3 months) and aged (>20 months) Balb/c mice i.m. with 10^8 EID50 heat-inactivated influenza A/PR/8/34 (H1N1), a mouse adapted strain. The CD4+ T cell response in the draining lymph nodes was then evaluated by flow cytometry, while the non-draining lymph nodes were used as negative controls (baseline).

Our data show that at least 2 times fewer CD4+ T cells express the T follicular helper (Tfh) markers CXCR5 and PD-1 in the draining lymph nodes of aged mice than in young mice at all time points assessed. The kinetic of accumulation of these cells was also impaired in the aged mice. While significant numbers of CXCR5+PD-1+CD4+ T cells can already be found by day 5 in young mice, we started to see an increase only at day 7 in the aged mice. Moreover, the number of CXCR5+PD-1+CD4+ remains higher than the baseline level (≥5 times) at least until day 15 post-immunization in young mice whereas it drops to baseline levels as early as day 11 post-immunization in aged mice. Similar results were obtained when evaluating the generation of germinal center B cells, suggesting that CD4+ T cells also had impaired cognate helper functions in the aged animals. This resulted in lower PR8-specific antibody and lower neutralizing antibody titers in the aged mice compared to the titers measured in young mice. Following the addition of an adjuvant, such as the toll-like receptor 3 agonist poly(I:C), the proportion of CD4+ T cells expressing CXCR5 and PD-1, as well as the proportion of germinal center B cells, increased significantly in both young and aged mice draining lymph nodes. This leads to higher PR8-specific antibody titers in both young and aged mice.

In summary, our data strongly support our hypothesis that the impairment of CD4+ T cells functions with aging is a leading cause of the defective humoral response to influenza immunization in the elderly. The addition of adjuvants improves vaccine efficacy partly by improving CD4+ T cell functions.

Poster No. 2
Mechanism of Neutralizing Antibodies Against the Binding Subunit (RTB) of Ricin

Anastasiya Yermakova\textsuperscript{1} and Nicholas J. Mantis \textsuperscript{1,2}

\textsuperscript{1}Department of Biomedical Sciences, University at Albany School of Public Health, and 
\textsuperscript{2}Division of Infectious Diseases, Wadsworth Center, New York State Department of Health, 
Albany, NY

Currently, there are no approved vaccines or therapeutics against the Category B select agent ricin. Ricin belongs to a family of A-B toxins, its B subunit (RTB) functions as a galactose-specific lectin to promote attachment and entry of the ricin enzymatic A subunit (RTA) into host cells. In efforts to design a therapeutic, we propose that the B subunit is an appealing target since antibodies directed against RTB will prevent the toxin’s entry into cells before intoxication can take place. In a screen of approximately 2,000 RTB-specific B cell hybridomas, a handful of neutralizing monoclonal antibodies (mAbs) were identified. A panel of four neutralizing mAbs SylH3, 24B11, JB4, and B/J F9 blocked ricin attachment to terminal galactose residues. These antibodies also prevented toxin binding to primary macrophages which suggested that they function by steric hindrance recognizing epitopes located adjacent to RTB’s galactose binding pockets located within two of RTB’s six sub-domains, \(1\alpha\) and \(2\gamma\). In contrast, analysis of four non-neutralizing mAbs revealed recognition of epitopes situated within sub-domains of RTB which are not involved in galactose recognition (i.e., \(1\beta\), \(2\alpha\)). Most importantly, two of the neutralizing mAbs, SylH3 and 24B11, were shown by passive immunization to protect mice against a 5xLD\textsubscript{50} dose of ricin. Although all four neutralizing mAbs were shown to inhibit ricin attachment to terminal galactose residues, they differed in the degree of inhibition. The SylH3 and JB4 antibodies inhibited attachment by >80\%, and the 24B11 and B/J F9 antibodies inhibited attachment by <40\%. These results suggest that anti-RTB mAbs may function to neutralize ricin toxin intracellularly as well as through steric hindrance. Ongoing studies include passive administration of JB4 and B/J F9 to mice to determine protection against ricin. Additionally, we are also tracking intracellular toxin-Ab interactions via fluorescence microscopy.

Poster No. 9
Blk-Haploinsufficiency Impairs the Development, but Enhances the Functional Responses, of MZ B-cells

Elizabeth M. Samuelson, Renee M. Laird, Alexander C. Maue¹, Rosemary Rochford, and Sandra M. Hayes
Department of Microbiology and Immunology, State University of New York Upstate Medical University, Syracuse, NY 13210
¹Current address: Enteric Diseases Department, Naval Medical Research Center, Silver Spring, MD 20910

Blk was identified two decades ago as a B cell-specific member of the Src family of tyrosine kinases. Recent studies, however, have discovered that Blk is expressed in many cell types outside of the B lineage, including early thymic precursors, IL-17-producing γδ T cells and pancreatic β-cells. In light of these recent discoveries, we performed a more comprehensive analysis of Blk expression patterns in hematopoietic cells and found that Blk is differentially expressed in mature B cell subsets, with marginal zone (MZ) B cells expressing high levels, B1 B cells expressing intermediate to high levels, and follicular (FO) B cells expressing low levels of Blk. To determine whether these differences in Blk expression levels reflected differential requirements for Blk in MZ, B1 and FO B cell development, we analyzed the effects of reducing and eliminating Blk expression on B cell development. We report that both Blkhaploinsufficiency and Blk-deficiency impaired the generation of MZ B cells. Moreover, although there were fewer MZ B cells in Blk+/- and Blk-/- mice compared to Blk+/+ mice, Blk mutant MZ B cells were hyper-responsive to B cell receptor stimulation, both in vitro and in vivo. Thus, this study has revealed a previously unappreciated role for Blk in the development and activation of MZ B cells.

Poster No. 4

47.
Identification of a Putative IgM Memory B-cell Population During Bacterial Infection

Jennifer Yates, Rachael Racine, Maura Jones, and Gary Winslow
Wadsworth Center, Albany, NY

Humoral immunity and B cell memory are essential components of the adaptive immune response. These elements of immunity have remained largely unexplored in intracellular bacterial infections. *Ehrlichia muris* is an obligate intracellular bacterium that generates a chronic infection in immunocompetent mice. Chronic infection with *E. muris* is characterized by long-term IgM production that confers antibody-mediated protection against fatal ehrlichial challenge. We have identified a novel CD19+ B cell population in the spleens of chronically infected mice based on the expression of the cell surface markers CD11c, CD73, and PD-L2. Upon stimulation with LPS *in vitro*, the CD11c+/CD73+/PD-L2+ B cells proliferated and produced antigen specific IgM. BrdU incorporation studies revealed that the population is largely quiescent. To ascertain the role of this population during chronic ehrlichial infection, we generated mice chimeric for B-cell deficient, and CD11c-diphtheria toxin receptor (CD11c-DTR) transgenic bone marrow. The CD11c-DTR transgene allows the primate diphtheria toxin receptor to be expressed in susceptible murine cells under control of the CD11c promoter. Thus, murine cells expressing the CD11c molecule are susceptible to toxin administration *in vivo*. Deletion of the CD11c+/CD73+/PD-L2+ B cells during chronic infection resulted in significantly reduced levels of protective antigen-specific IgM 7 days post toxin administration. Based on these data, we propose that the CD11c+/CD73+/PD-L2+ B cells we have identified during chronic infection are long-lived effector/memory cells responsible for the maintenance of long-term immunity during chronic ehrlichial infection.
Session V

Virus-Host Interactions
Human Cytomegalovirus Reprograms Monocytes to Function as Vehicles of Viral Dissemination

Gary C. Chan
SUNY Upstate Medical University, Syracuse, NY

Human cytomegalovirus (HCMV), a betaherpesvirus, is endemic throughout the world with seropositivity reaching 50 to 80% among urban populations in the United States. HCMV infection is generally asymptomatically in immunocompetent individuals, although HCMV is now believed to be a primary viral candidate in the etiology of several diseases including atherosclerosis, inflammatory bowel disease and glioblastoma multiforme. In immunocompromised individuals such as neonates, AID patients and transplant recipients, HCMV infection can lead to multiorgan failure resulting in significant morbidity and mortality. The myriad of organ diseases associated with HCMV infection is a direct pathological consequence of the systemic viral spread to and infection of multiple organ sites that occurs during either asymptomatic or symptomatic infections, which is necessary for the establishment of viral persistence within the infected host.

HCMV infection is characterized by a monocyte-associated viremia prior to the onset of viral pathogenesis, suggesting that HCMV may utilize these blood sentinels as vehicles to mediate hematogenous dissemination of the virus to several organ sites. In support, monocytes are the primary cell type infected in the blood during acute HCMV infection and are the predominant infiltrating cell type found in infected organs. However, despite monocytes being “at the right place, at the right time”, these cells have a short life span of approximately 2 days and are not permissive for viral replication. To overcome this biological quandary, we have recently shown HCMV infection to stimulate differentiation of short-lived, viral replication non-permissive monocytes into long-lived, viral replication permissive macrophages, which, to our knowledge, is the only identified viral pathogen that can directly induce monocyte-to-macrophage differentiation. The process of monocyte-to-macrophage differentiation involves a delicate balance between apoptosis and differentiation, where the levels of caspase 3 activity plays a central regulatory role. We now have data showing that HCMV temporally regulates the activation of caspase 3 differently from other differentiation factors, in order to concurrently induce monocyte survival and maturation following infection. Elucidating the precise mechanism of how HCMV uniquely modulates the monocyte survival/differentiation machinery, in order to promote viral dissemination and persistence, may provide insight into novel therapeutic cellular targets against HCMV-infected monocytes, where replication does not occur until weeks post infection.
Kaposi Sarcoma Associated Herpesvirus Infection of Primary Human Endothelial Cells Activates the Proto-oncogene STAT3

Christine A King¹ and Craig McCormick²

¹Department of Microbiology and Immunology, SUNY Upstate Medical University, Syracuse, NY, USA, 13210
²Department of Microbiology and Immunology, Dalhousie University, Halifax, NS, Canada, B3H 1X5

Kaposi’s sarcoma associated herpesvirus (KSHV) is the etiological agent for 3 AIDS-related cancers: Kaposi’s sarcoma (KS), primary effusion lymphoma, and multicentric Castleman’s disease. The molecular mechanisms used by KSHV to induce cancer are incompletely understood. KS lesions harbor proliferating latently-infected endothelial cells (ECs), large numbers of inflammatory cells, and marked neoangiogenesis. Considered the major driving force in the development of KS, these KSHV-infected ECs elaborate a variety of pro-inflammatory and angiogenic factors that contribute to tumourigenesis. Considerable evidence has accumulated suggesting a critical role for activated signal transducer and activator of transcription-3 (STAT3) in malignant transformation. STAT3 is a latent transcription factor that upon activation, drives the expression of a number of genes involved in cell proliferation, survival, and immune responses. Canonical STAT3 activation occurs via phosphorylation of Y705, dimerization, and nuclear translocation, followed by phosphorylation of S727 for maximal transcriptional activity. Activated STAT3 has been observed in a variety of malignancies and has been shown to induce fibroblast transformation in vitro suggesting that STAT3 is a proto-oncogene. Interestingly, evidence has accumulated suggesting a role for S727 mono-phosphorylated STAT3. Here we show that latent KSHV infection of primary human endothelial cells (ECs) in vitro activates STAT3, and identify a key latency protein, kaposin B, that contributes to this activation. Kaposin B expression in ECs causes STAT3 phosphorylation at S727, in the absence of significant Y705 phosphorylation, and enhanced expression of a subset of STAT3 target genes including CCL5. Recent work shows that the tripartite motif-containing protein 28 (TRIM28, a.k.a. TIF-1β, KAP-1) negatively regulates STAT3 by recruiting transcriptional silencing complexes. The repressive activity of TRIM28 is mediated by post-translational modifications and a key site in the regulation of repressor activity maps to S473. Phosphorylation of this residue disrupts the recruitment of transcriptional silencing complexes effectively deactivating the co-repressive function of TRIM28. Confocal microscopy and western blot analysis demonstrate phosphorylation of TRIM28 at S473 in KSHV latently infected and kaposin B expressing ECs. Taken together, our studies suggest kaposin B may contribute to tumourigenesis via constitutive activation of STAT3.
Multicolor Flow Cytometry: Optimizing Instrument Settings

Darci Gorgone, M.A.
Technical Applications Specialist
BD Biosciences

Multicolor flow cytometry rapidly reveals a large amount of biological information from a single sample. It often is the only means to adequately identify or functionally characterize complex populations of interest within the immune system. Over the past few years, the number of parameters (and consequently colors) simultaneously analyzed in typical flow cytometry experiments has increased. This is enabled by the availability of high performance instrumentation with additional laser and detector options and data computational power, along with advances in biochemistry that have led to more fluorochrome options. Not only has this increased the usefulness of flow cytometry, but it has increased the importance of proper experiment setup to ensure accurate and meaningful results. In this talk, we will provide some guidelines for consistent and reliable instrument set up.
Session VI

Inflammation
Susceptibility of Asthmatic Mice to Influenza A Virus Challenge

*Yoichi Furuya*¹, Sean Roberts¹, Alan Sanfilippo¹, Andrea Marias¹ and Dennis Metzger¹

¹Center for Immunology & Microbial Disease, Albany Medical College, Albany, NY 12208

Viral infections are responsible for approximately 85% of asthmatic exacerbations, and currently, there are no effective prophylactic therapies. Despite this, not much is known about how the asthmatic phenotype could influence immune responses against respiratory viral pathogens. Using a mouse model of asthma, we investigated the susceptibility of asthmatics to influenza A virus challenge. Unexpectedly, asthmatic mice were highly resistant to infection with the H1N1 2009 pandemic strain. However, although viral replications were initially delayed in asthmatic mice, viral lung titers ultimately reached comparable levels relative to non-asthmatic mice. Asthmatic mice also had increased numbers of immune cells in their lungs before and after influenza virus challenge. However, the lungs of asthmatic mice were found to have decreased levels of various cytokines including IFN-gamma, TNF, IL-12, and IL-5. Thus, increased survival rate did not correlate with viral titer; instead, increased resistance was associated with reduced pro-inflammatory lung cytokine levels and increased numbers of immune cells at the site of infection. Currently, we are elucidating the mechanisms responsible for enhanced resistance of asthmatic mice to influenza virus infection.
Insights into Inflammatory Bowel Disease Using *Toxoplasma gondii* as an Infectious Trigger

*Eric Y. Denkers*¹, *Kirk J. Maurer*²,³, *Sara B. Cohen*¹, *Matthias Mack*⁴, *Kenneth W. Simpson*⁵ and *Charlotte E. Egan*¹

Departments of *¹Microbiology and Immunology, and ²Biomedical Sciences, College of Veterinary Medicine, Cornell University, Ithaca, New York, USA; ³Center for Animal Resources and Education, College of Veterinary Medicine, Cornell University, Ithaca, New York, USA; ⁴Department of Internal Medicine II, University of Regensburg, Regensburg, Germany; ⁵Department of Clinical Sciences, College of Veterinary Medicine, Cornell University, Ithaca, New York, USA

Oral infection of C57BL/6 mice with *Toxoplasma gondii* triggers severe necrosis in the ileum within 7-10 days of infection. Lesion development is mediated by Th-1 cytokines, CD4⁺ T cells, and sub-epithelial bacterial translocation. As such, these features share similarity to Crohn’s disease. Recently, we uncovered a role for intraepithelial αβ and not γδ T cell IELs mediate intestinal damage. By adoptive transfer of mucosal T cells into naïve *Rag1⁻/⁻* mice, we demonstrate that IEL do not function alone to cause inflammatory lesions, but act with CD4⁺ T lymphocytes from the lamina propria. Furthermore, recipient mice pretreated with broad-spectrum antibiotics to eliminate intestinal flora resisted intestinal disease after transfer of IEL and lamina propria lymphocytes. Our data provide valuable new insight into mechanisms of intestinal inflammation, findings that have important implications for understanding human inflammatory bowel disease.
Apoptotic Cell ‘Find-me’ Signals in Cell Clearance and Inflammation

Michael R. Elliott, Ph.D.
University of Rochester

Cells undergoing apoptosis must be rapidly and efficiently cleared in order to prevent aberrant inflammation. Professional phagocytes such as macrophages, monocytes and dendritic cells are responsible for much of this clearance owing to their efficiency in moving through tissues and their high phagocytic capacity, although the signaling events that control these outcomes are poorly understood. Recently we discovered that apoptotic cells specifically release nucleotides ATP and UTP via pannexin-1 channels on the plasma membrane, and that these nucleotides act as apoptotic cell “find-me” signals that stimulate migration via P2Y receptors on the responding phagocyte, leading to the prompt and efficient clearance of apoptotic T cells in vivo. Extracellular nucleotides and nucleosides (i.e. adenosine) exert a wide range of biological effects through receptor-mediated autocrine and paracrine mechanisms in a process collectively referred to as purinergic signaling. From an immunological perspective, extracellular nucleotides can promote either pro- or anti-inflammatory outcomes depending on the target cell and microenvironment. While purinergic signaling is clearly important for the “find-me” response, how these factors specifically alter phagocyte function or the role they play in the larger context of apoptotic cell clearance and the inflammatory immune response stands as a new and important areas for investigation.
Session VII

Immune Response to Pathogens
IL-10 Maintains a Pool of Hepatic B-cells that Prevents Development of Severe Cardiopulmonary Disease in Schistosomiasis

Keke C. Fairfax, Eyal Amiel, Irah L. King, Markus Mohrs, and Edward J. Pearce
Trudeau Institute, Saranac Lake, NY 12983

In schistosomiasis patients, parasite eggs trapped in hepatic sinusoids become foci for Th2 cell-orchestrated granulomatous cellular infiltrates. Since the immune response is unable to clear the infection, the liver is subjected to ongoing cycles of focal inflammation and healing that lead to vascular obstruction and tissue fibrosis. This is mitigated by regulatory mechanisms that develop over time and which minimize the inflammatory response to newly deposited eggs. Exploring changes in the hepatic inflammatory infiltrate over time we found a marked accumulation of schistosome egg antigen-specific IgG1-secreting plasma B cells. This population of cells was significantly altered by blockade of the receptor for IL-10, a cytokine implicated in plasma cell development. Strikingly, IL-10R blockade also precipitated the development of embolic pulmonary hypertension due to the portal-systemic shunting of parasite eggs. This did not reflect more aggressive Th2 cell responsiveness, as we were unable to detect any changes in Th2 cell proliferation or IL-4 production as a result of treatment with anti-IL-10R antibody. Rather, a role for B cells in the prevention cardiopulmonary disease is reinforced by our finding that disease outcome in infected B cell-deficient mice phenocopies that which develops in infected wild type mice when IL-10R is blocked. Our data describe a role for IL-10-dependent B cell responses in the regulation of tissue damage during a chronic helminth infection.
cAMP-dependent IL-10 Production Determines the Severity of Lyme Disease

Bikash Sahay, Rebeca Patsey, Kathleen Bashant, Timothy Sellati
Center for Immunology and Microbial Disease, Albany Medical College, Albany, NY 12208

*Borrelia burgdorferi* is the causative agent of Lyme disease in humans and susceptibility varies widely in the population, which can be efficiently mimicked in a murine infection model. While C57Bl/6 (B6) mice are resistant to disease, C3H/HeN (C3H) mice are highly susceptible to Lyme Borreliosis, with features reminiscent of severe Lyme morbidity in humans. Disease resistance has been associated with the ability of macrophages (MΦ) from a B6 mouse to produce the anti-inflammatory cytokine IL-10, which is not adequately produced by C3H MΦ; rather, C3H MΦ produce interferon response factors such as *irf1*. Expression of *irfs* in C3H MΦ and *il10* in B6 MΦ is controlled by p38-dependent activation of STAT1 (Sahay 2009; Miller 2007) and STAT3 (Sahay 2011), respectively. Interestingly, both genotypes are equally efficient in activating *B. burgdorferi*-induced STAT1 and STAT3 but exhibit markedly different outcomes. Cyclic AMP (cAMP) is a second messenger that has been shown to inhibit the function of STAT1 without affecting its activation status and simultaneously activates cAMP response element binding protein (CREB) for IL-10 production. We evaluated cAMP levels between these two genotypes and found that B6 MΦ contain twice as much cAMP as compared to C3H MΦ. Upon exogenous addition of cAMP, C3H MΦ produced IL-10 in response to *B. burgdorferi* with a reduction in *irf1* transcription. Interestingly, inhibition of adenylate cyclase following recognition of *B. burgdorferi* did not reduce IL-10 release, suggesting that the initial accumulation, but not the generation, of cAMP is the primary difference between B6 and C3H MΦs. These findings suggest a role for cAMP in determining disease severity within the human population and represent a potential therapeutic target during Lyme disease.
Induction of a Polyreactive IgM Response During Bacterial Infection

Derek D. Jones, Greg Delulio, and Gary Winslow
Wadsworth Center, Albany, NY

Infection of mice with the intracellular bacterium *Ehrlichia muris* induces a protective T cell-independent IgM response. A unique property of the IgM elicited during acute infection is that it is polyreactive; that is, it binds to several foreign and self antigens, including influenza virus, *Borrelia burgdorferi*, lipopolysaccharide, single- and double-stranded DNA, insulin, and thyroglobulin. We hypothesized that serum polyreactivity was either due to cross-reactivity of antigen-specific IgM, or that the non-pathogen-specific antibodies were generated by polyclonal B cell activation. To address these hypotheses, we generated B cell hybridomas from spleen cells from infected mice, and determined whether the IgM produced from the hybridomas was ehrlichia-specific. The majority of clones (80%) produced pathogen-specific IgM; of these, 30% bound an immunodominant outer membrane protein, OMP-19. The IgM that bound OMP-19 was highly polyreactive relative to the IgM that bound other ehrlichial antigens, suggesting that the physical properties of the antigen influence the generation of polyreactive IgM. Infection-induced polyreactive IgM bound HEp-2 cells, a human cell line used clinically to identify anti-nuclear antibodies. Polyreactive autoantibodies were also detected in sera from human patients. Because anti-nuclear antibodies are characteristic of systemic lupus erythematosus, we plan to address whether infection-induced polyreactive IgM can modify disease in lupus-prone (NZB x NZW) F1 mice. Our data indicate that polyreactivity, often considered to be a property of natural IgM, is also characteristic of IgM elicited by pathogen-specific T cell-independent responses, and likely plays a role in host defense and inflammation.
Autophagy-mediated Defense Against *Francisella tularensis*

1,2Girish S. Kirimanjeswara, D.V.M., Ph.D. and 1Dennis W. Metzger, Ph.D.
1Center for Immunology and Microbial Disease, Albany Medical College, Albany, NY, 12208
2Current Address: Dept. of Veterinary and Biomedical Sciences, Pennsylvania State University, University Park, PA, 16802

Sterilizing immunity is critical for the control of pathogens that have the potential to become persistent within a host. Sterilizing immunity also contributes to a strong herd immunity and thus critical for the control of pathogens in a population. It is therefore important to understand the mechanism of intracellular killing of microbes in order to develop better prophylactics and therapeutics. Using a model intracellular bacterium *Francisella tularensis*, we have demonstrated that a synergistic activation of Fc and Interferon gamma receptors is essential for the rapid clearance of bacteria. Further elucidation of the intracellular killing of bacteria revealed that priming and triggering mechanism of macrophage activation leads to induction of microbiocidal nitric oxide and differential trafficking of phagocytosed bacterium. Specifically, autophagy was induced in primed macrophages and the bacteria phagocytosed by Fc receptors were targeted to autophagosomal compartment. The bacteria within the autophagosomes were eventually trafficked to lysosomal compartment, where they were killed. These results revealed that additional complex mechanisms, not just priming and triggering, are involved in killing pathogens that exploit the phagocyte intracellular environment for their survival.
Interactions Between *Salmonella enterica* serovar Typhimurium and Secretory IgA at Epithelial Surfaces

Jayaleka J. Amarasinghe and Nicholas J. Mantis  
Wadsworth Center, New York State Department of Health, Albany, NY 12208

Sal4 is a monoclonal, polymeric IgA antibody directed against the O-antigen (O-Ag) of *Salmonella enterica* serovar Typhimurium that has been shown to be sufficient in protecting mice against enteric infection. We recently reported that the exposure of *S*. Typhimurium to Sal4 results in bacterial paralysis (loss of flagella-based motility) and inhibition of type 3 secretion (T3S) system-mediated entry into epithelial cells. We now demonstrate that Sal4 has additional immediate and long-term (>1 hr) physiologic effects on *S*. Typhimurium, including the production of a putative cellulose-containing capsular exopolysaccharide (EPS) leading to enhanced biofilm formation. In an effort to understand the signal transduction pathway involved in sensing the Sal4, we screened a library of chromosomal deletion mutants for those that retained motility in the presence of Sal4. This screen led to the identification of a *yeaJ*, a gene encoding a putative inner membrane-localized diguanylate cyclase (DGC) that contains an extra-cytoplasmic sensory (Cache) domain and a GGDEF output domain. In preliminary studies, the *yeaJ* mutant (Δ*yeaJ*) strain demonstrates a severe defect in biofilm formation and production of cellulose in response to Sal4, as compared to the wild-type strain. Overexpression of YeaJ in an otherwise wild-type strain of *S*. Typhimurium resulted in enhanced biofilm formation, cellulose production and rdar (red,dry and rough) colony formation. Based on these and other data, we propose that YeaJ mediates a signal transduction pathway in response to Sal4 associated with suppression of motility and enhanced secretion of an EPS that leads to biofilm formation. We propose that the interaction between Sal4 and *S*. Typhimurium reveals a mechanism by which enteric pathogens may sense and respond to host immunity at mucosal surfaces.
Poster Abstracts
**14th Annual Upstate New York Immunology Conference**  
**Poster Number Assignments**

<p>| | |</p>
<table>
<thead>
<tr>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>Henry Ogbomo</td>
</tr>
<tr>
<td>2.</td>
<td>Julie S. Lefebvre*</td>
</tr>
<tr>
<td>3.</td>
<td>Katrina Simmons*</td>
</tr>
<tr>
<td>4.</td>
<td>Elizabeth M. Samuelson*</td>
</tr>
<tr>
<td>5.</td>
<td>Abigail L. Sedlacek</td>
</tr>
<tr>
<td>6.</td>
<td>Joanne Lim*</td>
</tr>
<tr>
<td>7.</td>
<td>Jennifer Yates*</td>
</tr>
<tr>
<td>8.</td>
<td>Joanne M. O’Hara</td>
</tr>
<tr>
<td>9.</td>
<td>Anastasiya Yermakova*</td>
</tr>
<tr>
<td>10.</td>
<td>Hristina Nedelkovska</td>
</tr>
<tr>
<td>11.</td>
<td>Lisbeth A. Boule</td>
</tr>
<tr>
<td>12.</td>
<td>Bethany Winans</td>
</tr>
<tr>
<td>13.</td>
<td>Jeffrey H. Mills*</td>
</tr>
<tr>
<td>14.</td>
<td>Deeqa Mahamed</td>
</tr>
<tr>
<td>15.</td>
<td>Bikash Sahay</td>
</tr>
<tr>
<td>16.</td>
<td>Antje Krenz</td>
</tr>
<tr>
<td>17.</td>
<td>Leah M. Alabanza</td>
</tr>
<tr>
<td>18.</td>
<td>Eyal Amiel</td>
</tr>
<tr>
<td>19.</td>
<td>Debarati Banik*</td>
</tr>
<tr>
<td>20.</td>
<td>Alan M. Sanfilippo</td>
</tr>
<tr>
<td>21.</td>
<td>Ellen B. Duffy</td>
</tr>
</tbody>
</table>

* Selected to give an oral poster presentation
Innovative Biotherapy for Brain Tumors: Myxoma Virus Synergizes with Natural Killer Cells to Clear Malignant Gliomas

Henry Ogbomo¹, Xueqing Lun¹, Jiqing Zhang¹, Grant Mcfadden², Christopher Mody³, and Peter Forsyth⁴*

¹Departments of Oncology, Biochemistry and Molecular Biology, University of Calgary, Calgary AB
²Tom Baker Cancer Centre, Southern Alberta Cancer Research Institute, Clark H. Smith Brain Tumor Centre, Calgary AB
³Department of Molecular Genetics and Microbiology, University of Florida College of Medicine, Gainesville FL
⁴Departments of Microbiology and Infectious Diseases, and Internal Medicine, University of Calgary and Calgary Health Region, Calgary AB
⁵Dept of Neurooncology, Moffitt Cancer Center and U. of Southern Florida, Tampa FL

Innovative biotherapeutic approaches for experimental malignant gliomas (MGs) and brain tumor stem cells (BTSCs) include using oncolytic viruses such as Myxoma Virus (Myxv) or immune cells such as Natural Killer (NK) cells. In this study, we combined the individual strengths of using NK cells and Myxv to treat MGs with a view of overcoming the potential limitations posed when each is used as monotherapy. All MGs (U87, U251, U118) and BTSCs (25EF and 48EF) investigated expressed varying levels of NK ligands including inhibitory MHC-I, activating ULBP1-3, MICA/B, and co-activating nectin-2, poliovirus receptor. Infection of MGs and BTSCs with Myxv resulted in the downregulation of MHC-I expression both on the cell surface and intracellularly. No effect was observed on the expression of NK activating ligands except for the downregulation of nectin-2. The downregulation of MHC-I and nectin-2 by Myxv was specifically mediated by M153 gene of Myxv as loss of M153 restored MHC-I and nectin-2 expression. Subsequently, NK cell killing of MGs was enhanced in Myxv-infected cells. Loss of M153 inhibited NK lysis of MGs. In vivo, using luciferase-expressing U87 cells in a mouse intracranial model, Myxv synergized with NK cells to accelerate tumor clearance. Combined intratumoral treatment with Myxv and NK of established U87 tumor accelerated tumor clearance when compared with single treatment with Myxv or NK. Tumor in 66% of mice (4 out of 6 mice) were cleared or significantly reduced within 14 days following combination treatment with Myxv and NK. No significant effect was observed in Myxv or NK singly-treated mice within the same time frame.
Impairment of CD4+ T-cell Cognate Helper Functions in Response to Influenza Immunization with Aging

Julie S. Lefebvre, Ashlee H. Petell, Paula A. Lanthier, Sheri M. Eaton, and Laura Haynes
Trudeau Institute, Saranac Lake NY

Influenza infection is a major cause of hospitalization and mortality in the elderly population. This population is therefore targeted for yearly vaccination against influenza, which is the principal method of disease prevention. The influenza vaccine, however, only provides 40-60% efficacy in people over 65 years old. This low efficacy results from low antibody titers with poor neutralizing activity. CD4+ T cells have been shown to be critical for germinal center formation, which is required for the generation of high affinity antibodies by B cells. Using model antigens such as ovalbumin and pigeon cytochrome c, our previous work showed that CD4+ T cells from old mice (>20 months) acquire age-associated defects that impair their cognate helper functions.

In the present studies, we aimed to determine whether defects in the CD4+ T cell response to influenza immunization in aged mice is responsible for the poor vaccine efficacy. To do so, we vaccinated young (2-3 months) and aged (>20 months) Balb/c mice i.m. with 10^8 EID50 heat-inactivated influenza A/PR/8/34 (H1N1), a mouse adapted strain. The CD4+ T cell response in the draining lymph nodes was then evaluated by flow cytometry, while the non-draining lymph nodes were used as negative controls (baseline).

Our data show that at least 2 times fewer CD4+ T cells express the T follicular helper (Tfh) markers CXCR5 and PD-1 in the draining lymph nodes of aged mice than in young mice at all time points assessed. The kinetic of accumulation of these cells was also impaired in the aged mice. While significant numbers of CXCR5+PD-1+CD4+ T cells can already be found by day 5 in young mice, we started to see an increase only at day 7 in the aged mice. Moreover, the number of CXCR5+PD-1+CD4+ remains higher than the baseline level (≥5 times) at least until day 15 post-immunization in young mice whereas it drops to baseline levels as early as day 11 post-immunization in aged mice. Similar results were obtained when evaluating the generation of germinal center B cells, suggesting that CD4+ T cells also had impaired cognate helper functions in the aged animals. This resulted in lower PR8-specific antibody and lower neutralizing antibody titers in the aged mice compared to the titers measured in young mice. Following the addition of an adjuvant, such as the toll-like receptor 3 agonist poly(I:C), the proportion of CD4+ T cells expressing CXCR5 and PD-1, as well as the proportion of germinal center B cells, increased significantly in both young and aged mice draining lymph nodes. This leads to higher PR8-specific antibody titers in both young and aged mice.

In summary, our data strongly support our hypothesis that the impairment of CD4+ T cells functions with aging is a leading cause of the defective humoral response to influenza immunization in the elderly. The addition of adjuvants improves vaccine efficacy partly by improving CD4+ T cell functions.

Poster No. 2
Effects of Vitamin D on Mammary Epithelial Cell Synthesis and Secretion of CD14

Katrina Simmons and JoEllen Welsh
Department of Biomedical Sciences, School of Public Health, University at Albany, Rensselaer, NY

Vitamin D has been shown to decrease the risk for infectious diseases by regulating the immune responses at the molecular level. The majority of the work in this area has focused on the direct effects of vitamin D on immune cells such as macrophages and monocytes. Very few studies have investigated how vitamin D alters the immune environment of epithelial cells in barrier tissues, such as the mammary gland, that are exposed to infectious agents. Through microarray screening of human mammary epithelial cells (hTERT-HME cell line, ATCC #CRL-4010) treated for 24 hours with 100nM 1,25D (the active form of vitamin D), we found an enrichment of genes involved in innate immunity, including CD14 which was elevated more than 60-fold. Based on these findings, we are examining the effects of vitamin D on the synthesis, secretion, and function of CD14 in hTERT-HME cells. CD14 is a pattern recognition receptor that binds lipopolysaccharide (LPS), a microbial cell wall component of gram-negative bacteria. Through quantitative PCR (qPCR), we confirmed that CD14 mRNA was significantly increased in hTERT-HME cells treated with either 1,25D or the circulating metabolite 25 hydroxyvitamin D (25D), which reflects overall vitamin D status. In association with up-regulation of CD14, we found down regulation of the pro-inflammatory cytokines IL-6, TNFα (alpha), and IL-1β (beta) in hTERT-HME cells treated with 25D. We next used a CD14 ELISA to examine whether vitamin D metabolites promoted the secretion of the soluble form of CD14 (sCD14) from mammary cells. Both 1,25D and 25D increased the accumulation of sCD14 in the conditioned media of hTERT HME cells in a dose and time dependent manner. Surprisingly, whereas vitamin D metabolites promoted the secretion of sCD14, purified LPS, a potent inducer of CD14 in the mammary gland and immune and non-immune cells, had no effect. This data suggests that the regulation of CD14 in mammary epithelial cells is distinct from that in other cell types. In conclusion, this data is the first to demonstrate effects of vitamin D on secretion of CD14 in mammary cells and support the central hypothesis that vitamin D promotes an anti-inflammatory environment via secretion of sCD14 from mammary epithelial cells.
Blk-Haploinsufficiency Impairs the Development, but Enhances the Functional Responses, of MZ B Cells

Elizabeth M. Samuelson, Renee M. Laird, Alexander C. Maue1, Rosemary Rochford, and Sandra M. Hayes
Department of Microbiology and Immunology, State University of New York Upstate Medical University, Syracuse, NY 13210
1Current address: Enteric Diseases Department, Naval Medical Research Center, Silver Spring, MD 20910

Blk was identified two decades ago as a B cell-specific member of the Src family of tyrosine kinases. Recent studies, however, have discovered that Blk is expressed in many cell types outside of the B lineage, including early thymic precursors, IL-17-producing γδ T cells and pancreatic β-cells. In light of these recent discoveries, we performed a more comprehensive analysis of Blk expression patterns in hematopoietic cells and found that Blk is differentially expressed in mature B cell subsets, with marginal zone (MZ) B cells expressing high levels, B1 B cells expressing intermediate to high levels, and follicular (FO) B cells expressing low levels of Blk. To determine whether these differences in Blk expression levels reflected differential requirements for Blk in MZ, B1 and FO B cell development, we analyzed the effects of reducing and eliminating Blk expression on B cell development. We report that both Blkhaploinsufficiency and Blk-deficiency impaired the generation of MZ B cells. Moreover, although there were fewer MZ B cells in Blk+/− and Blk−/− mice compared to Blk+/+ mice, Blk mutant MZ B cells were hyper-responsive to B cell receptor stimulation, both in vitro and in vivo. Thus, this study has revealed a previously unappreciated role for Blk in the development and activation of MZ B cells.
Tumor metastasis to the peritoneal cavity is frequently observed in patients with tumors of peritoneal origin such as ovarian, colon, or stomach cancer. Upon metastasis, the primary site of attachment is the omentum. The omentum is composed of immune aggregates embedded into adipose tissue and covered by a collagen matrix. It is to these immune aggregates specifically that tumor cells initially attach and divide. Given the proximity of tumor and immune cells, activation of these omental immune aggregates could potentially lead to an effective anti-tumor immune response.

Our work has shown that intraperitoneal immunization of C57BL/6 mice with syngeneic, lethally irradiated Colon38 cells prior to live tumor challenge prevents tumor growth on the omentum when mice are challenged with autologous Colon38 and interestingly non-autologous B16.F0. Whereas immunization results in an increase in the frequency and activation of CD8+ T cells on the omentum, only upon challenge with autologous Colon38 do we observe a specific CD8+ T cell response. This suggests that, following immunization, there are two mechanisms by which cells within the peritoneal cavity and on the omentum are capable of eliminating tumor: a tumor specific CD8+ T cell response and an alternate non-specific mechanism.
Exploring the Role of Type I Interferons in the Induction of Antitumor Response Following Radiation Treatment

Joanne Lim, Scott Gerber, and Edith Lord
University of Rochester

Although type I interferons (IFNs-I) play critical roles as factors that protect the host against viral infections, there has been increasing evidence that they are also essential for many immunological processes. Based on previous work by several investigators, there is clear indication that the responsiveness of hematopoietic cells to type I IFNs is important in generating antitumor immunity.

Our lab is interested in examining the importance of endogenous type I IFNs in the induction of antitumor immunity in response to radiation therapy. Using B16 melanoma mouse model, we have previously demonstrated that single high dose radiation therapy results in a dramatic delay in tumor progression, and this in turn is dependent on an increase in IFN-γ within the tumor microenvironment. We hypothesize that endogenous IFNs-I play a role in the induction of IFN-γ response following radiation therapy. We first compared the growth of B16 tumors between wild type (WT) mice and mice that lack functional IFN-α/β receptor (KO). Interestingly, tumor growth was faster in KO mice than in WT mice. When the tumors were treated with 15 Gy radiation, tumor progression in both KO and WT mice were slowed, but tumor burdens were still larger in the KO than WT mice. The induction of intratumoral IFN-γ mRNA levels by radiation therapy observed in WT mice, was drastically lower in KO mice. Importantly, the increase in CD8+ T cells as well as the increase in activation status of CD8+ T cells occurred only in mice that were able to signal through intact IFN-α/β receptors. Since dendritic cells (DCs) within tumor play a major role in the recruitment and activation of CD8+ T cells, we examined the activation status of these cells as well. MHC class I molecules, required for presentation of tumor antigen to CD8+ T cells, were upregulated on the surface of intratumoral DCs of WT mice in response to radiation treatment, but not in mice that lacked IFN-α/β receptors. Further, when intratumoral DCs were FACsorted and analyzed for chemokine mRNA levels, we observed that the relative levels of CXCL10 and CXCL11 were much higher in WT than KO cDCs, especially after radiation treatment. In conclusion, our data suggest that IFNs-I are required for complete activation of intratumoral cDCs and CD8+ T cells. However, whether the effects of type I IFNs on these cell types are direct or indirect is unclear, and we are interested in exploring these mechanisms in the future.
Identification of a Putative IgM Memory B-Cell Population During Bacterial Infection

Jennifer Yates, Rachael Racine, Maura Jones, and Gary Winslow
Wadsworth Center, Albany, NY

Humoral immunity and B cell memory are essential components of the adaptive immune response. These elements of immunity have remained largely unexplored in intracellular bacterial infections. *Ehrlichia muris* is an obligate intracellular bacterium that generates a chronic infection in immunocompetent mice. Chronic infection with *E. muris* is characterized by long-term IgM production that confers antibody-mediated protection against fatal ehrlichial challenge. We have identified a novel CD19+ B cell population in the spleens of chronically infected mice based on the expression of the cell surface markers CD11c, CD73, and PD-L2. Upon stimulation with LPS *in vitro*, the CD11c+/CD73+/PD-L2+ B cells proliferated and produced antigen specific IgM. BrdU incorporation studies revealed that the population is largely quiescent. To ascertain the role of this population during chronic ehrlichial infection, we generated mice chimeric for B-cell deficient, and CD11c-diphtheria toxin receptor (CD11c-DTR) transgenic bone marrow. The CD11c-DTR transgene allows the primate diphtheria toxin receptor to be expressed in susceptible murine cells under control of the CD11c promoter. Thus, murine cells expressing the CD11c molecule are susceptible to toxin administration *in vivo*. Deletion of the CD11c+/CD73+/PD-L2+ B cells during chronic infection resulted in significantly reduced levels of protective antigen-specific IgM 7 days post toxin administration. Based on these data, we propose that the CD11c+/CD73+/PD-L2+ B cells we have identified during chronic infection are long-lived effector/memory cells responsible for the maintenance of long-term immunity during chronic ehrlichial infection.
Specific Regions of the Ricin Toxin A Subunit are Involved in Eliciting Protective Antibodies: Implications for Vaccine Design

Joanne M. O’Hara¹, Robert N. Brey³, Nicholas J. Mantis¹,²
Department of Biomedical Science, School of Public Health, SUNY at Albany¹,
Wadsworth Center, New York State Department of Health, Albany, NY²
Soligenix Inc, Princeton, NJ³

Efforts to develop an effective vaccine against ricin are focused on engineering attenuated and stable recombinant forms of the toxin’s enzymatic A subunit (RTA). While several candidate antigens are in development, vaccine design and efficacy studies are being undertaken in the absence of a fundamental understanding of the regions of RTA that are critical in eliciting protective immunity. To address this issue, we recently identified six distinct immunodominant linear regions on RTA. Using monoclonal antibodies (MAbs) specific to each region, we determined that protective antibodies were directed against α-helices located in RTA folding domains 1 and 2, whereas non-neutralizing antibodies recognized random coils and loops that were primarily confined to folding domain 3. This suggests that neutralizing and non-neutralizing antibodies target distinct domains on RTA. In support of this hypothesis, we now report that the vast majority of conformation-dependent ricin neutralizing MAbs bind to RTA 1-33/44-198, a recombinant RTA deletion mutant lacking folding domain 3. Furthermore, preliminary studies in mice suggest that RTA 1-33/44-198 is more effective than an attenuated RTA point mutant at eliciting neutralizing antibodies. These data offer insights into the immunodominant and structural determinants on RTA that give rise to protective immunity, and for the first time provide an immunological rationale for ricin vaccine design.

Poster No. 8
Currently, there are no approved vaccines or therapeutics against the Category B select agent ricin. Ricin belongs to a family of A-B toxins, its B subunit (RTB) functions as a galactose-specific lectin to promote attachment and entry of the ricin enzymatic A subunit (RTA) into host cells. In efforts to design a therapeutic, we propose that the B subunit is an appealing target since antibodies directed against RTB will prevent the toxin’s entry into cells before intoxication can take place. In a screen of approximately 2,000 RTB-specific B cell hybridomas, a handful of neutralizing monoclonal antibodies (mAbs) were identified. A panel of four neutralizing mAbs SylH3, 24B11, JB4, and B/J F9 blocked ricin attachment to terminal galactose residues. These antibodies also prevented toxin binding to primary macrophages which suggested that they function by steric hindrance recognizing epitopes located adjacent to RTB’s galactose binding pockets located within two of RTB’s six sub-domains, 1a and 2g. In contrast, analysis of four non-neutralizing mAbs revealed recognition of epitopes situated within sub-domains of RTB which are not involved in galactose recognition (i.e., 1b, 2a). Most importantly, two of the neutralizing mAbs, SylH3 and 24B11, were shown by passive immunization to protect mice against a 5xLD$_{50}$ dose of ricin. Although all four neutralizing mAbs were shown to inhibit ricin attachment to terminal galactose residues, they differed in the degree of inhibition. The SylH3 and JB4 antibodies inhibited attachment by >80%, and the 24B11 and B/J F9 antibodies inhibited attachment by <40%. These results suggest that anti-RTB mAbs may function to neutralize ricin toxin intracellularly as well as through steric hindrance. Ongoing studies include passive administration of JB4 and B/J F9 to mice to determine protection against ricin. Additionally, we are also tracking intracellular toxin-Ab interactions via fluorescence microscopy.
Hsp70-mediated Nonclassical MHC Class Ib-restricted Anti-tumor T-cell Responses are 
Ag-dependent in the Frog Xenopus laevis

Hristina Nedelkovska and Jacques Robert 
Department of Microbiology and Immunology 
University of Rochester Medical Center, Rochester NY

The heat shock proteins (hsp) hsp70 are highly conserved molecular chaperones that elicit potent T cell responses against the antigens (Ags) they chaperone in both mammals and Xenopus. We have shown that frogs immunized with hsp70 generate CD8 T cell responses against the Xenopus thymic tumor 15/0 that expresses several nonclassical MHC class Ib genes, but not classical MHC class Ia. Therefore, we hypothesized that hsp70 can prime class Ib-mediated anti-tumor unconventional CD8 T cells in an Ag-dependent manner. To test this, we produced Xenopus recombinant tagged hsp70 proteins (both the cognate hsc73 and the inducible hsp72) from stable 15/0 tumor transfectants. Antigen binding in hsp70 is ATP dependent; therefore, peptides can be easily removed by ATP treatment. We used a cross-presentation assay to adoptively transfer hsp-pulsed antigen presenting cells and showed that both hsp72 and hsc73-Ag complexes, but not ATP treated Ag-negative hsp70s, elicit class Ib-mediated CD8 T cells responses resulting in protection from 15/0 tumor challenge. From expression studies, we further postulate that these responses involve one or a few class Ib gene products among which XNC10 is the prime candidate due to its lymphoid tissue distribution. To further elucidate the role of class Ib molecules in anti-tumor immunity, we developed a reverse genetic approach combining transgenesis and shRNA technology and obtained F1 transgenic Xenopus clones with silenced expression of β2-m. These transgenic clones in turn also have downregulated expression of class Ib molecules in vivo since interaction with β2-m is critical for their cell surface expression. In addition, we are currently in the process of generating transgenic animals with shRNAs targeting particular genes such as XNC10. This approach will not only demonstrate the importance of class Ib molecules in anti-tumor immunity but it will also allow us to distinguish which genes are critical for these interactions.

Research Support: NIH T32-AI 07285 (H.N.), 1R03-HD061671-01, R24-AI-059830-06

Poster No. 10
The role of the aryl hydrocarbon receptor (AhR) in the proper progression of an immune response has recently become a subject of focus in the immunological community. This cytosolic receptor is a transcription factor that translocates into the nucleus when bound by one of its ligands. Previous studies have examined the effects of AhR activation on CD4+ T cells in the context of acute exposure models wherein adult mice were treated with an AhR ligand prior to an immune challenge. Few studies have examined how inappropriate triggering of AhR during development alters the immune system; however, the studies to date suggest this is an important consideration. Using the pollutant 2,3,7,8 tetrachlorodibenzo-p-dioxin (TCDD) as a prototype AhR agonist, and influenza virus as a model antigen, we are currently investigating how developmental exposure to TCDD alters the function of the offspring’s immune system. We have found that developmental exposure to TCDD causes suppressed CD4+ T cell proliferation and IFNγ production. In contrast to IFNγ+ CD4+ T cells, the percent and number of CD4+ cells with a Treg phenotype did not appear to be affected by developmental exposure to TCDD. We plan to further characterize the CD4+ T cell response to influenza infection to elucidate the mechanism in which developmental AhR activation modulates the immune response.
Aryl Hydrocarbon Receptor Activation During Development Alters CD8+ T-cell Functional Response to Influenza A Virus

Bethany Winans and B. Paige Lawrence
Department of Environmental Medicine, University of Rochester Medical Center, Rochester NY

The developing immune system is susceptible to environmental insults, which can lead to altered immune function later in life. The aryl hydrocarbon receptor (AhR) is a transcription factor that plays a role in immune system development and function. Furthermore, the AhR acts as an environmental sensor, and among its numerous ligands binds many dioxins and polychlorinated biphenyls (PCBs), pollutants to which humans are constantly exposed. Human and animal data demonstrate that early life exposure to dioxins and PCBs leads to persistent alterations in immune function, supporting the idea that AhR activation impacts the developing immune system. Our laboratory has shown that mice exposed to a potent AhR agonist during gestation and lactation have persistent alterations in their immune response to influenza virus challenge at maturity. Specifically, these developmentally exposed mice have a reduced clonal expansion and activation of CD8+ T cells, and increased bronchopulmonary inflammation. However, it is not known how early life exposure to AhR agonists persistently alters immune function. We first sought to determine whether these alterations are due to direct AhR activation in the offspring, or if maternal activation of AhR alters the developing immune system of the offspring indirectly. Using both wild type and AhR knockout offspring of heterozygous dams, we show that the impaired CD8+ T cell response to influenza virus infection is the result of AhR activation in the developing offspring. Using bone marrow transplantation we determined that the impaired CD8+ T cell response tracks with the hematopoietic cell population, and that defective function persists even when the animals are no longer exposed to exogenous AhR agonists. These findings have led us to hypothesize that early life AhR activation impairs CD8+ T cell function via alterations in epigenetic regulation. Initial exploration of this idea reveals that CD8+ T cells from developmentally-exposed mice have increased levels of DNA methyltransferases (DNMTs), enzymes that regulate DNA methylation. We also have evidence that histone methylation is altered at the interferon-gamma (IFNγ) locus in these CD8+ T cells. These preliminary findings support the idea that early life activation of the AhR can reprogram the developing immune system, leading to persistent functional changes, and suggest that AhR may play a role in the epigenetic regulation of the immune system.

Poster No. 12
Extracellular Adenosine as a Master Regulator of Immune Cell Migration into the Central Nervous System via Induction of CX3CL1

Jeffrey H. Mills¹, Leah Alabanza¹, Deeqa Mahamed¹, and Margaret S. Bynoe¹
¹Department of Microbiology and Immunology, Cornell University College of Veterinary Medicine, Ithaca, NY 14853

Having the ability to modulate lymphocyte entry into the central nervous system (CNS) would benefit patients with neuroinflammatory diseases. We have previously shown that extracellular adenosine regulates CNS entry of lymphocytes during experimental autoimmune encephalomyelitis (EAE), the animal model for the CNS inflammatory disease multiple sclerosis. For instance, while extracellular adenosine levels are vastly increased following inflammatory cellular damage (from the hydrolysis of released cytoplasmic ATP by CD39 and CD73), mice lacking CD73 or given adenosine receptor (AR) antagonists have significantly reduced CNS lymphocyte entry during EAE. We now show through detailed genetic studies that AR signaling regulates lymphocyte migration into the CNS through induction of CX3CL1, a specialized chemokine that acts as both an adhesion molecule and chemoattractant for lymphocytes, monocytes, and NK cells. We show that AR signaling is necessary and sufficient to induce CNS expression of CX3CL1 (as compared to over 40 other chemokines and adhesion molecules). AR regulation of CX3CL1 is critical for EAE progression, as daily anti-CX3CL1 treatments prevent CNS lymphocyte infiltration during EAE. Importantly, AR signaling induces CX3CL1 expression on the choroid plexus, a known CNS entry point for lymphocytes. As AR signaling can promote lymphocyte transmigration across a choroid plexus transwell barrier, we conclude that adenosine is a master regulator of CNS lymphocyte entry.

F32 NS 066682 (J.H.M.) and R01 NS 063011 (M.S.B)
As an obligate intracellular pathogen, the apicomplexan parasite *Toxoplasma gondii* evades immune system-mediated clearance by undergoing stage differentiation to persist indefinitely in susceptible hosts. Previously, we found that mice deficient in the ecto-enzyme CD73, which generates adenosine in the extracellular matrix, were significantly resistant to acute and chronic toxoplasmosis after oral infection with the *T. gondii* type II ME49 strain. Resistance in CD73-knockout mice was due to a reduction in immune-mediated pathology in the intestine, and a delay in parasite differentiation in the CNS. To further clarify the role of CD73 and extracellular adenosine in *T. gondii* pathogenesis, we infected wildtype and CD73KO mice with ME49 cysts by the intraperitoneal (IP) route. In contrast to oral infection, IP-infected CD73KO mice were highly susceptible to immune-mediated pathology, with significantly increased infiltration of neutrophils and T cells into the peritoneal cavity. Peritoneal exudate cells from infected CD73KO mice generated higher levels of the inflammatory mediators nitric oxide, TNFa, and IL1b, without enhanced parasite killing or clearance. In addition, mice deficient in the adenosine receptors A1 or A2A were more susceptible to immunopathology during intraperitoneal infection with *T. gondii*, compared to wildtype mice. Thus extracellular adenosine is a key molecule that regulates the immune response to an intracellular pathogen and promotes host survival.
cAMP-dependent IL-10 Production Determines the Severity of Lyme Disease

Bikash Sahay, Rebeca Patsey, Kathleen Bashant, Timothy Sellati
Center for Immunology and Microbial Disease, Albany Medical College, Albany, NY 12208

*Borrelia burgdorferi* is the causative agent of Lyme disease in humans and susceptibility varies widely in the population, which can be efficiently mimicked in a murine infection model. While C57BL/6 (B6) mice are resistant to disease, C3H/HeN (C3H) mice are highly susceptible to Lyme Borreliosis, with features reminiscent of severe Lyme morbidity in humans. Disease resistance has been associated with the ability of macrophages (MΦ) from a B6 mouse to produce the anti-inflammatory cytokine IL-10, which is not adequately produced by C3H MΦ; rather, C3H MΦ produce interferon response factors such as *irf1*. Expression of *irfs* in C3H MΦ and *il10* in B6 MΦ is controlled by p38-dependent activation of STAT1 (Sahay 2009; Miller 2007) and STAT3 (Sahay 2011), respectively. Interestingly, both genotypes are equally efficient in activating *B. burgdorferi*-induced STAT1 and STAT3 but exhibit markedly different outcomes. Cyclic AMP (cAMP) is a second messenger that has been shown to inhibit the function of STAT1 without affecting its activation status and simultaneously activates cAMP response element binding protein (CREB) for IL-10 production. We evaluated cAMP levels between these two genotypes and found that B6 MΦ contain twice as much cAMP as compared to C3H MΦ. Upon exogenous addition of cAMP, C3H MΦ produced IL-10 in response to *B. burgdorferi* with a reduction in *irf1* transcription. Interestingly, inhibition of adenylate cyclase following recognition of *B. burgdorferi* did not reduce IL-10 release, suggesting that the initial accumulation, but not the generation, of cAMP is the primary difference between B6 and C3H MΦs. These findings suggest a role for cAMP in determining disease severity within the human population and represent a potential therapeutic target during Lyme disease.
Chronic Inflammation in a Mouse Model of Alzheimer’s Disease

Antje Krenz, Margaret S. Bynoe
Department of Microbiology and Immunology, Cornell University, Ithaca, NY

Neurodegenerative diseases share common characteristics in general. One major commonality between neurodegenerative diseases is the role of inflammation in disease pathogenesis. Multiple sclerosis (MS) is a neuroinflammatory disease of the central nervous system that is characterized by immune mediated damage to myelin and resultant disability. The hallmarks of Alzheimer’s disease (AD) are the accumulation of beta-amyloid protein into amyloid plaques and neurofibrillary tangles accompanied by chronic inflammation. Besides inflammation, blood-brain barrier (BBB) dysfunction is another common aspect of these two neurodegenerative disorders. It has been proposed that inflammation can modulate the clearance of beta-amyloid from the brain parenchyma via the BBB, leading to intracerebral accumulation of beta-amyloid. Experimental autoimmune encephalomyelitis (EAE) models the chronic inflammation found in MS and is commonly used as an animal model for MS.

We used the EAE mouse model to investigate the impact of chronic inflammation on AD disease progression in the transgenic APPswe/PS1dE9 mouse model of AD. APPswe/PS1dE9 mice, challenged with EAE, showed a typical course of the disease. Interestingly, APPswe/PS1dE9 mice had a significantly increased mortality compared to wild type littermates. This unusual response to EAE suggests that APPswe/PS1dE9 mice are more vulnerable to an autoimmune mediated pathology. We characterized this phenomenon further by comparing immune cell composition and cytokine profiles as well as demyelination and lymphocyte infiltration into the CNS of APPswe/PS1dE9 and wild type mice.

Levels of beta-amyloid deposition as plaques and cerebral amyloid angiopathy as well as axonal dystrophy and BBB permeability of aged EAE-challenged mice APPswe/PS1dE9 compared to naïve, age-matched APPswe/PS1dE9 mice will be analyzed to determine whether an early chronic inflammatory event like EAE exacerbates these AD symptoms.

Poster No. 16
Activated Protein C (APC) is an anticoagulant that is involved in the complex two-way interaction between the coagulation and immune systems. APC is widely known to be anti-inflammatory; most notably it has been clinically developed for treatment of severe sepsis. The anti-inflammatory properties of APC include conferring endothelial barrier protection, thus limiting the extravasation of inflammatory immune cells into tissues. Given that the major pathological component of multiple sclerosis (MS) and its animal model, experimental autoimmune encephalomyelitis (EAE), is the extravasation of autoreactive immune cells into the central nervous system (CNS) as a result of blood brain barrier (BBB) dysfunction, we investigated whether inhibiting endogenous APC can affect the pathogenesis of EAE. We observed that blocking APC in the circulation during EAE resulted in considerable immune cell infiltration in the CNS. Interestingly, however, these mice exhibited mild to no outward signs of EAE as assessed by paralysis severity. The anti-APC treated mice also had significantly higher percentage of CD4+Foxp3+ cells both in the CNS and in the periphery compared to controls. Moreover, the cytokine profile of leukocytes from the anti-APC mice was skewed to T helper-2, which is known to be protective in EAE. These data show that lowered APC levels during EAE can lead to divergent effects on EAE pathogenesis, specifically leading to increased BBB dysfunction while simultaneously inducing immunological conditions that are non-pathogenic in EAE.
mTOR Inhibition Increases Dendritic Cell Lifespan and Improves Autologous DC Vaccine Efficacy in a Melanoma Tumor Model

Eyal Amiel, Bart Everts, Tori Freitas, Irah King, Edward Pearce
Trudeau Institute, Saranac Lake, NY

In vitro generated Dendritic cells (DCs) are a major tool used in autologous cell vaccine therapies. The short lifespan of DCs following their activation is a major hurdle of DC vaccine therapeutic approaches. Recent reports demonstrate that genetic modulation DC longevity can improve the potency of DC vaccines in animal tumor models. In this work, we assess the role of mTOR inhibition in regulating DC lifespan and activation, and assessing whether this approach can improve the efficacy of autologous DC vaccination in establishing anti-tumor immunity. Our data demonstrate that mTOR inhibition improves survival of TLR-activated DCs and prolongs expression of co-stimulatory molecules on these cells. Furthermore, autologous DC vaccination with rapamycin-treated DCs results in improved generation of antigen-specific CD8+ T-cells in vivo and improved anti-tumor immunity compared to conventionally activated DCs.
Role of MMP3 in Tumor Progression and IRF8 Mediated Regulation of MMP3 Function

Debarati Banik and Dr. Scott Abrams
Dept. of Immunology, Roswell Park Cancer Institute

Matrix metalloproteinases (MMP) are involved in tissue remodeling under various physiological or pathological conditions. A Stromelysin family member MMP3 (or Str1) is significantly involved in several physiological processes mediating enzymatic modification on a broad range of substrates. In cancer, MMP3 has demonstrated a significant role in the initiation of tumor development. However, whether MMP3 plays important roles in the stages of tumor progression and metastasis, remain unexplored. Therefore our first aim is to test the hypothesis that MMP3 can be a determining factor in neoplastic progression, focusing on primary tumor growth and metastasis. We approached the problem using loss and gain of function methods. We used a sarcoma cell line (CMS4) that showed detectable levels of MMP3 expression at both mRNA and protein levels. Thus far, our results showed that: 1) silencing MMP3 with shRNA in the parental tumor population significantly reduced local tumor growth, and 2) over-expression of MMP3 significantly increased tumor growth, which suggests that MMP3 can affect the neoplastic process beyond initiation. Next we plan to extend our study to experimental or spontaneous metastasis models to determine if MMP3 is causally linked to metastatic events. Our next aim is to identify a novel mechanism of regulation of MMP3. Ongoing studies in our lab suggest that the transcription factor, Interferon regulatory factor-8 (IRF-8), serves as a negative regulator of MMP3. IRF8 previously has been shown to mediate antitumor responses in both hematopoietic and non-hematopoietic models. Disruption of IRF8 in CMS4 cells significantly augments its aggressiveness in vivo. A number of experimental data (differential microarray, RT-PCR, qPCR and Western blot analyses) revealed up-regulation of MMP3 as a result of alterations in IRF8 function in CMS4 cells. MMP3 reporter assays reveal that IRF8 is a negative regulation of MMP3 transcription; CMS4 cells experimentally lacking IRF8 were more efficient in driving MMP3 promoter activity than CMS4 expressing IRF8. Thus we hypothesize that IRF-8 behaves as a negative regulator of MMP3 by either directly binding to MMP3 promoter or indirectly through interactions with other IRF-8 binding partners. Ongoing studies are testing the hypothesis that altered MMP3 activity, at least in part, accounts for differences in tumor growth between IRF-8-deficient and IRF-8-expressing tumor cells. Understanding the importance of MMP3 and its regulation may have important implications for cancer therapy.

Poster No. 19
Allergic Lung Inflammation and Susceptibility to *Streptococcus pneumoniae*

Alan M. Sanfilippo and Dennis W. Metzger
Albany Medical College, Albany, NY

*Streptococcus pneumoniae* is the most common cause of bacterial pneumonia which results in 175,000 hospitalizations as well as 40,000 deaths in the United States annually. With the introduction of a conjugate vaccine, overall disease burden from *S. pneumoniae* has been on the decline. However, recent work has identified a link between asthma and invasive pneumococcal disease (IPD) prompting the Advisory Committee on Immunization Practices to recommend that all adults with asthma receive the pneumococcal conjugate vaccine. To study the underlying mechanism responsible for an increased risk of IPD for individuals with asthma, we used an acute murine model of ovalbumin (OVA)-induced allergic lung inflammation. We hypothesized that mice with acute OVA-induced allergic lung inflammation would be more susceptible than naïve mice to *S. pneumoniae*. Following OVA challenge, BALB/c mice were infected intranasally (i.n.) with a lethal dose of *S. pneumoniae* and morbidity and mortality were monitored daily. Bronchoalveolar lavage fluids and lung tissue samples were obtained for cytokine and histological analysis following lung function testing using mechanical ventilation. Surprisingly, mice with acute allergic lung inflammation were less susceptible to infection than mice that did not receive OVA sensitization and challenge. Histological analysis showed that following OVA challenge, inflammatory infiltration was most apparent immediately following the final challenge, though not completely cleared 10 days later. We then tested IPD resistance following chronic allergen challenge and found that chronic OVA challenged mice lost the resistance to IPD seen using the acute OVA challenge model. Finally, we tested acute OVA sensitized and challenged mice of different BALB/c substrains and found disease resistance to vary. These results suggest a complex association between asthma and IPD.
Inflammasome Activation by *Francisella tularensis* in Human Cells is Mediated in Part by NLRP3

*Ellen B. Duffy*, Maninjay K. Atianand, and Jonathan A. Harton
Albany Medical College, Albany, NY

*Francisella tularensis* (*Ft*), an intracellular bacterium, causes the often fatal respiratory disease tularemia. The low infectious dose and inhalational route of infection makes *Ft* a potential biothreat agent. An understanding of how *Ft* overcomes the innate immune response during intracellular growth is important to facilitating development of immunization strategies. Towards this end, we examined the role of various cytoplasmic sensors, mostly members of the NOD-like receptor (NLR) family, of pathogen-associated molecular patterns (PAMPS) that are necessary to elicit a pro-inflammatory response. One member of this family, NLRP3, is known to respond to several intracellular signals including reactive oxygen species (ROS), K⁺ ion efflux, and Cathepsin B release from damaged lysosomal compartments, all of which are known or likely consequences of cellular *Ft* infection. Using genetic reconstitution of NLR-specific inflammasomes in human epithelial cells, visualization of inflammasome "speck" formation, and siRNA mediated knock-down in the human THP-1 monocyte cell line, we implicate NLRP3 as a sufficient sensor of *Francisella* infection. We further test the effect(s) of established inhibitors of the NLRP3 inflammasome on *Francisella* induced IL-1beta production in PMA-differentiated THP-1 cells. Human epithelial cells showed increased IL-1beta production and speck formation upon transfection with NLRP3, but not with other NLRs. siRNA mediated knock-down of NLRP3 THP-1 cells decreased IL-1beta production. In all cases, addition of an inhibitor of NLRP3 inflammasome showed a decrease in IL-1beta production by at least 50% relative to the control samples. While recent reports implicate Aim2 as the sole mediator of IL-1beta response against *Ft* infection in mouse macrophages, activated by cytosolic DNA, we demonstrate a role for NLRP3 inflammasome in human cells.

Poster No. 21
Attendee Contact Information

Alabanza, Leah
Cornell University
201-362-8776
lma53@cornell.edu

Amarasinghe, Jayaleka
Wadsworth Center
518-402-4081
jja07@wadsworth.org

Amiel, Eyal
Trudeau Institute
518-891-3080
eamiel@trudeauinstitute.org

Banik, Debarati
Roswell Park Cancer Institute
716-845-3352
debarati.banik@roswellpark.org

Bellville, Dawn
Albany Medical College
518-262-5365
bellvid@mail.amc.edu

Berwin, Brent
Dartmouth Medical School
603-650-6899
berwin@dartmouth.edu

Boule, Lisbeth
University of Rochester
585-275-2013
lisbeth_boule@urmc.rochester.edu

Brincks, Erik
Trudeau Institute
518-891-3080
ebrincks@trudeauinstitute.org

Bynoe, Margaret
Cornell University
607-253-1023
msb76@cornell.edu

Byrne, Katelyn
Dartmouth Medical School
603-653-3688
katelyn.t.byrne@dartmouth.edu

Chan, Gary
SUNY Upstate Medical University
315-464-7682
chang@upstate.edu

Chin, Shu Shien (Mandy)
University at Buffalo
716-881-7490
shuchin@buffalo.edu

Denkers, Eric
Cornell University
607-253-4022
eyd1@cornell.edu

Drake, James
Albany Medical College
518-262-9337
drakej@mail.amc.edu

Duffy, Ellen
Albany Medical College
518-262-0052
duffye@mail.amc.edu

Elliott, Michael
University of Rochester
585-273-4793
michael_elliott@urmc.rochester.edu

Evans, Sharon
Roswell Park Cancer Institute
716-845-3421
sharon.evans@roswellpark.org

Fiering, Steven
Dartmouth Medical School
603-653-9966
fiering@dartmouth.edu
Foti, Shawn
Johns Hopkins University
617-899-6401
sfoti1@jhu.edu

Furuya, Yoichi
Albany Medical College
518-262-6220
furuyay@mail.amc.edu

Gerber, Scott
University of Rochester
585-275-6747
scott_gerber@urmc.rochester.edu

Gorgone, Darci
BD Biosciences
781-820-9896
darci_gorgone@bd.com

Harden, Jamie
University at Buffalo
716-829-2681
jlharden@buffalo.edu

Harton, Jonathan
Albany Medical College
518-262-4445
hartonj@mail.amc.edu

Hayes, Sandy
SUNY Upstate Medical University
315-464-7692
hayessa@upstate.edu

Jang, Meishaiang
Astellas Research Institute
847-933-7435
Mei-shiang.jang@us.astellas.com

Jones, Derek
Wadsworth Center
518-486-4393
djones@wadsworth.org

Kalia, Vandana
Pennsylvania State University
814-863-8533
vkalia@psu.edu

Kennedy, Jeff
Wadsworth Center
518-486-4395
jsk07@health.state.ny.us

King, Christine
SUNY Upstate Medical University
315-464-5465
kingch@upstate.edu

Kirimanjeswara, Girish
Pennsylvania State University
814-863-5350
gsk125@psu.edu

Krenz, Antje
Cornell University
607-253-4024
akrenz@cornell.edu

Laird, Renee
SUNY Upstate Medical University
315-464-7690
lairdr@upstate.edu

Lefebvre, Julie
Trudeau Institute
518-891-3080
jlefebvre@trudeauinstitute.org

Leifer, Cynthia
Cornell University
607-200-4117
cal59@cornell.edu

Lee, Kelvin
Roswell Park Cancer Institute
716-845-4106
kelvin.lee@roswellpark.org
Lim, Joanne  
University of Rochester  
585-478-9325  
joanne.h_lim@urmc.rochester.edu

Lord, Edith  
University of Rochester  
585-275-5855  
edith_lord@urmc.rochester.edu

MacNamara, Katherine  
Albany Medical College  
518-262-0921  
macnamk@mail.amc.edu

Mahamed, Deeqa  
Cornell University  
607-253-4052  
dam267@cornell.edu

Mantis, Nicholas  
Wadsworth Center, NYSDOH  
518-402-2750  
nmantis@wadsworth.org

Metzger, Dennis  
Albany Medical College  
518-262-6750  
metzged@mail.amc.edu

Mills, Jeffrey  
Cornell University  
607-253-4052  
jhm49@cornell.edu

Nedelkovska, Hristina  
University of Rochester  
585-330-9575  
hristina_nedelkovska@urmc.rochester.edu

Norbury, Christopher  
Pennsylvania State University  
717-531-7204  
cen1@psu.edu

O’Hara, Joanne  
Wadsworth Center  
845-901-6787  
joanne.m.ohara@gmail.edu

Patankar, Yash  
Dartmouth Medical School  
603-650-6899  
yash.patankar@dartmouth.edu

Pelletier, Marianne  
Merck  
518-791-0008  
marianne_pelletier@merck.com

Periasamy, Sivakumar  
Albany Medical College  
518-496-8026  
periasa@mail.amc.edu

Princiotto, Michael  
SUNY Upstate Medical University  
315-464-7683  
princiom@upstate.edu

Sahay, Bikash  
Albany Medical College  
518-262-8141  
sahayb@mail.amc.edu

Sanfilippo, Alan  
Albany Medical College  
518-462-6220  
sanfila@mail.amc.edu

Sedlacek, Abigail  
University of Rochester  
585-275-6747  
abigail_sedlacek@urmc.rochester.edu
Sellati, Timothy
Albany Medical College
518-262-8140
sellatt@mail.amc.edu

Simmons, Katrina
Wadsworth Center, 518-210-6077
ksimmons004@gmail.com

Sun, Keer
Albany Medical College
518-262-6220
sunk@mail.amc.edu

Thanavala, Yasmin
Roswell Park Cancer Institute
716-845-8536
yasmin.thanavala@roswellpark.org

Vance, Dave
Wadsworth Center
978-877-1435
djv05@wadsworth.org

Ventullo, Stephanie
BD Biosciences
617-306-6028
stephanie_ventullo@bd.com

Winans, Bethany
University of Rochester
585-275-2013
bethany_winans@urmc.rochester.edu

Winslow, Gary
Wadsworth Center, NYSDOH
518-473-2795
gary.winslow@wadsworth.org

Yates, Jennifer
Wadsworth Center
518-339-2556
huntingt@wadsworth.org

Yermakova, Anastasiya
Wadsworth Center
518-402-4081
anastasiya.yermakova@gmail.com

Zhang, Yi
Dartmouth Medical School
603-650-6899
yi.zhang@dartmouth.edu
Authors Index
O - Oral Poster Presentation
P# - Poster Number
S - Speaker

Abrams, Scott ...................................................................................................................................... 36, 83
Alabanza, Leah M. (P#17) ................................................................................................................. 35, 40, 77, 81
Amiel, Eyal (P#18) ............................................................................................................................... 58, 81, 82
Amarasinghe, Jayaleka J. (S) ............................................................................................................. 62
Appenheimer, Michelle M. .................................................................................................................. 44
Atianand, Maninjay K. ....................................................................................................................... 85
Baird, Jay ........................................................................................................................................... 28
Banik, Debarati (O, P#19) .................................................................................................................. 36, 83
Bashant, Kathleen .............................................................................................................................. 59, 79
Bauman, Heinz ................................................................................................................................... 44
Bergsagel, P. Leif .................................................................................................................................. 32
Boise, Lawrence H. ............................................................................................................................ 32
Boule, Lisbeth A. (P#11) ................................................................................................................... 75
Brey, Robert N. .................................................................................................................................. 72
Brincks, Erik L. (S) ............................................................................................................................ 39
Bynoe, Margaret S. (S) ....................................................................................................................... 35, 40, 77, 78, 80, 81
Byrne, Katelyn T. (S) ......................................................................................................................... 38
Bzik, David ........................................................................................................................................ 28
Carlson, Louise M. ............................................................................................................................. 32
Chae, Wook-Jin ................................................................................................................................... 40
Chan, Gary C. (S) .................................................................................................................................. 50
Chan-Khan, Asher A. ............................................................................................................................ 32
Chen, Qing ......................................................................................................................................... 44
Chin, Shu Shien (S) .............................................................................................................................. 29
Cohen, Sara B. .................................................................................................................................... 55
Conejo-Garcia, Jose .............................................................................................................................. 28
Cookenham, Tres .................................................................................................................................. 39
Deluio, Greg .......................................................................................................................................... 60
Denkers, Eric Y. (S) ............................................................................................................................ 55
Duffy, Ellen B. (P#21) ....................................................................................................................... 85
<table>
<thead>
<tr>
<th>Name</th>
<th>Page Numbers</th>
</tr>
</thead>
<tbody>
<tr>
<td>Eaton, Sheri M.</td>
<td>45, 66</td>
</tr>
<tr>
<td>Egan, Charlotte E.</td>
<td>55</td>
</tr>
<tr>
<td>Egilmez, Nejat</td>
<td>42</td>
</tr>
<tr>
<td>Elliott, Michael R. (S)</td>
<td>56</td>
</tr>
<tr>
<td>Evans, Sharon S. (S)</td>
<td>44</td>
</tr>
<tr>
<td>Everts, Bart</td>
<td>82</td>
</tr>
<tr>
<td>Fairfax, Keke C. (S)</td>
<td>58</td>
</tr>
<tr>
<td>Fiering, Steven (S)</td>
<td>28</td>
</tr>
<tr>
<td>Fisher, Daniel T</td>
<td>44</td>
</tr>
<tr>
<td>Forsyth, Peter</td>
<td>65</td>
</tr>
<tr>
<td>Freitas, Tori</td>
<td>82</td>
</tr>
<tr>
<td>Furuya, Yoichi (S)</td>
<td>54</td>
</tr>
<tr>
<td>Garett-Sinha, Lee Ann</td>
<td>29</td>
</tr>
<tr>
<td>Gerber, Scott A. (S)</td>
<td>30, 34, 69, 70</td>
</tr>
<tr>
<td>Gorgone, Darci (S)</td>
<td>52</td>
</tr>
<tr>
<td>Green, Nathanile</td>
<td>26</td>
</tr>
<tr>
<td>Harden, Jamie L. (S)</td>
<td>42</td>
</tr>
<tr>
<td>Harton, Jonathan A.</td>
<td>85</td>
</tr>
<tr>
<td>Hayes, Sandra M.</td>
<td>47, 68</td>
</tr>
<tr>
<td>Haynes, Laura</td>
<td>45, 66</td>
</tr>
<tr>
<td>John, Shinu</td>
<td>29</td>
</tr>
<tr>
<td>Jones, Derek D. (S)</td>
<td>60</td>
</tr>
<tr>
<td>Jones, Maura</td>
<td>48, 71</td>
</tr>
<tr>
<td>Kalia, Vandana (S)</td>
<td>41</td>
</tr>
<tr>
<td>Keifer, Kerstin</td>
<td>26</td>
</tr>
<tr>
<td>King Christine A. (S)</td>
<td>51</td>
</tr>
<tr>
<td>King, Irah L.</td>
<td>58, 82</td>
</tr>
<tr>
<td>Kirimanjeswara, Girish S. (S)</td>
<td>61</td>
</tr>
<tr>
<td>Kohlmeier, Jacob E.</td>
<td>29</td>
</tr>
<tr>
<td>Koorella, Chandana</td>
<td>32</td>
</tr>
<tr>
<td>Krenz, Antje (P#16)</td>
<td>80</td>
</tr>
<tr>
<td>Laird, Renee M.</td>
<td>47, 68</td>
</tr>
<tr>
<td>Lanthier, Paula A.</td>
<td>45, 66</td>
</tr>
<tr>
<td>Lawrence, B. Paige</td>
<td>75, 76</td>
</tr>
<tr>
<td>Lefebvre, Julie S. (O, P#2)</td>
<td>45, 66</td>
</tr>
<tr>
<td>Lee, Kelvin P. (S)</td>
<td>32</td>
</tr>
<tr>
<td>Lim, Joanne (O, P#6)</td>
<td>34, 70</td>
</tr>
</tbody>
</table>
Lord, Edith M. ................................................................. 34, 69, 70
Lun, Xueqing ................................................................. 65
Mack, Matthias ............................................................. 55
Mahamed, Deeqa (P#14) ............................................... 35, 77, 78
Mantis, Nicholas J. ......................................................... 46, 62, 72, 73
Marias, Andrea ............................................................. 54
Marshak-Rothstein, Ann (S) ......................................... 26
Maue, Alexander C. .................................................... 47, 68
Maurer, Kirk J. ............................................................ 55
McCormick, Craig ........................................................ 51
Mcfadden, Grant ........................................................... 65
Metzger, Dennis W. ..................................................... 54, 61, 84
Mills, Jeffrey H. (O, P#13) ................................................ 35, 77, 78
Mody, Christopher ....................................................... 65
Mohrs, Markus ............................................................ 58
Moody, Krishna ........................................................... 26
Mueller, Cynthia .......................................................... 40
Muhitch, Jason B. ........................................................ 44
Nair, Jayakumar ........................................................... 32
Nedelkovska, Hristina (P#10) ....................................... 74
O’Hara, Joanne M. (P#8) ................................................ 72
Patsey, Rebeca ............................................................ 59, 79
Pearce, Edward J. ........................................................ 58, 82
Petell, Ashlee H. .......................................................... 45, 66
Racine, Rachael ........................................................... 48, 71
Repasky, Elizabeth A. .................................................. 44
Robert, Jacques ........................................................... 74
Roberts, Alan D. .......................................................... 39
Roberts, Sean ............................................................... 54
Rochford, Rosemary .................................................... 47, 68
Rose-John, Stefan ....................................................... 44
Rozanski, Cheryl H. .................................................... 32
Russell, Lisa ................................................................. 29
Sahay, Bikash (S, P#15) .................................................. 79
Samuelson, Elizabeth M. (O, P#4) ................................. 47, 68
Sanfilippo, Alan (P#20) ................................................ 84
Sarkar, Surojit .............................................................. 41
Schoenberger, Stephen P. ................................................................. 32
Sedlacek, Abigail L. (P#5) ................................................................. 69
Sellati, Timothy J. ............................................................................ 59, 79
Simmons, Katrina (O, P#3) .............................................................. 33, 67
Simpson, Kenneth W. ................................................................. 55
Skitzki, Joseph J. ............................................................................. 44
Solomon, Benjamin D. ................................................................. 40
Turk, Mary Jo ................................................................................. 38
Vardam, Trupti D. ........................................................................... 44
Welsh, Jo Ellen ................................................................................ 67
Winans, Bethany (P#12) ................................................................. 75, 76
Winslow, Gary ................................................................................ 48, 60, 71
Woodland, David L. ....................................................................... 39
Yates, Jennifer (O, P#7) ................................................................. 48, 71
Yermakova, Anastasiya (O, P#9) .................................................... 46, 73
Zhang, Jiqing .................................................................................. 65
Zhou, Lei .......................................................................................... 44
<table>
<thead>
<tr>
<th>General Information</th>
<th>Yes</th>
<th>No</th>
</tr>
</thead>
<tbody>
<tr>
<td>This was my first year attending the Upstate New York Immunology Conference (NYIC).</td>
<td></td>
<td></td>
</tr>
<tr>
<td>I would recommend this Conference to fellow colleagues and/or students.</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Even if I leave my current location, I would consider attending this Conference in the future.</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Conference Organization</th>
<th>Yes</th>
<th>No</th>
</tr>
</thead>
<tbody>
<tr>
<td>Web site information was updated regularly</td>
<td></td>
<td></td>
</tr>
<tr>
<td>The scientific content was diversified</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Keynote presentations were of personal value</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Graduate Students and Postdoctoral Fellows were given ample opportunities to present their research and interact with PIs</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Poster Session/Vendor Fair was well organized</td>
<td></td>
<td></td>
</tr>
<tr>
<td>The overall meeting was well organized and met or exceeded my expectations.</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Anticipated Outcomes</th>
<th>Yes</th>
<th>No</th>
</tr>
</thead>
<tbody>
<tr>
<td>I have established a new contact with someone at the Conference that I believe will lead either to collaboration or future information sharing, data or techniques.</td>
<td></td>
<td></td>
</tr>
<tr>
<td>I learned something specific at this Conference that I anticipate will save me time or money, or accelerate a research project or goal.</td>
<td></td>
<td></td>
</tr>
<tr>
<td>I have made leads about current and/or future positions/openings and anticipate making further contacts.</td>
<td></td>
<td></td>
</tr>
<tr>
<td>I had an opportunity to present my research by either presenting a poster or giving an oral presentation, which resulted in thought-provoking questions that will enhance my research project.</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Since last year’s Conference…</th>
<th>Yes</th>
<th>No</th>
<th>Did not attend</th>
</tr>
</thead>
<tbody>
<tr>
<td>I have been in contact with someone on a current or future collaboration.</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>I have collaborated with one or more attendees, based on contact(s) made.</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>I shared information with colleagues who did not attend last year’s Conference.</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>I have submitted or published data related to topics discussed during last year’s Conference.</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Since last year’s Conference… | Yes | No | Did not attend
---|---|---|---
A new idea or concept (either openly stated or created in your mind), which I became aware of during the Conference, has altered the direction of my research. | | | |
There has been a major advance in this area of research, either by me or others in the field, based upon an idea or concept that was revealed or conceived during the Conference. | | | |
I have used something specific that I learned at last year’s Conference that has saved me time or money, or has accelerated a research objective. | | | |
Someone with whom I established contact during the Conference has been helpful to my research or career. | | | |
I have obtained a Postdoctoral Fellowship or Faculty position through networking among institutions represented at the Conference. | | | |
Do you plan to attend the 15th Annual Upstate New York Immunology Conference? Yes No
Suggestions for future Keynote Speakers (Name and Institution):

Comments/Suggestions:

The following questions are voluntary/optional. The information here is confidential and represent statistics only. This information is gathered solely for the purpose of applying for the NYIC grant renewal. If you choose to answer, please check any/all that apply.

| I prefer not to respond | Check or leave blank |
| | |

<table>
<thead>
<tr>
<th>Race</th>
<th>Check or leave blank</th>
</tr>
</thead>
<tbody>
<tr>
<td>White</td>
<td></td>
</tr>
<tr>
<td>Asian</td>
<td></td>
</tr>
<tr>
<td>More than one race</td>
<td></td>
</tr>
<tr>
<td>Black or African American</td>
<td></td>
</tr>
<tr>
<td>Native Hawaiian or Pacific Islander</td>
<td></td>
</tr>
<tr>
<td>American Indian/Alaskan</td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Ethnicity</th>
<th>Check or leave blank</th>
</tr>
</thead>
<tbody>
<tr>
<td>Non-Hispanic or Latino</td>
<td></td>
</tr>
<tr>
<td>Hispanic or Latino</td>
<td></td>
</tr>
</tbody>
</table>

OPTIONAL

Name:  
Date: