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Message from the Chairman

It is a great pleasure for me to welcome you to the Sue Golding Graduate Division (SGGD) and our Department of Microbiology and Immunology at the Albert Einstein College of Medicine. Our laboratories in the Department of Microbiology and Immunology cover a wide range of topics spanning the fields of virology, bacteriology, host immune responses to infection and cancer, inflammation and autoimmune disease. The department warmly and enthusiastically welcomes PhD candidates and postdoctoral fellows who are interested in pursuing research projects and potentially developing careers in these areas. I welcome all new students in the SGGD and postdoctoral candidates to contact me or any of the department's faculty directly to explore the terrific range of opportunities available to you in the Department of Microbiology and Immunology.

My best wishes for success in your new adventure!

Professor & Chairman
Department of
Microbiology and Immunology

MICROBIOLOGY AND IMMUNOLOGY

FACULTY
2019 – 2020

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MICROBIOLOGY AND IMMUNOLOGY

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DR. JOAN W. BERMAN

Dr. Berman's laboratory examines the mechanisms that mediate HIV entry into the CNS and how viral and inflammatory mediators damage neurons and other CNS cells. More than 40 million people worldwide are HIV infected. As a result of antiretroviral therapies, HIV infected people are living longer. HIV enters the CNS early after infection and despite therapy, persists within the CNS. Prevalence of NeuroAIDS and its associated cognitive impairment is increasing. An understanding of mechanisms that mediate these effects are critical to the development of therapeutic strategies.

HIV infection of the CNS can have devastating consequences, often resulting in cognitive impairment and severe neurological complications. The basis of this impairment is poorly understood. Although its development is associated with early viral infiltration of the CNS, the number of activated monocytes/macrophages within the CNS appears to be a better indicator of neurologic compromise than viral load, suggesting that leukocyte infiltration and cognitive impairment are tightly correlated. How infected monocytes cross the blood brain barrier (BBB) and infiltrate the CNS is not well understood. This process is critical to the development of NeuroAIDS as it brings leukocytes into the brain where they activate and infect microglia, and effect damage to the BBB and other CNS cells. The mechanisms of HIV-infected monocyte transmigration across the BBB have only been minimally characterized. We are characterizing several of the steps in this transmigration process using a tissue culture model of the human BBB. We analyze the mechanisms that mediate attachment and diapedesis of HIV-infected monocytes across the BBB to identify markers that contribute to brain infection and BBB disruption, such as adhesion molecules, tight junction and adherens proteins, chemokines and their receptors. The lab has a major translational component, examining sera and CSF from HIV infected individuals for predictors of cognitive impairment, as well as patient cells for unique markers of this impairmentand for their ability to transmigrate across the blood brain barrier. We examine tissue from HIV-infected individuals for altered proteins. The overall goal is to identify targets for therapeutic intervention to limit the entry of HIV into the CNS.

Many HIV-infected people who abuse drugs have more extensive CNS damage associated with significant cognitive impairment. As many drugs of abuse cause an increase in extracellular dopamine, we examine the effects of dopamine on HIV infection of macrophages. We demonstrated that dopamine increases HIV infection of human macrophages and are addressing the mechanisms by which dopamine causes this increase as well as alterations in macrophage function. We also study the impact of buprenorphine and methadone,therapies for Opiate abuse, in the context of NeuroAIDS.

PUBLICATIONS

Eugenin EA, Martiney JA, Berman JW. (2019) The malaria toxin hemozoin induces apoptosis in human neurons and astrocytes: potential role in the pathogenesis of cerebral malaria. Brain Res. 2019 Jul 2:146317. doi: 10.1016/j.brainres.2019.146317.
PMID: 31276637

Williams, DW, Calderon, TM, Lopez, L, Carvallo, L, Gaskill, PJ, Eugenin, EA, Moregllo, S, Berman, JW. (2013) Mechanisms of HIV Entry Into the CNS: Increased Sensitivity of HIV Infected CD14+CD16+ Monocytes to CCL2 and Key Roles of CCR2, JAM-A, and ALCAM in Diapedesis. PLOS One. Jul 26;8(7):e69270.

Williams, DW, Eugenin, EA, Calderon, TM, Berman, JW. (2012) Monocyte Maturation, HIV

Susceptibility, and Transmigration Across the Blood Brain Barrier are Critical in HIV Neuropathogenesis. Journal of Leukocyte Biology, Mar;91 (3):401-15.

Buckner CM, Calderon TM, Williams DW, Belbin TJ, Berman JW. (2011) Characterization of monocyte maturation/differentiation that facilitates their transmigration across the blood-brain barrier and infection of HIV: Implications for NeuroAIDS. *Cell Immunol;* 267(2):109-23.

Eugenin E.A., Clements J.E, Zink M, and Berman J.W.(2011) HIV infection of human astrocytes disrupts blood brain barrier integrity by a gap junction dependent mechanism. The Journal of Neuroscience. 31(26):9456-65.

King JE, Eugenin EA, Hazleton JE, Morgello S and Berman JW (2010) Mechanisms of HIV Tat Induces phosphorylation of NMDA Receptor Subunit 2A in Human Primary Neurons: Implications for NeuroAIDS Pathogenesis. *Am.J Pathology*, June17:2819-30.

Roberts TK, Eugenin EA, Morgello S, Clements JE, Zink MC and Berman JW, (2010) PrP^{C}, the Cellular Isoform of the Human Prion Protein, is a Novel Biomarker of HIV-Associated Neurocognitive Impairment and Mediates Neuroinflammation, *Am. J.Pathology*, Oct 2010; 177(4):1848-60.

Gaskill PJ, Calderon TM, Luers AJ, Eugenin EA, Javitch JA, Berman JW, (2009) Human Immunodeficiency Virus (HIV) Infection of Human Macrophages Is Increased by Dopamine. A Bridge between HIV-Associated Neurologic Disorders and Drug Abuse,.*Am J Pathol*. September 175(3):1148-59.

Eugenin EA, Osieki K, Lopez L, Goldstein H, Calderon TM, Berman JW, (2006) CCL2/Monocyte chemoattractant protein-1 mediates enhanced transmigration of human immunodeficiency virus (HIV)-infected leukocytes across the blood-brain barrier: a potential mechanisms of HIV-CNS invasion and NeuroAIDS. *J Neurosci* 26(4):1098-1106.

Dr. Michael Berney

The Berney lab seeks to understand how the human pathogen *Mycobacterium tuberculosis* adapts bioenergetically and metabolically to the host environment. Using an integrated approach of genetic engineering, metabolomics, biochemical assays, transcriptomics and animal experiments we are pursuing fundamental questions such as which nutrients and energy sources do pathogenic microorganisms use in the host, or how do aerobic pathogens survive in host tissues that are hypoxic. Our goal is to identify vulnerable pathways that can be exploited for drug discovery.

Suffocating *M. tuberculosis*

In recent years the energy generating machinery of *M. tuberculosis* has received considerable attention as a new drug target space resulting in the discovery of some promising new (and old) drug candidates. The most promising candidate drugs are inhibitors of the ATP synthase (Bedaquiline) and bc_1-cytochrome *c* reductase (Q203). However, these drugs have relatively low to no early bactericidal acitivity (first 7 days) and we hypothesized that this is partially due to cytochrome *bd* oxidase and its ability to uncouple electron flux from proton pumping. Recently we demonstrated that inactivation of the terminal oxidase Cyt-*bd* (ΔcydAB) leads to rapid killing and clearance of *Mtb ΔcydAB* in mouse lungs treated with the inhibitor Q203 (Kalia et al. 2017, *PNAS*). Our goals are now to understand the role of Cyt-*bd* in TB pathogenesis and to develop drugs that exploit this synthetic lethality.

Trapping *M. tuberculosis* in its nutritional autonomy.

Co-evolution of pathogens and host has led to many different metabolic strategies employed by intracellular pathogens to deal with the immune response and the scarcity of food during infection. Therefore, it is not a surprise that maintenance of metabolic homeostasis contributes to the virulence of intracellular pathogens. Recently we identified a metabolic vulnerability of *M. tuberculosis* that leads to persister-free killing *in vitro* and *in vivo* (Berney et al. 2015, *PNAS*). We could show that methionine biosynthesis, a branch of the aspartate family amino acid biosynthesis (the “aspartate pathway”), is essential for host infections, because *M. tuberculosis* is unable to scavenge methionine from the host. The aspartate pathway is a core anabolic pathway that provides the cell with the proteinogenic amino acids lysine, threonine, isoleucine and methionine, the peptidoglycan precursor diaminopimelate and S-adenosylmethionine, the main cofactor in cellular methyltransferase reactions. Sensing and regulating the abundance of these essential intermediates allows *M. tuberculosis* to integrate information from cell wall biosynthesis, translation and one carbon metabolism to maintain cellular homeostasis. Interestingly, aspartate and glutamate, the two essential precursors of this pathway are both thought to be taken up from the host, which links the regulation of this pathway to external signals from the host. Such metabolic interplay between *M. tuberculosis* and the host are still poorly understood. In this project we study the metabolic regulation and vulnerability of the aspartate pathway during active and latent mouse infections, determine intermediates that are exchanged in crosstalk with the host and investigate the allosteric and transcriptional control that is needed to maintain pathway homeostasis and guarantee balanced growth of *M. tuberculosis*.

Publications

Mycobacterium tuberculosis' capacity to survive iron-starvation might enable it to persist in iron-deprived microenvironments of human granulomas.
Kurthkoti K, Amin H, Marakalala MJ, Ghanny S, Subbian S, Sakatosb A, Livnye J, Fortune SM, **Berney M**, Rodriguez GM.
mBio (2017 IN PRESS)

Exploiting the synthetic lethality between terminal respiratory oxidases to kill *Mycobacterium tuberculosis* and clear host infection.
Kalia NP, Hasenoehrl EJ, Ab Rahman NB, Koh VH, Ang MLT, Sajorda DR, Hards K, Grüber G, Alonso S, Cook GM, **Berney M*,+**, Pethe K*,+.
Proceedings of the National Academy of Sciences (*EE June 26th* 2017) (+equal contributions, *corresponding author)

Mycobacterium tuberculosis in the face of host-imposed nutrient limiation.
Berney M* & Berney-Meyer L.
In ***Tuberculosis and the Tubercle Bacillus, Second Edition*****,** ASM Press. (2017) Jun;5(3).

OXPHOS as a new target space for tuberculosis: success, failures, and future directions
Cook GM, Hards K, Greening C, Heikal A, Dunn E, Nakatani Y, Pethe K, Crick D, **Berney M.**
In ***Tuberculosis and the Tubercle Bacillus, Second Edition***, ASM Press. (2017) Jun;5(3).

Central Role of Pyruvate Kinase in Carbon Co-Catabolism of Mycobacterium tuberculosis.
Tahel Noy, Olivia Vergnolle, Travis E Hartman, Kyu Y Rhee, William R Jacobs Jr., **Michael Berney***, and John S Blanchard*
Journal of Biological Chemistry (2016) 291/13: 7060–7069

Essential roles of methionine and S-adenosylmethionine in the autarkic lifestyle of Mycobacterium tuberculosis
Berney M*, Berney-Meyer L, Wong K, Chen B, Chen M, Kim J, Wang J, Chan J, Wang F, Jacobs WR*.
Proceedings of the National Academy of Sciences (2015) 112: 10008–10013
Featured with a spotlight article in Trends in Microbiology

An obligately aerobic soil bacterium activates hydrogen production to survive reductive stress during hypoxia
Berney M*, Greening C, Conrad R., Jacobs WR, Cook GM.
Proceedings of the National Academy of Sciences (2014) 111: 11479-11484
Featured with an interview in The Pathologist (https://thepathologist.com/issues/0114/the-survival-artists/)

Full publist: https://www.ncbi.nlm.nih.gov/myncbi/michael.berney.1/bibliography/40584568/public/)

DR. ROBERT BURK

Human Papillomavirus (HPV) and Microbiome Translational Science: Molecular Epidemiology, Pathogenesis and Evolution.

The main focus of the Burk laboratory is to understand viral-host evolution and the emergence of HPV types that are highly pathogenic and cause multiple cancers in humans (e.g., cervix and oropharynx). In addition, the lab is also testing hypotheses on the role of epigenetic changes in the viral and host (human) genome and its relationship to precancer and cancer development. Concomitant with viral-host relationships is a new emerging area of interest, the cervical microbiome/mycobiome and its relationship to viral-host interactions and other outcomes. These investigations extend from clinical studies where we obtain exfoliated cervix cellular material (Pap cells) and evaluate the HPV genome, CpG methylation and the composition of the microbiome to cell based biochemistry studies of viral proteins and molecular evolution of viral sequences.

Papillomaviruses are 8.0 kb double stranded DNA viruses readily amenable to amplification and sequencing, making this system ideal as a model for DNA virus evolution and identification of pathogenic genetic signatures. Over 200 HPV types exist and further characterization of HPVs infecting the population (i.e., from our large sample repository) have allowed us to explore the virus as a species, characterization of the frequency and heterogeneity of HPV types and variants in the population, and the role of viral evolution in pathogenicity. The lab uses phylogenic methods and other analytic strategies to test hypotheses about the relationship and characteristics of HPV genomes and disease. Exploration of natural selection of papillomaviruses has led us to the conclusion that the viruses are evolving through complex means yet to be discovered. Our major collaboration with investigators at the National Cancer Institute, NIH has provided an ideal translational team of world-class epidemiologists, biostatisticians and clinical investigators. In combination with evolutionary biologists at the American Museum of Natural History, our integrative group provides a unique prospective for intellectual growth of students that want to “think outside the box”. More recently, we have investigated epigenetic changes to the HPV genome and have demonstrated very significant results on the association of these changes with neoplastic progression. The identification of HPV in specific biological niches has challenged us to explore the microbiome through barcoding and parallel sequencing using Next-Gen methods. We have recently developed the methodologies and computer software to test hypothesis on the influence of the micriobiome on cervix cancer development in HPV positive women.

Other research areas include a human genetic project to identify the gene(s) for hyperhidrosis (excessive sweating). This is a fascinating disorder that is strongly associated with a family history of excessive sweating. There seems to be at least two phenotypes, excessive sweating from the palms and soles, and excessive sweating from the underarms, body, face and groin areas. The analyses of families with this disorder suggest genetic and/or allelic heterogeneity. To date, we have collected over 1500 DNA samples from affected individuals and families. We are in the process of exome-sequencing the coding regions using Next-Gen technologies to identify candidate mutations associated with this disorder.

Lastly, as part of our goal to understand genes and cancer, we have for many years studied the von Hippel-Lindau (VHL) gene that is a driver of kidney cancer. We investigate the function of the VHL protein, in part, an oxygen sensor. Our recent observations indicate that intact VHL is required for primary cilia formation and function in renal cells. We have localized VHL and other

proteins known to interact to cilium. Further studies will investigate the function of VHL in the cilium.

PUBLICATIONS

Amaro-Filho SM, Gradissimo A, Usyk M, Moreira FCB, de Almeida LM, Moreira MAM, Burk RD. HPV73 a nonvaccine type causes cervical cancer. Int J Cancer. 2019 Apr 9. doi: 10.1002/ijc.32315. PMID: 30963559

Ho, G.Y.F., Bierman, R., Beardsley, L., C.J. Chang, and **Burk, R.D.** Natural history of cervicovaginal papillomavirus infection in young women. N. Engl. J. Med. 338:423-428, 1998.

Chen, Z., Terai, M., Fu, L., Herrero, R., DeSalle, R. and **Burk, R.D**. Diversifying selection in human papillomavirus type 16 (HPV16) lineages based on complete genome analysis. J. Virol. 79:7014-7023, 2005.

Schiffman, M., Herrero, R., DeSalle, R., Hildesheim, A., Wacholder, S., Rodriguez, A.C., Bratti, M.C., Sherman, M.E., Morales, J., Guillen, D., Alfaro, M., Hutchinson, M., Wright, T.C., Solomon, D., Chen, Z., Schussler, J., Castle, P.E. and **Burk, R.D.** The carcinogenicity of human papillomavirus types reflects viral evolution. Virology 337:76-84, 2005.

Bottalico, D., Chen, Z., Dunne, A., Ostoloza, A., McKinney, S., Sun, C., Schlecht, N.F., Fatahzadeh, M., Herrero, R., Schiffman, M. and **Burk, R.D.** The oral cavity contains abundant known and novel Human papillomaviruses from the *Betapapillomavirus* and *Gammapapillomavirus* genera. J. Inf. Dis. 204:787-792, 2011. PMID: 21844305; PMCID: PMC3156102.

Chen, Z., Schiffman, M., Herrero, R., DeSalle, R., Anastos, K., Segondy, M., Sahasrabuddhe, V., Gravitt, P.E., Hsing, A. and **Burk, R.D.** Evolution and taxonomic study of HPV16-related *Alphapapillomavirus* variant genomes: HPV31, HPV33, HPV35, HPV52, HPV58 and HPV67. PLoS ONE 6(5): e20183. PMID:2167391.

Smith, B., Chen, Z., Reimers, L., van Doorslaer, K., Schiffman, M., DeSalle, R., Herrero, R., Yu, K., Wang, T. and **Burk R.D.** Sequence imputation of HPV16 genomes for genetic association studies. PLoS ONE 6(6):e21375. Epub 2011 Jun 23. PMID:21731721.

Smith, B.C., McAndrew, T., Chen, Z., Harari, A., Barris, D.M., Viswanathan, S., Rodriguez, A.C., Castle, P., Herrero, R., Schiffman, M. and **Burk, R.D.** The cervical microbiome over 7 years and a comparison of methodologies for its characterization. PLoS One. 2012;7(7):e40425. PMID: 22792313.

Burk, R.D., Harari, A. and Chen, Z. Human papillomavirus genome variants. Virology 445:232-243, 2013. PubMed PMID: 23998342; PMCID: PMC3979972.

Mirabello, L., Frimer, M., Harari, A., McAndrew, T., Smith, B. Chen, Z., M., Wentzensen, Wacholder, S., Castle, P.E., Raine-Bennett, T., Schiffman, M. and **Burk, R.D.** HPV16 methyl-haplotypes determined by a novel next-generation sequencing method are associated with cervical precancer. Int. J. Cancer 136:E146-53, 2015. PMID: 25081507.

Van Doorslaer, K., DeSalle, R., Einstein, M.H. and **Burk, R.D.** Degradation of Human PDZ-

proteins by Human Alphapapillomaviruses represents an evolutionary adaptation to a novel cellular niche. PLoS Pathog. 2015 Jun 18;11(6):e1004980. PubMed PMID: 26086730.

Harari, A., Chen, Z. and **Burk, R.D.** Human papillomavirus genomics: past, present and future. Curr. Probl. Dermatol. 45:1-18, 2014. PubMed PMID: 24643174; PMCID: PMC4430864.

Stern, J.M., Moazami, S., Qiu, Y., Kurland, I., Chen, Z., Agalliu, I., Burk, R., Davies, K.P. Evidence for a distinct gut microbiome in kidney stone formers compared to non-stone formers. Urolithiasis. 2016 PubMed PMID: 27115405.

Agalliu, I., Gapstur, S., Chen, Z., Wang, T., Anderson, R.L., Teras, L., Kreimer, A.R., Hayes, R.B., Freedman, N.D., Burk, R.D. Associations of Oral α-, β-, and γ-Human Papillomavirus Types With Risk of Incident Head and Neck Cancer. JAMA Oncol. 2:599-606, 2016. PubMed PMID: 26794505; PMCID: PMC4956584.

Harari, A., Chen, Z., Rodríguez, A.C., Hildesheim, A., Porras, C., Herrero, R. Wacholder, S., Panagiotou, O.A., Befano, B., *Burk, R.D. and *Schiffman, M. Cross-protection of the bivalent human papillomavirus (HPV) vaccine against variants of genetically related high-risk HPV infections. J. Infect. Dis. 213:939-47, 2016. PMID: 26518044; PMCID: PMC4760417.

Mirabello, L., Yeager, M., Cullen, M., Boland, J.F., Chen, Z., Wentzensen, N., Zhang, X., Yu, K., Yang, Q., Mitchell, J., Roberson, D., Bass, S., Burdette, L., Raine-Bennett, T., Lorey, T., Castle, P.E., Burk, R.D.*, Schiffman, M.* HPV16 sublineage associations with histology-specific cancer risk using HPV whole-genome sequences in 3,200 women. J. Natl. Cancer Inst. 2016 Apr 29;108(9). PMID: 27130930

Other studies (VHL, hyperhidrosis and prostate cancer)

Lutz, M.S. and **Burk, R.D.** Primary cilium formation requires von Hippel-Lindau gene function in renal derived cells. Cancer Res. 66:6903-6907, 2006. (Highlighted article in July 15, 2006 Cancer Res.)

Keller, S.M., Bello, R., Vibert, B., Swergold, G. and **Burk, R**. Diagnosis of palmar hyperhidrosis via questionnaire without physical examination. Clin. Auton. Res. 19:175-181, 2009.

DR. JOHN CHAN

Our lab studies how *Mycobacterium tuberculosis* (Mtb), the causative agent of tuberculosis (TB), interacts with the host; on-going studies include:

Tuberculous latency and reactivation: Gene profiling studies have identified a set of Mtb genes (the dormancy regulon) that are regulated by exposure to low oxygen tension and nitric oxide, two latency-promoting environmental signals. Deletion of a dormancy regulon gene, *rv2623* (a member of the USP [**u**niversal **s**tress **p**rotein] family), results in hypervirulence in infected hosts. Rv2623 also regulates Mtb growth in vitro. The growth-regulating attribute has been linked to the ATP-binding capacity of Rv2623. Understanding how Rv2623 regulates the growth of Mtb and how members of the dormancy regulon promote latency is a focus of our laboratory.

We are collaborating with JoAnn Tufariello to study the role of the five Mtb Rpf proteins, putative peptidoglycan hydrolyases, in regulating tuberculous latency and reactivation. Mutagenesis studies showed that Mtb doubly deficient for RpfA and RpfB displays defects in persistence and reactivation, both in in vivo and in vitro models of tuberculous latency and disease recrudescence. Studies have been initiated to characterize how Mtb Rpf's regulate mycobacterial growth.

The mechanisms that regulate the granulomatous reaction: Our Studies using the B cell-deficient μMT mouse model and specific Fcγ receptor knockout mice have shown that humor immune response regulates the tuberculous granulomatous reaction and is required for optimal control of Mtb. There is also evidence that B cells can regulate T cell response in infected hosts. We are conducting experiments aimed at characterizing the roles of B cells in regulating the host granulomatous response to Mtb.

TNF is essential in controlling TB. However, TNF also contributes to the development of immunopathology. TNF neutralization reactivates TB in both mice and humans with persistent infection. We are using this TNF neutralization reactivation model to study the role of TNF in regulating the tuberculous granulomatous reaction. We are also generating cell-specific conditional TNF knockout mouse strains that will be used to decipher the relative contribution of cell-specific TNF in the protective immunity and immunopathogenesis of this cytokine in the host.

TB vaccines: We will apply the knowledge gained from the above-listed investigation to rationally design safe and effective TB vaccines in collaboration with the Jacobs and Porcelli labs. For example, understanding how to manipulate the humoral response to the advantage of the host may lead to the development of vaccines with increased efficacy. Knowing how Mtb regulates host production of TNF may lead to the design of Mtb-derived vaccines with enhanced immunogenicity.

PUBLICATIONS

Drumm JE, Mi K, Bilder P, Sun M, Lim J, Bielefeldt-Ohmann H, Basaraba R, So M, Zhu G, Tufariello JM, Izzo AA, Orme IM, Almo SC, Leyh TS, **Chan J**. 2009. Mycobacterium tuberculosis universal stress protein Rv2623 regulates bacillary growth by ATP-Binding: requirement for establishing chronic persistent infection. PLoS Pathog 5:e1000460

Russell-Goldman E, Xu J, Wang X, **Chan J**, Tufariello JM. 2008. A double Rpf knockout Mycobacterium tuberculosis strain exhibits profound defects in reactivation from chronic tuberculosis and innate immunity phenotypes. Infect Immun. 76:4269-81.

Maglione PJ, Xu J, Casadevall A, **Chan J**. 2008. Fcgamma receptors regulate immune activation and susceptibility during Mycobacterium tuberculosis Infection. J Immunol 180:3329-38.

Chakravarty SD, Zhu G, Tsai MC, Mohan VP, Marino S, Kirschner DE, Huang L, Flynn J, **Chan J**. 2008. Tumor necrosis factor blockade in chronic murine tuberculosis enhances granulomatous inflammation and disorganizes granulomas in the lungs. Infect Immun. 76:916-26.

Dao DN, Sweeney K, Hsu T, Gurcha, SS, Nascimento IP, Roshevsky D, Besra GS, **Chan J**, Porcelli S, Jacobs WR Jr. 2008. Mycolic acids of trehalose dimycolate modified by MmaA4 repress IL-12p40 production. Plos Pathogens 4(6):e1000081.

Maglione PJ, Xu J, **Chan J**. 2007. B cells moderate inflammatory progression and enhance bacterial containment upon pulmonary challenge with *Mycobacterium tuberculosis*. J Immunol 178:7222-34.

Chakravarty SD, Xu J, Lu B, Gerard C, Flynn J, **Chan J**. 2007. The chemokine receptor CXCR3 attenuates the control of chronic Mycobacterium tuberculosis infection in BALB/c mice. J Immunol 178:1723-35.

Hinchey J, Lee S, Jeon BY, Basaraba RJ, Venkataswamy MM, Chen B, **Chan J**, Braunstein M, Orme IM, Derrick SC, Morris SL, Jacobs Jr WR, Porcelli SA. 2007. Enhanced priming of adaptive immunity by a proapoptotic mutant of *Mycobacterium tuberculosis*. J Clin Invest 117:2279-88.

DR. KARTIK CHANDRAN

As our world grows more interconnected and humans impinge on the few remaining wild habitats, infections caused by the accidental transmission of viruses from their natural animal hosts to humans are increasingly of concern. The unprecedented 2013–2015 Ebola virus disease epidemic in western Africa provides a particularly apt example. Few specific antiviral treatments are available for Ebola and other emerging agents, and our ability to develop them is challenged by a poor understanding of exactly how viruses co-opt our own cells at the molecular level.

The Chandran Lab at Einstein strives to understand this molecular warfare between virus and cell, and to apply what we learn to the development of antiviral treatments. Filoviruses, such as Ebola virus and Marburg virus, and hantaviruses, such as Sin Nombre virus and Hantaan virus, are major topics of study in our group. Working collaboratively with our partners on three continents, we have helped uncover critical host factors required for cell invasion by Ebola virus, including the long-sought viral receptor, Niemann-Pick C1 (NPC1). One of our recent interests is to decipher how genetic variation in host-encoded factors—in NPC1, for example—can influence the susceptibility of humans and animals to viral infection and the likelihood of animal-to-human 'host-jumping' events. Our ongoing efforts also include translational studies to develop anti-Ebola drugs, including small molecule and antibody therapeutics, targeting host factors critical for viral invasion.

SELECTED PUBLICATIONS

Mechanism of Ebola virus entry and infection:

Bornholdt ZA, Ndungo E, Fusco ML, Flyak AI, Crowe JE Jr, **Chandran K*** and Saphire EO. 2016. Host-primed Ebola virus GP exposes a hydrophobic NPC1 receptor-binding pocket, revealing a target for broadly neutralizing antibodies. *mBio* 7. pii: e02154-15.

Spence JS, Krause TB, Mittler E, Jangra RK, **Chandran K***. 2016. Direct visualization of Ebola virus fusion triggering in the endocytic pathway. *mBio* 7. pii: e01857-15.

Ng M, Ndungo E, Kaczmarek ME, Herbert AS, Binger T, James R, Jangra RK, Hawkins JA, Gifford RJ, Biswas R, Demogines A, Kuehne AI, Yu M, Brummelkamp TR, Drosten C, Wang LF, Kuhn JH, Müller MA, Dye JM, Sawyer SL, and **Chandran K***. 2015. NPC1 contributes to species-specific patterns of filovirus susceptibility in bats. *eLife* 4. pii: e11785.

Miller EH, Obernosterer G, Raaben M, Herbert AS, Deffieu M, Krishnan A, Ndungo E, Sandesara RG, Carette JE, Kuehne AI, Ruthel G, Pfeffer SR, Dye JM*, Whelan SP*, Brummelkamp TR*, **Chandran K***. 2012. Ebola virus entry requires the host-programmed recognition of an intracellular receptor. *EMBO J* 31:1947-1960.

Carette JE, Raaben M, Wong AC, Herbert AS, Obernosterer G, Mulherkar N, Kuehne AI, Kranzusch PJ, Griffin AM, Ruthel G, Dal Cin P, Dye JM, Whelan SP, **Chandran K***, Brummelkamp TR. 2011. Ebola virus entry requires the cholesterol transporter Niemann-Pick C1. Nature 477:340-343.

Coté M, Misasi J, Ren T, Bruchez A, Lee K, Filone CM, Hensley L, Li Q, Ory D, **Chandran K**, Cunningham J*. 2011. Small molecule inhibitors reveal Niemann-Pick C1 is essential for ebolavirus infection. *Nature* 477:344-348.

Chandran K, Sullivan NJ, Felbor U, Whelan SP, Cunningham JM. 2005. Endosomal proteolysis of the Ebola virus glycoprotein is necessary for infection. Science 308:1643-1645.

Mechanism of hantavirus entry and infection:

Kleinfelter LM, Jangra RK, Jae LT, Herbert AS, Mittler E, Stiles KM, Wirchnianski AS, Kielian M, Brummelkamp TR, Dye JM, and **Chandran K***. 2015. Haploid Genetic Screen Reveals a Profound and Direct Dependence on Cholesterol for Hantavirus Membrane Fusion. *mBio* 6:e00801-15.

Antiviral strategies and therapeutics:

Wec AZ, Herbert, AS, Murin CD, Nyakatura EK, Abelson DM, Fels JM, He S, James RM, de La Vega M-A, Zhu W, Bakken RR, Goodwin E, Turner HL, Jangra RK, Zeitlin L, Qiu X, Lai JR, Walker LM, Ward AB, Dye JM, **Chandran K***, Bornholdt ZA. 2017. Antibodies from a human survivor define sites of vulnerability for broad protection against ebolaviruses. *Cell* 169:878-890.e15.

Wec AZ, Nyakatura EK, Herbert AS, Howell KA, Holtsberg FW, Bakken RR, Mittler E, Christin JR, Shulenin S, Jangra RK, Bharrhan S, Kuehne AI, Bornholdt ZA, Flyak AI, Saphire EO, Crowe JE Jr, Aman MJ, Dye JM, Lai JR, **Chandran K***. 2016. A "Trojan horse" bispecific antibody strategy for broad protection against ebolaviruses. *Science* pii: aag3267.

DR. JOHANNA P. DAILY

Our primary research interest is in pathogenesis in the *Plasmodium falciparum.* Patients infected with this parasite can be completely asymptomatic or develop severe disease resulting in death. The goal of our research has been to define the molecular mechanisms that underlie this variation in disease outcomes in *P. falciparum.* Toward this goal, we have developed a new pathogenesis model through the analysis of *in vivo* parasite biology and associated host factors using a whole genome approach. We have identified novel parasite biology when it resides in the human host; this biology has not been reported under *in vitro* cultivation and may play a role in enhanced virulence and/or transmission capacity. Using longitudinal studies we have characterized individual responses to malaria during a severe and subsequent mild infections to understand how patient's immune responses may change and lessen immunopathology. The long term goal is to identify parasite and host processes involved in disease to serve as targets for vaccine or chemotherapeutic development. We carry out field based translational studies in cohorts infected with malaria in Africa and these inform our experimental work using molecular biology, whole transcriptional, metabolomic and cellular approaches in the laboratory.

I. Define factors that protect against parasite sequestration to human brain microvasculature.

II. Define host immunity associated with protection from clinical disease.

III. Comprehensive analysis of host and parasite small molecules to study pathogenesis and develop biomarkers of cerebral malaria associated brain swelling.

Selected Publications

Gupta A, Seydel K, Miranda M, Feintuch C, Saidi A, Kim R, Birbeck G, Taylor T, **Daily JP**. Extensive Alterations of Blood Metabolites in Pediatric Cerebral Malaria. 2017 PloS ONE *12(4):e0175686.*

Feintuch CM, Saidi A, Seydel K, Chen G, Goldman-Yassen A, Mita N, Kim RS, Frenette PS, Taylor T, **Daily JP.** Activated neutrophils are associated with pediatric cerebral malaria vasculopathy in Malawian children. mBio 2016; 16 February, vol 7. No 1 e01300-15

Pappa V, Seydel K, Gupta S, Feintuch CM, Potchen MJ, Kampondeni S, Goldman-Yassen A, Veenstra M, Lopez L, Kim RS, Berman JW, Taylor T, **Daily JP.** Lipid metabolites of the phospholipase A2 pathway and inflammatory cytokines are associated with brain volume in paediatric cerebral malaria. Malar J. 2015 Dec 21;14(1):513.

Subramaniam KS, Spaulding E, Ivan E, Mutimura E, Kim RS, Liu X, Dong C, Feintuch CM, Zhang X, Anastos K, Lauvau G, **Daily JP**. The T cell inhibitory molecule BTNL2 is upregulated in mild Plasmodium falciparum infection and is protective during experimental cerebral malaria. J Infect Dis. 2015. Oct 15;212(8):1322-31.

Subramaniam KS, Skinner J, Ivan E, Mutimura E, Ryung S. Kim RS, Feintuch C, Portugal S, Anastos K, Crompton PD, **Daily JP.** HIV malaria co-infection is associated with atypical memory B cell expansion and a reduced antibody response to a broad array of Plasmodium falciparum antigens in Rwandan adults PLoS One. 2015 Apr 30;10(4):e0124412.

Lakshmanan V, Rhee KY, Wang W, Yu Y, Khafizov K, Fiser A, Wu P, Ndir O, Mboup S, Ndiaye D, **Daily JP**. Metabolomic Analysis of Patient Plasma Yields Evidence of Plant-Like α-Linolenic Acid Metabolism in Plasmodium falciparum. *J Infect Dis.* 2012 Jul;206(2):238-48.

Krupka M, Seydel K, Manix C, Yee K, Kim R, Lin C, Calder R, Petersen C, Taylor T, **Daily J**. Mild *Plasmodium falciparum* malaria following an episode of severe malaria is associated with induction of the interferon pathway in Malawian children. *Infection and Immunity* 2012;80(3):1150-5

LeRoux M, Lakshmanan V and **DailyJP.** *Plasmodium falciparum* biology: analysis of *in vitro* versus *in vivo* growth condition , Trends in Parasitology 2009 Oct;25(10):474-81.

Daily JP. Novel in vivo parasite biology--implications for pathogenesis.Pediatric Res. 2008 Apr; 63(4):339.

Daily JP, Scanfeld D, Pochet N, Le Roch K, Plouffe D, Kamal M, Sarr O, Mboup S, Ndir O, Wypij D, Levasseur K, Thomas E, Tamayo P, Dong C, Zhou Y, Lander ES, Ndiaye D, Wirth D, Winzeler EA, Mesirov JP, Regev A. Distinct physiological states of Plasmodium falciparum in malaria-infected patients. Nature. 2007 Dec 13;450(7172):1091.

DR. TERESA DiLORENZO

Type 1 diabetes is an organ-specific autoimmune disease characterized by T cell-mediated destruction of the insulin-producing beta cells of the pancreatic islets. While insulin therapy allows for continuation of life, it neither cures the disease nor prevents its devastating complications. Studies utilizing the nonobese diabetic (NOD) mouse model of the disease have shown that T cells, recognizing autoantigenic peptides bound to major histocompatibility complex (MHC) molecules, are absolutely required for disease development. T cells specific for beta cell antigens can also be detected in the peripheral blood and islets of type 1 diabetes patients. Our laboratory utilizes an extensive collection of mouse models to investigate the antigenic specificities, pathogenicity, and immunobiology of T cells in type 1 diabetes. These models are also being used to develop and optimize therapeutic strategies. New, increasingly "humanized" mouse models are continually in development in our group, as are strategies to utilize human samples to translate our findings to patients.

Our research program seeks to develop an improved understanding of the immunopathogenesis of type 1 diabetes, with the objective of applying new knowledge to the goals of improved diagnostic, monitoring, and therapeutic strategies for this disease. Our efforts have resulted in several important contributions in this area. Our laboratory was the first to identify islet-specific glucose-6-phosphatase catalytic subunit-related protein (IGRP) as a major beta cell antigen recognized by CD8 T cells in NOD mice. In work that was inspired by our discovery in NOD mice, peptides derived from IGRP were subsequently shown to be T cell targets in patients. Our group has also been a leader in characterizing humanized NOD-based models of type 1 diabetes. Our study of HLA-A*02:01-transgenic NOD mice led to the identification of new T cell epitopes of IGRP and insulin, several of which have since been shown to be markers of beta cell autoreactivity in HLA-A*02:01-positive patients. We developed and characterized models in which HLA-A*02:01 expression is accompanied by reduced thymic expression of insulin, as is seen in patients, resulting in improved humanized models for type 1 diabetes. We discovered that mature human CD8 and CD4 T cells can be "reprogrammed" to be beta cell-specific upon transduction with a lentivirus encoding a T cell receptor that recognizes a beta cell antigen; this will allow human T cells to be incorporated into future models of type 1 diabetes. We were the first to explore the potential of delivery of beta cell antigens to steady-state dendritic cells *in vivo* in NOD mice for the purpose of inducing tolerance. Using antigen-linked anti-DEC-205, we showed that transferred T cell receptor-transgenic T cells could be deleted, and endogenous beta cell-specific T cells reduced in number, even in the context of ongoing autoimmunity in NOD mice having known T cell tolerance defects. We identified undersized peptides as previously unknown targets of autoreactive CD8 T cells and devised a strategy for their identification as antigens in autoimmune disease. Two United States patents related to our work have been granted.

PUBLICATIONS

Lieberman, S. M., A. M. Evans, B. Han, T. Takaki, Y. Vinnitskaya, J. A. Caldwell, D. V. Serreze, J. Shabanowitz, D. F. Hunt, S. G. Nathenson, P. Santamaria, and **T. P. DiLorenzo**. 2003. Identification of the β cell antigen targeted by a prevalent population of pathogenic $CD8^+$ T cells in autoimmune diabetes. *Proc. Natl. Acad. Sci. USA 100:8384-8388.*

Mukhopadhaya, A., T. Hanafusa, I. Jarchum, Y. G. Chen, Y. Iwai, D. V. Serreze, R. M. Steinman, K. V. Tarbell, and **T. P. DiLorenzo**. 2008. Selective delivery of β cell antigen to dendritic cells *in vivo* leads to deletion and tolerance of autoreactive $CD8^+$ T cells in NOD mice. *Proc. Natl. Acad. Sci. USA 105:6374-6379.*

Jarchum, I., and **T. P. DiLorenzo**. 2010. Ins2 deficiency augments spontaneous HLA-A*0201-restricted T cell responses to insulin. *J. Immunol. 184:658-665.*

Brims, D. R., I. Jarchum, L. Mikesh, E. Palmieri, U. A. Ramagopal, V. N. Malashkevich, R. J. Chaparro, T. Lund, M. Hattori, J. Shabanowitz, D. F. Hunt, S. G. Nathenson, S. C. Almo, and **T. P. DiLorenzo**. 2010. Predominant occupation of the class I MHC molecule H-2K^{wm7} with a single self peptide suggests a mechanism for its diabetes-protective effect. *Int. Immunol. 22:191-203.*

Samanta, D., G. Mukherjee, U. A. Ramagopal, R. J. Chaparro, S. G. Nathenson, **T. P. DiLorenzo***, and S. C. Almo. 2011. Structural and functional characterization of a single-chain peptide-MHC molecule that modulates both naive and activated CD8$^+$ T cells. *Proc. Natl. Acad. Sci. USA 108:13682-13687*. ***co-corresponding author**

Antal, Z., J. C. Baker, C. Smith, I. Jarchum, J. Babad, G. Mukherjee, Y. Yang, J. Sidney, A. Sette, P. Santamaria, and **T. P. DiLorenzo**. 2012. Beyond HLA-A*0201: New HLA-transgenic nonobese diabetic mouse models of type 1 diabetes identify the insulin C-peptide as a rich source of CD8$^+$ T cell epitopes. *J. Immunol. 188:5766-5775.*

Mukherjee, G., A. Geliebter, J. Babad, P. Santamaria, D. V. Serreze, G. J. Freeman, K. V. Tarbell, A. Sharpe, and **T. P. DiLorenzo**. 2013. DEC-205–mediated antigen targeting to steady-state dendritic cells induces deletion of diabetogenic CD8 T cells independently of PD-1 and PD-L1. *Int. Immunol. 25:651-660*.

Lamont, D., G. Mukherjee, P. R. Kumar, D. Samanta, C. G. McPhee, T. W. H. Kay, S. C. Almo, **T. P. DiLorenzo***, and D. V. Serreze. 2014. Compensatory mechanisms allow undersized anchor-deficient class I MHC ligands to mediate pathogenic autoreactive T cell responses. *J. Immunol. 193:2135-2146*. ***co-corresponding author**

Babad, J., G. Mukherjee, A. Follenzi, R. Ali, B. O. Roep, L. D. Shultz, P. Santamaria, O. O. Yang, H. Goldstein, D. L. Greiner, and **T. P. DiLorenzo**. 2015. Generation of β cell-specific human cytotoxic T cells by lentiviral transduction and their survival in immunodeficient HLA-transgenic mice. *Clin. Exp. Immunol. 179:398-413.*

Mukherjee, G., R. J. Chaparro, J. Schloss, C. Smith, C. D. Bando, and **T. P. DiLorenzo**. 2015. Glucagon-reactive islet-infiltrating CD8 T cells in NOD mice. *Immunology 144:631-640.*

Babad, J., R. Ali, J. Schloss, and **T. P. DiLorenzo**. 2016. An HLA-transgenic mouse model of type 1 diabetes that incorporates the reduced, but not abolished, thymic insulin expression seen in patients. *J. Diabetes Res. 2016:7959060*.

Sidney, J., J. Schloss, C. Moore, M. Lindvall, A. Wriston, D. F. Hunt, J. Shabanowitz, **T. P. DiLorenzo**, and A. Sette. 2016. Characterization of the peptide binding specificity of the HLA class I alleles B*38:01 and B*39:06. *Immunogenetics 68:231-236.*

Ali, R., J. Babad, A. Follenzi, J. A. Gebe, M. A. Brehm, G. T. Nepom, L. D. Shultz, D. L. Greiner, and **T. P. DiLorenzo.** 2016. Genetically modified human CD4$^+$ T cells can be evaluated in vivo without lethal graft versus host disease. *Immunology 148:339-351*.

DR. DAVID FOOKSMAN

The goals of my laboratory are to understand the regulation of plasma cell differentiation, migration, survival and function. Plasma cells are terminally-differentiated B cells that secrete high-affinity antibodies constitutively, following immunization and exposure to a pathogen. The quality, magnitude and longevity of the antibody response are dependent upon the differentiation and survival of these cells, which involves many signaling factors and auxiliary cell types. We have used intravital two-photon imaging to study plasma cell differentiation and migration in the lymph node and have found that that these cells exhibit a highly linear migration that is independent of $g_{\alpha i}$ chemotaxis. This migration is unique among lymphocytes and enables these cells to travel long distances crossing heterogeneous microenvironments to reach niches critical for their survival. In some cases, plasma cells may undergo malignant transformation during differentiation leading to neoplasms in humans such as multiple myeloma. Despite their critical role in immune function and disease, many fundamental questions remain regarding the physiology of plasma cells in vivo. We are using two-photon intravital imaging in combination with modern cellular and immunological tools to visualize and better understand the physiology of these cells under normal and pathological conditions. The current topics in the laboratory are focused on:

1. Plasma cell differentiation. What factors regulate selection and differentiation of germinal center B cells to plasma cell?
2. What factors control plasma cell migration to the bone marrow and subsequent long-lived survival and retention?
3. What factors control myeloma cell retention and migration in the bone marrow, which enables tumor progression?

PUBLICATIONS

(* corresponding author, # senior author)

Pitt L, Tikhonova A, Trimarchi T, King, B, Hu H, Gong, Y, Tsirigos A, Littman DR, Ferrando A, Morrison S, **Fooksman DR#**, Aifantis I#, Schwab S#. CXCL12-producing vascular endothelial niches control acute T cell leukemia maintenance. Cancer Cell. 2015 Jun 8;27(6):755-68.

Zolla V, Nizamutdinova IT, Scharf B, Clement CC, Maejima D, Akl T, Nagai T, Luciani P, Leroux JC, Halin C, Stukes S, Tiwari S, Casadevall A, Jacobs WR, Entemberg D, Zawieja D, Condeelia J, **Fooksman DR,** Gashev AA, Santambrogio L. Aging-related anatomical and biochemical changes in lymphatic collectors impair lymph drainage, fluid homeostasis, and pathogen transport. Aging Cell. 2015 Aug;14(4):582-94

Fooksman DR*. Organizing MHC class II Presentation. Front. Immunol., 10 April 2014;

Fooksman DR*, Nussenzweig MC, Dustin ML. Myeloid Cells Limit Production of Antibody-Secreting Cells after Immunization in the Lymph Node. J Immunol. 2014 Feb 1;192(3):1004-12

Schwickert TA, Victora GD, **Fooksman DR**, Kamphorst AO, Mugnier MR, Gitlin AD, Dustin ML, Nussenzweig MC. A dynamic T cell-limited checkpoint regulates affinity-dependent B cell entry into the germinal center. J Exp Med. 2011 Jun 6;208(6):1243-52. Epub 2011 May 16.

Victora GD, Schwickert TA, **Fooksman DR**, Kamphorst AO, Meyer-Hermann M, Dustin ML, Nussenzweig MC. Germinal Center Dynamics Revealed by Multiphoton Microscopy with a Photoactivatable Fluorescent Reporter. Cell Nov 2010; 143(4): 592-605.

Fooksman DR, Schwickert TA, Victora GD, Dustin ML, Nussenzweig MC, Skokos D. Development and migration of plasma cells in the mouse lymph node. Immunity. 2010 Jul 23;33(1):118-27.

Fooksman DR, Vardhana S, Vasiliver-Shamis G, Liese J, Blair DA, Waite J, Sacristán C, Victora GD, Zanin-Zhorov A, Dustin ML. Functional anatomy of T cell activation and synapse formation. Annu Rev Immunol. 2010 Mar;28:79-105. Review.

Fooksman DR*, Shaikh SR, Boyle S, Edidin M. Cutting edge: phosphatidylinositol 4,5-bisphosphate concentration at the APC side of the immunological synapse is required for effector T cell function. J Immunol. 2009 May 1;182(9):5179-82. PubMed PMID: 19380760; PubMed Central PMCID: PMC2799928.

Fooksman DR*, Edidin M, Barisas BG. Measuring rotational diffusion of MHC class I on live cells by polarized FPR. Biophys Chem. 2007 Oct;130(1-2):10-6.

Fooksman DR, Grönvall GK, Tang Q, Edidin M. Clustering class I MHC modulates sensitivity of T cell recognition. J Immunol. 2006 Jun 1;176(11):6673-80.

DR. NIKOLAOS FRANGOGIANNIS

Our laboratory studies the cell biological processes and the molecular pathways involved in cardiac repair, remodeling and fibrosis. The adult mammalian heart has negligible regenerative capacity and heals through formation of a collagen-based scar. Repair of the infarcted heart is dependent on induction and timely suppression of inflammatory signals, and on recruitment of reparative cells (fibroblasts and vascular cells). Dysregulation of the inflammatory and fibrotic responses causes adverse remodeling of the heart and results in heart failure. Using cell-specific genetic manipulations, established mouse models of cardiac injury and remodeling, and cell biological assays (using isolated cardiomyocytes, fibroblasts and macrophages), we explore the molecular circuitry of myocardial repair and fibrosis. Ongoing studies address the following questions:

1. What are the signals implicated in suppression and resolution of the post-infarction inflammatory reaction?

Timely inhibition and spatial containment of inflammatory signaling are critical for cardiac repair. We study the role of macrophage-specific inhibitory signals in suppression and resolution of the post-infarction inflammatory reaction.

2. Which the molecular signals are responsible for fibroblast activation and de-activation in infarcted and remodeling hearts?

In the infarcted heart, fibroblasts critically regulate cardiac repair by transdifferentiating into myofibroblasts and by producing extracellular matrix proteins. However, excessive or dysregulated activation of fibroblasts results in extension of fibrosis and causes diastolic ventricular dysfunction. We study the molecular signals that activate and de-activate fibroblasts in cardiac repair, focusing primarlily on the role and regulation of the TGF-beta cascade.

3. How does the extracellular matrix modulate the phenotype of cells involved in repair and fibrosis?

The extracellular matrix is not simply a structural scaffold, but actively participates in transduction of signaling responses. Specialized components of the matrix are induced following injury and modulate cytokine and growth factor-mediated responses, signaling through integrins or syndecan receptors. Our lab is particularly interested in the biology of these “matricellular proteins” in cardiac repair and remodeling.

4. How does metabolic disease cause cardiac fibrosis?

Diabetes and obesity are associated with profound alterations in cardiac function causing diastolic heart failure. Our lab studies the effects of metabolic dysregulation on cardiac fibroblasts and explores the mechanisms of fibrosis and capillary rarefaction in diabetic hearts.

5. What is the fate and role of pericytes in the infarcted and remodeling heart?

Pericytes are abundant in the mammalian heart and may regulate angiogenic and fibrogenic responses. Our lab will study the fate and role of pericytes in myocardial infarction and in diabeytic cardiomyopathy using lineage tracing strategies and cell-specific loss-of-function approaches.

The ultimate goal of our research is to identify therapeutic targets for attenuation of adverse remodeling following cardiac injury, thus preventing the development of heart failure.

PUBLICATIONS

On the role of inflammatory signaling in cardiac repair and remodeling:

M Bujak, G Ren, HJ Kweon, M Dobaczewski, A Reddy, G Taffet, XF Wang, **and NG Frangogiannis**. Essential Role of Smad3 in Infarct Healing and in the Pathogenesis of Cardiac Remodeling. ***Circulation*** 2007; 116:2127-38.

P Huebener, T Abou-Khamis, P Zymek, M Bujak, Ying X, K Chatila, S Haudek, G Thakker, **and NG Frangogiannis**, CD44 is critically involved in infarct healing by regulating the inflammatory and fibrotic response. ***J Immunol*** 2008; 180:2625-33.

M Bujak, M Dobaczewski, K Chatila, L Mendoza, N Li, A Reddy, **and NG Frangogiannis**. Interleukin-1 type (IL-1RI) signaling critically regulates infarct healing and cardiac remodeling. ***Am J Pathol*** 2008; 173:57-67.

M Dobaczewski, Y Xia, M Bujak, C Gonzalez-Quesada, **and NG Frangogiannis**. CCR5 signaling suppresses inflammation and reduces adverse remodeling following myocardial infarction mediating recruitment of regulatory T cells. ***Am J Pathol*** 2010; 176: 2177-87.

W Chen, A Saxena, N Li, J Sun, DW Lee, Q Tian, M Dobaczewski **and NG Frangogiannis**. Endogenous IRAK-M attenuates post-infarction remodeling through effects on macrophages and fibroblasts. ***Arterioscl Thromb Vasc Biol*** 2012; 32:2598-608.

A Saxena, W Chen, Y Su, V Rai, OU Uche, N Li, **and NG Frangogiannis**. IL-1 induces proinflammatory leukocyte infiltration and regulates fibroblast phenotype in the infarcted myocardium. ***J Immunol*** 2013; 191(9):4838-48.

NG Frangogiannis. The inflammatory response in myocardial injury repair and remodeling. ***Nat Rev Cardiol*** 2014; 11:255-265.

A Saxena, M Dobaczewski, V Rai, Z Haque, W Chen, N Li, **and NG Frangogiannis**. Regulatory T cells are recruited in the infarcted myocardium and may regulate fibroblast phenotype and function. ***Am J Physiol Heart Circ Physiol*** 2014; 307(8):H1233-42.

AV Shinde **and NG Frangogiannis**. Mediators secreted by myeloid cells may protect and repair the infarcted myocardium ***Circ Res*** 2015; 117:10-12.

NG Frangogiannis. Emerging roles of macrophages in cardiac injury: cardioprotection, repair and regeneration. ***J Clin Invest*** 2015: 2927-30.

On the biology of the fibroblast:

M Bujak, M Dobaczewski, C Gonzalez-Quesada, Y Xia, T Leucker, P Zymek, V Veeranna, A Tager, A Luster, **and NG Frangogiannis**. Induction of the CXC Chemokine Interferon-γ-Inducible Protein (IP)-10 Regulates the Reparative Response Following Myocardial Infarction. ***Circulation Research*** 2009; 105:973-83.

M Dobaczewski, M Bujak, N Li, C Gonzalez-Quesada; LH Mendoza, XF Wang, **and NG Frangogiannis**. Smad3 signaling critically regulates fibroblast phenotype and function in healing myocardial infarction. ***Circulation Research*** 2010; 107 (3): 418-28.

A Biernacka, M Dobaczewski **and NG Frangogiannis**. TGF-beta signaling in fibrosis. ***Growth Factors*** 2011; 29: 196-202.

W Chen **and NG Frangogiannis**. Fibroblasts in post-infarction inflammation and cardiac repair. ***BBA Mol Basis Dis***. 2013; 1833:945-53.

P Kong, P Christia, A Saxena, Y Su **and NG Frangogiannis**. Lack of specificity of Fibroblast Specific Protein (FSP)1 in cardiac remodeling and fibrosis. ***Am J Physiol Heart Circ Physiol*** 2013;305(9):H1363-72.

AV Shinde **and NG Frangogiannis**. Fibroblasts in myocardial infarction. A role in inflammation and repair. ***J Mol Cell Cardiol*** 2014; 70:74-82.

P Kong, P Christia **and NG Frangogiannis**. The pathogenesis of cardiac fibrosis. ***Cell Mol Life Sci*** 2014 71:549-74.

A Saxena, M Bujak, O Frunza, M Dobaczewski, C Gonzalez-Quesada, B Lu, C Gerard **and NG Frangogiannis**. CXCR3-independent actions of the CXC chemokine CXCL10 in the infarcted myocardium and in isolated cardiac fibroblasts are mediated through proteoglycans. ***Cardiovasc Res*** 2014; 103: 217-227.

A Biernacka, M Cavalera, J Wang, I Russo, A Shinde, P Kong, C Gonzalez-Quesada, V Rai, M Dobaczewski, DW Lee, XF Wang **and NG Frangogiannis**. Smad3 signaling promotes fibrosis, while preserving cardiac and aortic geometry in obese diabetic mice. ***Circulation Heart Failure*** 2015; 8: 788-798.

On the biology of the matricellular proteins:

Y Xia, M Dobaczewski, C Gonzalez-Quesada, W Chen, A Biernacka, N Li, D Lee **and NG Frangogiannis**. Endogenous Thrombospondin-1 protects the pressure-overloaded myocardium by modulating fibroblast phenotype and matrix metabolism. ***Hypertension*** 2011; 58: 902-911.

NG Frangogiannis. Matricellular proteins in cardiac adaptation and disease. ***Physiol Rev*** 2012; 92:635-688.

P Kong, C Gonzalez-Quesada, N Li, M Cavalera, D-W Lee **and NG Frangogiannis**. Thrombospondin-1 regulates adiposity and metabolic dysfunction in diet-induced obesity enhancing adipose inflammation and stimulating adipocyte proliferation. ***Am J Physiol Endocrinol Metab*** 2013; 305:E439-450.

C Gonzalez-Quesada, M Cavalera, A Biernacka, P Kong, DW Lee, A Saxena, O Frunza, M Dobaczewski, AV Shinde, **and NG Frangogiannis**. Thrombospondin-1 induction in the diabetic myocardium stabilizes the cardiac matrix while promoting vascular rarefaction through angiopoietin-2 upregulation. ***Circulation Research*** 2013; 113(12):1331-44.

M Cavalera, J Wang **and NG Frangogiannis**. Obesity, metabolic dysfunction and cardiac fibrosis: pathophysiologic pathways, molecular mechanisms and therapeutic opportunities. ***Transl Res*** 2014. Pp: S1931-5244(14)00164-9.

DR. DAVID GOLDMAN

The medical community has long recognized fungi as important allergens for patients with asthma. Interestingly, fungal sensitization is more common in children and has been linked to severe asthma resulting in death. The accepted paradigm is that fungal sensitization occurs as a result of recurrent, transient environmental exposures. Yet, increasing evidence suggests that fungi may interact with people in unrecognized ways to promote asthma. My lab is interested in understanding the role of subclinical fungal infections in asthma and their potential contribution to the high prevalence of asthma in urban areas.

We have demonstrated that the majority of Bronx children older than 2 years have serologic evidence of cryptococcal infection. *Cryptococcus neoformans* is an encapsulated fungus that is well suited to serve as co-factor in urban asthma. *C. neoformans* colonizes pigeon droppings and is endemic to urban areas. Once inhaled, this fungus causes persistent, subclinical infections. Cryptococcal infection induces TH2 inflammation in animal models. In a rat model, we have shown that cryptococcal pulmonary infection acts a co-factor to enhance allergic inflammation to allergen challenge and promotes airway hyper-responsiveness, both hallmark features of asthma. Pulmonary cryptococcosis also induces chitinase expression, which has recently been implicated as an essential mediator of allergic inflammation.

In addition to fungal studies, my lab is interested in anthrax pathogenesis. *Bacillus anthracis* is widely recognized as a potential agent of bioterrorism as evidenced by the 2001 anthrax attack. The toxins of *B. anthracis* are essential to virulence. In collaborations with Drs. Arturo Casadevall and Jurgen Brojatsch, we have studied the mechanisms by which *Bacillus anthracis* toxins contribute to host death. We have identified a previously unrecognized protease in human serum that inactivates the protective antigen component of lethal toxin *in vitro*. The precise protease and its role in the host response and susceptibility to anthrax remain to be determined. We have also identified a potential role for platelet activating factor (PAF) in mediating the lethal effects of toxin, including the alterations in vascular permeability which is characteristic of anthrax. Together, these observations may have important implications in developing new approaches to the treatment of anthrax.

PUBLICATIONS

Vicencio AG, Muzumdar H, Tsirilakis K, Kessel A, Nandalike K, **Goldman DL**. Severe asthma with fungal sensitization in a child: response to itraconazole therapy. Pediatrics. 2010 May;125(5):e1255-8. Epub 2010 Apr 12.

Goldman DL, Huffnagle GB. Potential contribution of fungal infection and colonization to the development of allergy. Med Mycol. 2009;47(5):445-56. Review.

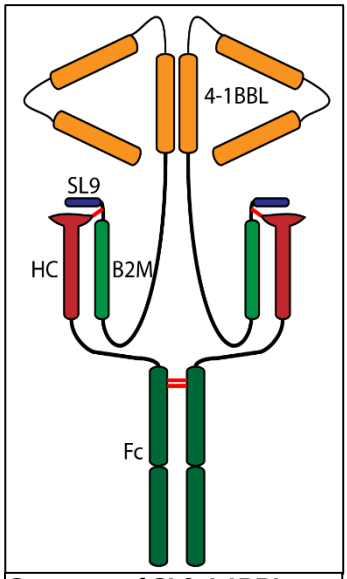
Vicencio AG, Narain S, Du Z, Zeng WY, Ritch J, Casadevall A, **Goldman DL**. Pulmonary cryptococcosis induces chitinase in the rat. Respir Res. 2008 May 15;9:40.

Goldman DL, Zeng W, Rivera J, Nakouzzi A, Casadevall A. Human serum contains a protease that protects against cytotoxic activity of Bacillus anthracis lethal toxin in vitro. Clin Vaccine Immunol. 2008 Jun;15(6):970-3. Epub 2008 Apr 30.

Goldman DL, Davis J, Bommarito F, Shao X, Casadevall A. Enhanced allergic inflammation and airway responsiveness in rats with chronic Cryptococcus neoformans infection: potential role for fungal pulmonary infection in the pathogenesis of asthma. J Infect Dis. 2006 Apr 15;193(8):1178-86. Epub 2006 Mar 9.

DR. HARRIS GOLDSTEIN

Our laboratory is utilizing novel molecular, cellular and biochemical approaches to "hack" the human immune system and amplify its activity to enable it to recognize and eliminate reactivated latent HIV-infected T cells (LHITC) and thereby achieve the functional cure of HIV-1 infection. Our lab is part of the NIH-funded *Martin Delaney Collaboratory: Towards an HIV-1 Cure* program established to fulfill President Barack Obama's pledge to invest in HIV cure research.



Structure of SL9:4-1BBL synTac with dimeric 4-1BBL.

The goal of the Delaney program is to unite and synergize the research programs of highly talented investigators to accelerate the development of a safe and scalable cure for HIV. One novel approach we are pioneering is the application of new immune checkpoint modulating biologics, which stimulate the immune system to kill tumor cells and have revolutionized cancer treatment, to amplify the cytotoxic activity of HIV-specific CD8+ T cells and increase their capacity to eliminate HIV-infected cells. For this purpose, we are applying synTac (artificial immunological synapse for T-cell activation), a novel class of soluble precision-targeted immunomodulatory biologics developed by the Almo lab. The synTac fusion proteins use a single MHC α and β2 microglobin chain containing a defined peptide (sc-pMHC) linked to a costimulatory or cytokine domain to integrate the specificity of antigen-receptor and potency of costimulatory and/or cytokine signaling (*see Figure*). Antigen specific T cell subpopulations are specifically activated by sc-pMHC binding to its cognate TCR which provides the primary activation signal as well as precisely delivering the costimulatory and/or cytokine signal by the linked costimulatory or cytokine domains. We have validated this approach for HIV by constructing synTac fusion proteins which target the HLA-A*0201-restricted HIV Gag epitope, SL9, linked to the costimulatory 4-1BBL molecule, SL9:4-1BBL-synTac. The SL9:4-1BBL-synTac specifically bound an SL9-specific CTL clone TCR and stimulated INF-γ and TNF-α secretion and cellular proliferation. We are extending those very promising results by developing synTac-based therapeutics linked to different costimulatory ligands and/or cytokines to identify the optimal costimulatory and cytokine signals required for the specific *in vitro* and *in vivo* activation and expansion of HIV-specific CD8+ T cells with the potential to eliminate reactivated LHITC.

Another novel strategy to cure HIV we are investigating and optimizing is amplifying NK cell cytotoxic function by treatment with various structurally enhanced cytokine constructs combined with targeting them to specifically kill reactivated latent HIV-1 infected cells by parallel treatment with molecularly engineered HIV-specific broadly neutralizing antibodies, bispecific antibodies and/or fusion proteins. We are also molecularly engineering the antigen specificity of CD8+ T cells to recognize HIV-specific epitopes and generate potent HIV-specific CD8+ cytotoxic T lymphocytes (CTLs) by transducing them with lentiviruses expressing the genes encoding the alpha and beta chain of TCRs derived from potent HIV-specific CD8+ CTLs. We are further amplifying the cytotoxic capacity of these molecularly engineered HIV-specific CTLs by evaluating which added anti-apoptotic genes or cytotoxic genes added to the TCR-expressing lentiviral vectors improved the their capacity to eliminate reactivated latent infected cells.

Two major impediments prevent the testing of strategies to reduce HIV reservoirs and inhibit viral rebound after the cessation of antiretroviral therapy (ART): 1. The rarity of latent infected cells capable of producing infectious HIV-1; and 2. The difficulty in distinguishing latent infected cells from the majority of infected cells which contain integrated defective proviruses that

cannot produce infectious virus. To evaluate the efficacy of immune system “hacking” strategies to deplete the HIV reservoir, we have established a novel humanized mouse model consisting of highly immunodeficient NSG mice intrasplenically injected with CD4+ memory T cells isolated from HIV-infected patients who are virally suppressed by ART treatment (viral loads <50 copies/ml) which include a population of latent HIV-infected cells. We demonstrated that the transplanted HIV reservoir was activated *in vivo* as reported for ART-suppressed individuals during treatment interruption. These mice displayed plasma viremia within 1 week after injection which rapidly rose over the next month. We will be using this mouse model to examine the effects of the aforementioned immune amplification strategies to reduce the HIV reservoir as indicated by elimination of viremia, temporal delay in the onset of the viremia and/or reduction in the amplitude of the viremia. We have also developed another humanized mouse model infectible with an infectious HIV expressing a luciferase reporter that enables us to serially visualize HIV infection in live mice by the intensity of the luciferase signal (see Figure).

The mechanisms by which HIV infection and meth disrupt the blood-brain barrier (BBB), stimulate migration of HIV infected monocytes into the CNS and induce neuroinflammation and the impact of ART on these processes are not fully delineated. This is a highly significant area of research relevant to NIH high priority topics of HIV/AIDS research, understanding the basic biology of HIV pathogenesis causing immune dysfunction and chronic inflammation and addressing the impact of HIV-associated comorbidities including neurological complications. We are also using a novel transgenic mouse we developed, hu-CD4/R5/cT1 mice, which circumvents major entry and transcription blocks preventing murine HIV-1 infection by targeting transgenic expression of human CD4, CCR5 and cyclinT1 genes to $CD4^+$ T cells and myeloid-committed cells. These mice develop disseminated HIV-1 infection after intravenous HIV-1 injection and local HIV-1 infection after intravaginal inoculation. We are utilizing these mice to evaluate the in vivo efficacy of novel HIV-1 vaccines. In addition, we are using these transgenic mice to evaluate the mechanisms by which co-infection facilitates HIV-1 acquisition and to determine the efficacy of different preventive therapies. By crossing the hu-CD4/R5/cT1 mice with another novel transgenic mouse line we developed, which expresses a full-length HIV provirus that produces infectious HIV, we are investigating the effect of drugs of abuse on disrupting the BBB and facilitating the entry of HIV-infected inflammatory cells into the brain and evaluating the capacity of antibodies to adhesion molecules to prevent the transmigration of HIV-infected inflammatory cells into the brain

Recent Publications

Bardhi A, Wu Y, Chen W, Zhu Z, Zheng JH, Wong H, Jeng E, Jones J, Ochsenbauer C, Kappes JC, Dimitrov DS, Ying T, **Goldstein H**. Potent in vivo NK cell-mediated elimination of HIV-1-Infected cells mobilized by a gp120-bispecific and hexavalent broadly neutralizing fusion protein. ***J. Virology*** 2017: pii: JVI.00937-17. doi: 10.1128/JVI.00937-17. [Epub ahead of print].

Flerin NC, Chen H, Glover TD, Lamothe PA, Zheng JH, Fang JW, Ndhlovu ZM, Newell EW, Davis MM, Walker BD, **Goldstein H**. T-Cell Receptor (TCR) Clonotype-Specific Differences in Inhibitory Activity of HIV-1 Cytotoxic T-Cell Clones Is Not Mediated by TCR Alone. ***J. Virology*** 2017;91:e02412-16. doi: 10.1128/JVI.02412-16.

Thomas T, Seay K, Zheng JH, Zhang C, Ochsenbauer C, Kappes JC, **Goldstein H.** High-Throughput Humanized Mouse Models for Evaluation of HIV-1 Therapeutics and Pathogenesis. *Methods Mol Biol*. 2016;1354:221-35. doi: 10.1007/978-1-4939-3046-3_15

Seay K, Khajoueinejad N, Zheng JH, Kiser P, Ochsenbauer C, Kappes JC, Herold B, **Goldstein H.** The vaginal acquisition and dissemination of HIV-1 infection in a novel transgenic mouse model is facilitated by coinfection with HSV-2 and is inhibited by microbicide treatment. *J. Virology* 2015;89:9559-9570. doi: 10.1128/JVI.01326-15.

Seay K, Church C, Zheng JH, Deneroff K, Ochsenbauer C, Kappes JC, Liu B, Jeng EK, Wong HC, **Goldstein H**. *In vivo* activation of human NK Cells by treatment with an Interleukin-15 superagonist potently inhibits acute *in vivo* HIV-1 infection in humanized mice. *J. Virology* 2015;89:6264-6274. doi: 10.1128/JVI.00563-15.

Costantini LM, Irvin SC, Kennedy SC, Guo F, **Goldstein H**, Herold BC, Snapp EL. Engineering and exploitation of a fluorescent HIV-1 gp120 for live cell CD4 binding assays. *Virology.* 2015 476:240-248. doi: 10.1016/j.virol.2014.12.019.

Nixon B, Fakioglu E, Stefanidou M, Wang Y, Dutta M, **Goldstein H**, Herold BC. Genital herpes simplex virus type 2 infection in humanized HIV-transgenic mice triggers HIV shedding and is associated with greater neurological disease. *J Infect Dis*. 2014;209:510-22. doi: 10.1093/infdis/jit472.

Seay K, Qi X, Zheng JH, Zhang C, Chen K, Dutta M, Deneroff K, Ochsenbauer C, Kappes JC, Littman DR, **Goldstein H**. Mice transgenic for CD4-specific human CD4, CCR5 and cyclin T1 expression: a new model for investigating HIV-1 transmission and treatment efficacy. *PLoS ONE*. 2013;8(5):e63537. doi: 10.1371/journal.pone.0063537.

DR. CLAUDIA GRAVEKAMP

Our laboratory is focused on the development and testing of cancer immunotherapy and non-immune-based cancer therapies. Since most cancer deaths occur by metastases (primary tumors can often be removed by surgery, chemotherapy, or radiation), our therapies are focused on the treatment of metastases. We have developed various therapies using different novel approaches, in preclinical mouse tumor models with metastatic breast and pancreatic cancer. For instance, we use an attenuated bacterium *Listeria monocytogenes* as a platform for the delivery of anticancer agents to the tumor microenvironment and into tumor cells such as radioactivity, tumor-associated antigens, or small molecules like alphagalactosylceramide, or we kill tumor cells through cryoablation by freezing and thawing tumor cells, combined with various adjuvants targeting myeloid-derived suppressor cells (MDSC) such as stimulator of interferon genes (STING)-ligand cyclic di-guanylate (c-di-GMP, Curcumin, and AMD3100. Since MDSC play a major role in immune suppression in the tumor microenvironment, MDSC are an important target in cancer immunotherapies. We also focus on the age factor since most cancer patients are old and elderly react less efficient to vaccines than young adults. The MDSC are present in blood of patients and mice with cancer. This MDSC population is strongly increased in the tumor microenvironment particularly at older age, and contributes to the age-related T cell unresponsiveness.

Listeria-based cancer vaccines

Attenuated *Listeria monocytogenes* is a weakened facultative anaerobic bacterium (non-toxic and non-pathogenic) and has been used to deliver antigens into antigen-presenting cells. We developed various Listeria-based constructs expressing tumor-associated antigens including Mage-b, Survivin, p53 etc and tested these constructs in mice with metastatic breast and pancreatic cancer, and demonstrated a significant reduction in metastases and tumor growth. In addition, we have further improved the efficacy of the Listeria-Mage-b vaccine with help of MDSC-targeting adjuvants like c-di-GMP and Curcumin. However, in 2009 our lab discovered that Listeria infects and kills tumor cells by the generation of reactive oxygen species (ROS) through the activation of the NADPH-oxidase pathway, and left healthy tissues unharmed. Based on this tropism for the tumor microenvironment we started using Listeria as a platform for the selective delivery of anti-cancer agents to the tumor microenvironment. For more detail see Kim et al, Cancer Res 2009; Chandra et al, Cancer Immunology Research, 2014.

Mechanisms that contribute to the selective survival and multiplication of Listeria in the tumor microenvironment

We have analyzed potential mechanisms explaining why Listeria survived and multiplied in the TME and not in healthy tissues. We discovered that Listeria is protected from immune clearance in the TME through strong immune suppression, but is rapidly killed in healthy tissues that lack immune suppression. In addition, we found that MDSC play an important role in the selective delivery and survival of Listeria in the tumor microenvironment. MDSC are selectively attracted by the primary tumor through the production of attractants such as granulocyte macrophage-colony stimulating factor (GM-CSF), interleukin (IL)-6, A100. Listeria infects, survives and multiplies in MDSC of tumor-bearing mice, and is protected from immune clearance because of the immune suppressive character of MDSC. We have shown that Listeria, once at the tumor site, infects (and kills) tumor cells directly or spreads from MDSC into tumor cells through the cell-to-cell spread mechanism specific for Listeria. For more detail see Quispe-Tintaya et al, PNAS 2013**;** Chandra et al, BJC, 2013.

Radioactive Listeria for the treatment of pancreatic cancer

Ninety six percent of patients diagnosed with pancreatic cancer have only 6 months to live, despite aggressive treatments. This underlines the urgent need for new effective therapies. In collaboration with Dr. Ekaterina Dadachova (Department of Radiation, Einstein), we developed a radioactive Listeria for the treatment of pancreatic cancer, by coupling ^{188}Rhenium to anti-Listeria antibodies followed by incubation with Listeria bacteria. This resulted in the synergistic destruction of cancer cells through Listeria-induced ROS and through ionizing radiation of the ^{188}Re. The number of metastases was reduced by 50% in mice treated with Listeria alone, and by 90% in mice treated with Listeria-^{188}Re. This correlated with the accumulation of radioactivity in the metastases. This was the first time that a live attenuated bacterium was successfully used to deliver radioactivity selectively to the tumor microenvironment. The potential of the radioactive Listeria for the treatment of pancreatic cancer was discussed in several high profile journals like Science, Nature, as well as lay Journals like The Economist, Forbes Magazine and many others. Currently, we are testing Listeria with other radioisotopes. For more detail see Quispe-Tintaya et al, PNAS 2013. Recently, we developed Listeria-32P by incorporating 32P-phosphorus directly into the Listeria during culture. Most likely of the longer half-life of 32P (14 days) than of 188Rhenium (16.9 hrs), Listeria-32P was also highly effective against tumors and metastases in syngeneic and transgenic mouse models of pancreatic cancer. For more detail see Oncotarget 2017.

Listeria incorporated with alphagalactosylceramide

In collaboration with Dr. Steven Porcelli (Department Microbiology and Immunology, Einstein), we incorporated alphagalactosylceramide (αGC) into the Listeria bacteria simply during culture. This method was originally developed for mycobacteria. αGC is a marine sponge that activates natural killer T cells (NKT) cells, which in turn stimulates other immune cells like natural killer (NK) cells and T cells. We demonstrated that Listeria expressing tumor-associated antigen Mage-b incorporated with αGC created an immune-stimulating environment that attracted the NKT cells to the metastases, resulting in improved activation of CD8 T cells to Mage-b and a dramatic reduction in the number of metastases in a mouse model of metastatic breast cancer (4T1). For more detail see Singh et al, BJC 2014.

Cryoablation combined with adjuvants

Cryoablation involves killing of tumor cells through freezing and thawing, resulting in recruitment of tumor-specific T cells. Since MDSC strongly inhibits these T cells we have the cryoablation combined with MDSC-targeting adjuvants like STING ligand c-di-GMP, Curcumin, and AMD3100. c-di-GMP reduces the number of MDSC and converts a subpopulation of MDSC into an immune-stimulating phenotype producing IL-12 (stimulates T cells). Curcumin reduces IL-6 produced by MDSC and breast tumors. IL-6 strongly inhibits T cells in the tumor microenvironment. AMD3100 is a small molecule that prevents the interaction of CXCR4 on MDSC and stromal cell-derived factor (sdf-1) on tumor cells. Currently, these combination therapies are under investigation in mice with metastatic breast cancer in collaboration with the Anticancer Fund (Brussels, Belgium). Preliminary results are extremely promising. Cryoablation and Meriva (Curcumin derivate) significantly delayed the onset of metastases and eliminated completely the primary tumor, prolonged the survival rate compared to the control groups in correlation with improved CD8 T cell responses to multiple tumor-associated antigens. For more detail see Chandra et al, OnoImmunology 2015.

Feasibility of cancer vaccination at older age

Cancer is a disease of the elderly. When cancer becomes metastatic, it often needs aggressive second-line treatment, for which the options are limited. This is particularly challenging for frail, elderly cancer patients in which comorbidity plays an antagonistic role. Immunotherapy is the most promising and benign option for preventing or curing metastatic cancer in such patients. Unfortunately, cancer immunotherapy is less effective at old than at young age, due to T cell unresponsiveness, especially in the tumor microenvironment (TME). Various causes have been described for T cell unresponsiveness at old age, such as lack of naïve T cells at older age, deficiency in the upregulation of co-stimulatory molecules on aged dendritic cells (DCs), and most recently, the increase in the population of MDSC in the TME of old compared to young mice, among other age-related immune impairments. As mentioned above, Listeria has an intimate relationship with MDSC. We have shown that Listeria-based vaccination was equally effective in young and old mice with metastatic breast cancer by targeting MDSC. The Listeria killed the tumor cells directly through ROS, and Listeria-activated T cells killed the infected tumor cells presenting the Listeria antigens. For more detail see Chandra et al, BJC, 2013.

Most recent peer-reviewed publications relevant to the field of cancer vaccination and cancer therapies selected out of 54

Kim SH, Castro F, Paterson Y, **Gravekamp C.** High efficacy of a Listeria-based vaccine against metastatic breast cancer reveals a dual mode of action. Cancer Res. 2009; 69(14): 5860-5866. PMID 19584282

Castro F, Leal B, Denny A, Bahar R, Lampkin S, Reddick R, Lu S, and **Gravekamp C.** Vaccination with Mage-b DNA induces CD8 T cell responses at young but not at old age in mice with metastatic breast cancer. British Journal of Cancer, 2009; 101:1329-1337. PMID 19826426

Quispe-Tintaya W*, Chandra D*, Jahangir A, Harris M, Casadevall A, Dadachova E, and **Gravekamp C.** A non-toxic radioactive Listeria^{at} is a highly effective therapy against metastatic pancreatic cancer. PNAS, 2013; 110(21):8668-73. PMID: 23610422.*Both authors contributed equally to the manuscript.

Singh M, RamosI, Asafu-AdjeiD, Quispe-TintayaW, ChandraD, JahangirA, ZangX, AggarwalBB, and **Gravekamp. C.** Curcumin Improves the Therapeutic Efficacy of Listeria^{at}-Mage-b Vaccine in mice with Triple Negative Breast Cancer (TNBC) in correlation with improved T cell responses in blood. Cancer Medicine, 2013; 2(4): 571-582 PMID: 24156030

Chandra D*, Quispe-Tintaya W, Jahangir A*, Singh M, and **Gravekamp C.** Myeloid-derived suppressor cells have a central role in attenuated Listeria monocytogenes-based immunotherapy against metastatic breast cancer in young and old mice. Brit J Cancer 2013, 108(11): 2281-90. PMID: 23640395. *Both authors contributed equally to the manuscript.

Chandra D, Quispe-TintayaW, Jahangir A, Asafu-AdjeiD, Ramos I, Sintim, HO, Zhou J, Hayakawa, Y, Karaolis D, and **Gravekamp C.** STING ligand c-di-GMP improves cancer vaccination against metastatic breast cancer. Cancer Immunology Research, 2014; 2(9): 901-910. (DOI: 10.1158/2326-6066.CIR-13-0123). PMID: 24913717

Singh M, Quispe-Tintaya, W, Chandra D, Jahangir A, Venkataswamy MM, Ng TW, Sharma S, Carreno LJ, Porcelli SA, and **Gravekamp C.** Direct incorporation of the NKT cell activator a-galactosylceramide into a recombinant *Listeria monocytogenes* improves breast cancer vaccine

efficacy. British Journal of Cancer 2014; (doi: 10.1038/bjc.2014.486). PMID: 25314062

Valdor R, Mocholi E, Botbol Y, Guerrero-Ros I, Chandra D, Koga H, **Gravekamp C,** Cuervo AM, Macian F. Chaperone-mediated autophagy regulates T cell responses through targeted degradation of negative regulators of T cell activation. Nat Immunol. 2014 Nov;15(11):1046-54. doi: 10.1038/ni.3003. PMID: 25263126

Chandra D, Jahangir A, Cornelis F, Rombauts K, Meheus L, Jorcyk CL, and **Gravekamp C.** Cryoablation and Meriva has strong therapeutic effect on triple negative breast cancer. OncoImmunology. 2015. In Press. Doi: 10.1080/2162402X.2015.1049802.

Chandra D, Selvanesan CB, Yuan Z, Libutti SK, Koba W, Beck A, Casadevall A, Dadachova E, **Gravekamp C.** 32-Phosphorus selectively delivered by Listeria to pancreatic cancer demonstrates strong therapeutic effect. Oncotarget. 2017 Feb 6. doi: 10.18632/oncotarget.15117. PMID: 28186976

Jahangir A, Chandra D, Quispe-Tintaya W, Singh M, and **Gravekamp C.** Immunotherapy with Listeria reduces metastatic breast cancer in young and old mice through different mechanisms. OncoImmunology, 2017. http://www.tandfonline.com/eprint/dnkC4emkjuSeYUeJtZGz/full

Most recent invited publications relevant to the field of cancer vaccination at older age selected out of 16

Gravekamp C. 2009. Cancer Immunotherapy and aging: Lessons from the mouse. In: Handbook on Immunosenescence: basic understanding and clinical applications, Eds Fulop T, Pawelec G, Franceschi C, and Hirokawa K. Springer. Pp1217-1243.

Gravekamp C, Kim SH, and Castro F. Cancer vaccination: manipulation of immune responses at old age. Mechanism of Ageing and Development 2009:130(1-2): 67-75. PMID 18561984

Gravekamp C. The importance of the age factor in cancer vaccination at older age. Cancer Immunology and Immunotherapy 2009; 58(12): 1969-1977. PMID 19259666.

Pawelec G, Lustgarten J, Ruby C, and **Gravekamp C.** Impact of aging on cancer immunity and immunotherapy. Cancer Immunol Immunother 2009: 58: 1907-1908. PMID 19787858

Gravekamp C, and Paterson Y. Harnessing Listeria monocytogenes to target tumors. Cancer Biol and Ther 9: 1-9, 2010. PMID 20139702.

Gravekamp C. The impact of aging on cancer vaccination. COII 2011; 23: 555-560. PMID:21763118.

Gravekamp C. Cancer vaccination in older age. Interdiscip Top Gerontol. 2013;38:28-37. PMID: 23503513.

Chandra D and Gravekamp C. Myeloid-derived suppressor cells: cellular missiles to target tumors. Oncoimmunology 2013; 1;2(11):e26967. PMID: 24427545

Gravekamp C and Jahangir A. Is cancer vaccination feasible at older age? Exp Gerontol 2014 Jun;54C:138-144. PMID: 24509231.

Gravekamp C and Chandra C. Targeting STING pathways for the treatment of cancer. OncoImmunology, 2015. In press. doi: 10.4161/2162402X.2014.988463.

Kohanski RA, Deeks SG, **Gravekamp C,** Halter JB, High K, Hurria A, Fuldner R, Green P, Huebner R, Macchiarini F, Sierra F. Reverse geroscience: how does exposure to early diseases accelerate age-related decline in health? Ann N Y Acad Sci. 2016 Dec;1386(1):30-44. doi: 10.1111/nyas.13297. Epub 2016 Dec 1.
PMID: 27907230

RESEARCH IMPACT

Commentary on paper about Radioactive Listeria by Quipe-Tintaya et al. in PNAS 2013. Stritzker J, and Szalay AA. Single-agent combinatorial cancer therapy. www.pnas.org/cgi/doi/10.1073/pnas.1305832110.

Websites of Journals and magazines that discussed the impact of the radioactive Listeria for therapeutic treatment of pancreatic cancer:

http://www.nature.com/nrgastro/journal/vaop/ncurrent/full/nrgastro.2013.81.html

http://news.sciencemag.org/sciencenow/2013/04/radioactive-microbes-nuke-tumor-.html

http://txchnologist.com/post/48768770496/radioactive-metal-hitches-ride-on-bacteria-to-fight

http://www.economist.com/news/science-and-technology/21576628-surprising-new-way-kill-tumours-three-wrongs-make-right

https://www.sciencenews.org/article/microbes-can-redeem-themselves-fight-disease

DR. FELIPE DIAZ-GRIFFERO

key words: HIV-1 uncoating and reverse transcription, restriction factors TRIM5alpha TRIMCyp, transportin-3 (TNPO3), CPSF6, SAMHD1, MxB, SERINC5, elite controllers, and HIV-1 T cell restriction factors.

My research program is focused on understanding early events of HIV-1 infection such as uncoating, reverse transcription and nuclear import. To this end, we have exploited a group of proteins that are expressed by the host, and block HIV-1 infection at early stages. These proteins, known as restriction factors, have allowed us to understand fundamental processes in the HIV-1 life cycle. Besides assisting the understanding of fundamental problems on HIV-1 biology, restriction factors represent a new frontier in the search for an effective HIV-1 cure. The following sections explain our past findings, ongoing and planned research.

1) HIV-1 Uncoating

HIV-1 uncoating occurs early in infection, and is the shedding of monomeric capsid from the HIV-1 core, which is composed of 1500 monomers of capsid protein assembled into a conical structure containing the RNA viral genome. Our investigations revealed, contrary to an old dogma, that HIV-1 reverse transcription occurs before or during uncoating but not after (Roa et al., 2012)(Fig. 1). Furthermore, we demonstrated that genetic or pharmacological inhibition of reverse transcription inhibits the uncoating process during infection (Yang et al., 2012). These experiments suggested that internal rearrangements inside the core start the uncoating process. In agreement, we found that cytosolic extracts stabilized the HIV-1 core during infection in vivo and in vitro (Fricke et al., 2013a). Overall, our work suggests that HIV-1 cores are stable in the cytosol, and that initiation of uncoating is triggered from inside the core.

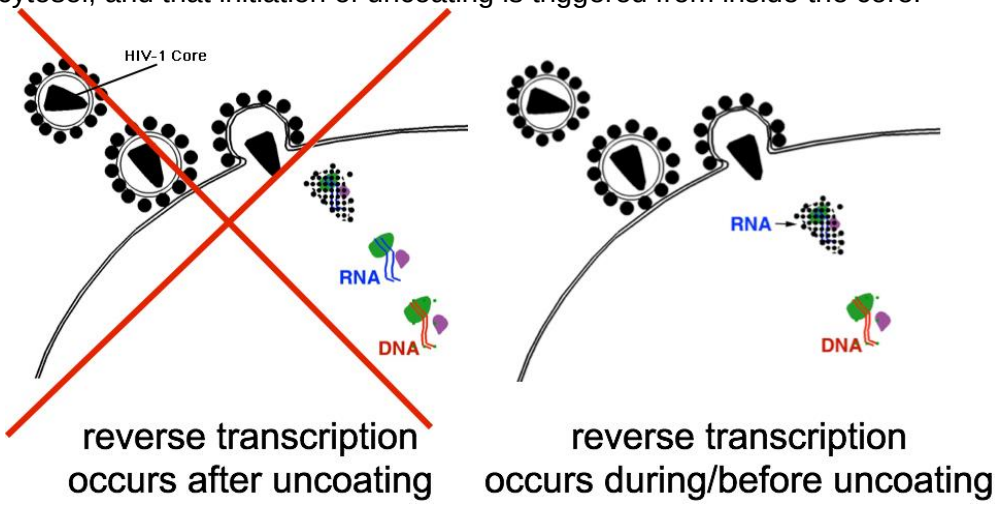


Figure 1. Current model for the occurrence of HIV-1 reverse transcription and uncoating. Our investigations showed that HIV-1 reverse transcription is completed inside the viral core during/before unocating. This is in stark contrast of a past dogma that suggested that reverse transcription occurs after uncoating.

The study of HIV-1 uncoating in vitro has been hindered by the unstable nature of the HIV-1 core outside the cellular environment. This evidence together with work mentioned above

suggests that the HIV-1 core is stabilized by cellular factors. Future work on this area will use biochemical and genetic approaches to identify factors that stabilize the HIV-1 core in the cellular environment. To this end, we will biochemically isolate HIV-1 cores from infected cells and identify the proteins associated to the core by mass spectrometry. We will compare the protein content of HIV-1 cores stabilized by different conditions: using reverse transcription inhibitors (Yang et al., 2012), viruses containing a defective reverse transcriptase enzyme (Yang et al., 2012), the microtubule disruptive drug nocodazol (Lukic et al., 2014; Malikov et al., 2015), cells expressing cytoplasmic CPSF6(Fricke et al., 2013b), and cells expressing MxB/Mx2(Fricke et al., 2014). As a negative control, we will mock isolate cores from cells expressing rhesus TRIM5a, which accelerates uncoating (Diaz-Griffero et al., 2007a; Perron et al., 2007; Stremlau et al., 2005). The assays we have developed to study capsid stability in vitro and in vivo will be used to confirm these interactions(Fricke et al., 2013a). Finally, the contribution to uncoating and infection will be evaluated in cells where the candidate proteins are knockout using the Cas9/CRISPR technology that is already working in our lab. Finding proteins that stabilize the HIV-1 core during infection will provide fundamental understanding on the uncoating process of HIV-1.

2) TRIM5α

The HIV-1 restriction factor TRIM5α is composed of four domains: RING, B-box-2, coiled-coil and PRYSPRY domains (Fig. 2A). To understand the contribution of these different domains to restriction, we have solved the structure of the RING, B-Box-2 and PRYSPRY domains (Biris et al., 2013; Diaz-Griffero et al., 2009; Lienlaf et al., 2011; Roa et al., 2012). Our structure-function studies revealed: 1) the RING domain provides E3 ligase activity, which is necessary for restriction(Lienlaf et al., 2011; Roa et al., 2012), 2) the B-box-2 domain regulates the ability of TRIM5a to form higher-order complexes(Diaz-Griffero et al., 2009; 2007b), which is an essential function for the ability of TRIM5α to form an array of protein in the surface of the core (Fig.2B)(Ganser-Pornillos et al., 2004). 3) the PRYSPRY domain is the domain that comes in direct contact with the HIV-1 core (Yang et al., 2014). In summary our findings suggested that the rhesus macaque protein TRIM5α binds to the surface of the HIV-1 core by forming an array protein (Fig. 2B). Formation of this complex recruits Ubc13, which is an E2 enzyme required for restriction(Pertel et al., 2011). Subsequently, an unknown activity leads to acceleration of uncoating (Diaz-Griffero et al., 2007a; Stremlau et al., 2006). Although, we have recently solved the structure of Ubc13 interacting with the RING domain (Yudina et al., 2015), the mechanism and energy source by which this complex leads to acceleration of uncoating is unknown (Fig.2B).

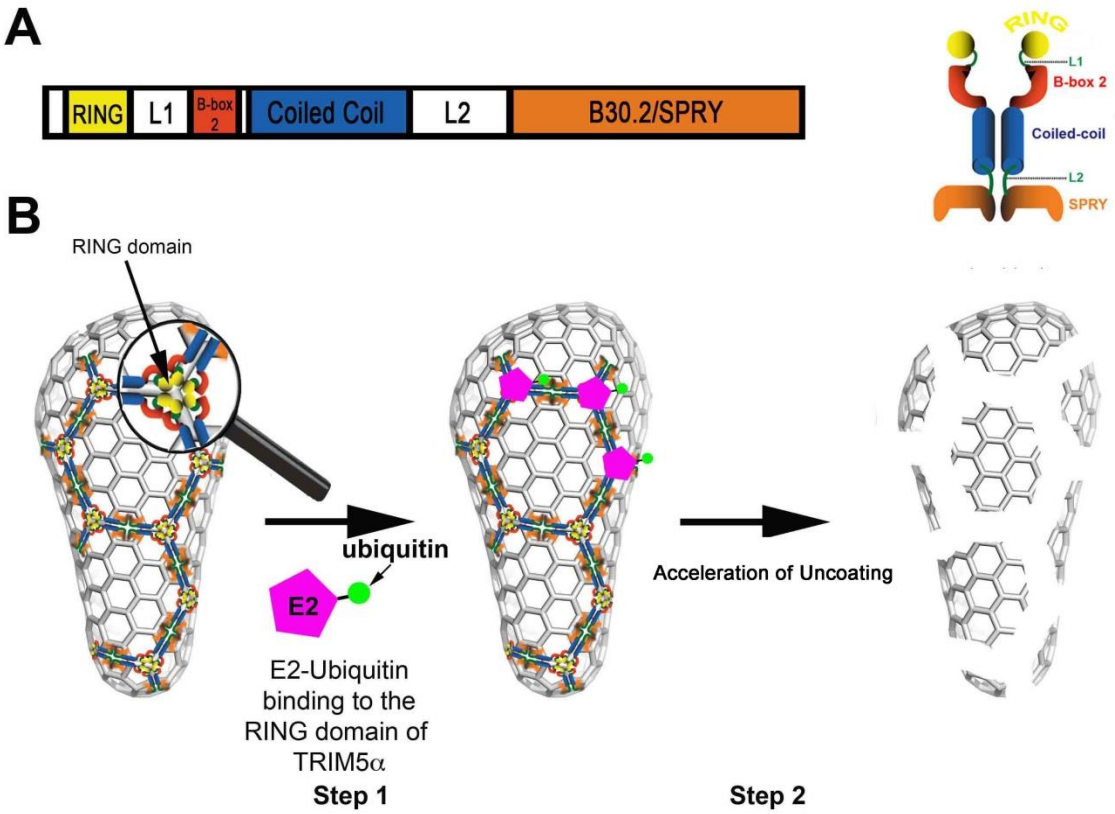


Figure 2. Inhibition of HIV-1 infection by TRIM5α. (A) The different domains of TRIM5α are shown, and a small cartoon depicting the TRIM5α protein is shown on the right side. **(B)** TRIM5a proteins assembled forming a hexagonal pattern on the surface of the HIV-1 core, and the RING domain of TRIM5α recruits an E2 enzyme (Ubc13). Subsequently the core is disassembled (acceleration of uncoating) and infection is aborted. The mechanism and source of energy for this process is unknown.

Interestingly, the structure of the PRYSPRY domain, which is the domain that directly interact with the HIV-1 core, exhibit a flexible loop in the region that interacts with the HIV-1 core (Fig. 3), as established by genetic experiments (Li et al., 2006; Yap et al., 2005). These observations suggested that movement of the loop is providing the energy necessary for the complex to accelerate uncoating. Future experiments will test the hypothesis that a flexible loop is required for acceleration of uncoating. To this end, we will identify TRIM5α mutations on the PRYSPRY domain that decrease the flexibility of the loop but preserve binding to the HIV-1 core. These particular variants will be tested for their ability to block HIV-1 and accelerate uncoating in human cells. These studies will be complemented by experiments that will measure the ability of the PRYSPRY domain mutants in solution to disassemble in vitro assembled HIV-1 CA complexes (Fricke et al., 2013a). Overall these experiments will sort out the role of loop flexibility in acceleration of uncoating.

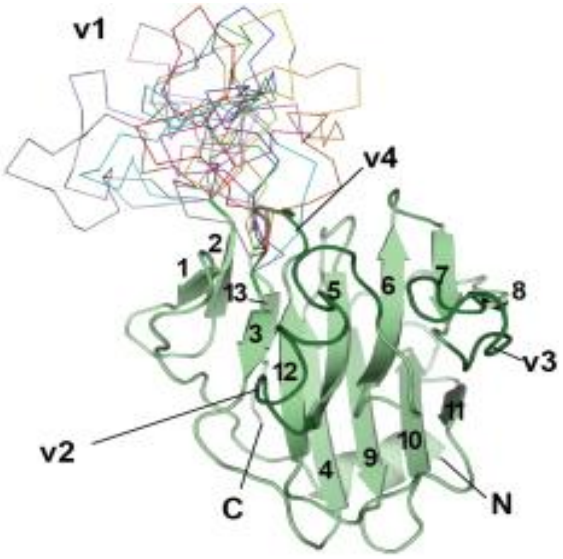


Figure 3. Structure of the PRYSPRY domain of rhesus monkey TRIM5α. The structure of the PRYSPRY domain is shown. The four variable loops of the protein are indicated as V1, V2, V3 and V4. The V1 loop, which directly interact with the HIV-1 core, exhibited hundred of different conformations, as indicated by the strands in different colors. These observations suggested that the PRYSPRY domain of TRIM5α exhibit great plasticity, which might allow the binding to different epitopes on the surface of the HIV-1 core. In addition, the movement of the V1 loop might be the energy necessary for accelerating the uncoating process of HIV-1.

3) MxB/Mx2

The restriction factor MxB is an interferon-a inducible protein that blocks HIV-1 infection in T cells (Goujon et al., 2013; Kane et al., 2013; Liu et al., 2013). Our investigations revealed that MxB blocks HIV-1 infection by inhibiting the uncoating process of HIV-1(Fricke et al., 2014)(Fig. 4). We found that MxB directly interacts with the HIV-1 core by using a triple arginine in the N-terminal domain of MxB (Schulte et al., 2015). Our studies also showed that oligomerization of MxB is essential for the ability of MxB to bind to the HIV-1 core and restrict HIV-1(Buffone et al., 2015). Overall, these results suggested that MxB binding to the core is forming an array on the surface of the HIV-1 core. Future experiments will test the hypothesis that MxB forms an array of protein on the surface of the HIV-1 core, which leads to inhibition of uncoating. To this end, we will perform Electron Microscopy of pure MxB protein overlaid on in vitro assembled HIV-1 CA that forms flat sheets. These experiments will show whether MxB cages the HIV-1 core in order to prevent HIV-1 uncoating. We are currently testing our purified MxB protein from human cells for its ability to interact with HIV-1 cores.

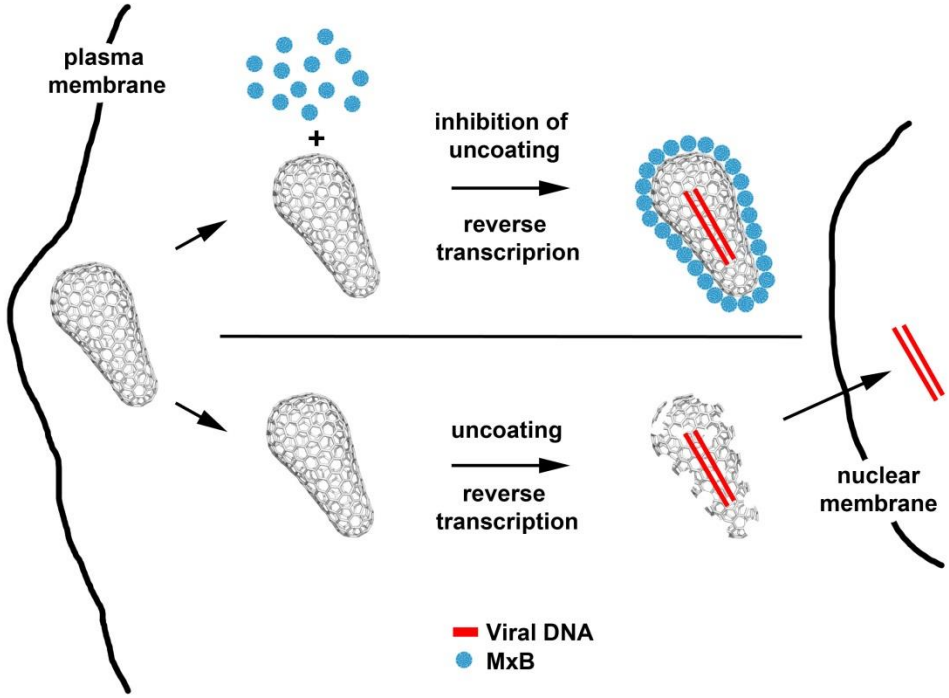


Figure 4. Inhibition of HIV-1 by MxB/Mx2. Our investigations revealed that MxB directly interacts with the HIV-1 core and prevents the uncoating process of HIV-1 terminating infection.

4) SAMHD1

The restriction factor SAMHD1 prevents HIV-1 infection of macrophages, dendritic cells, and resting T cells (Baldauf et al., 2012; Hrecka et al., 2011; Laguette et al., 2011). Our investigations revealed that SAMHD1 is regulated by phosphorylation of T592(White et al., 2013a; 2013b)(Fig. 5). The unphosphorylated form of SAMHD1 potently blocks HIV-1 infection. By contrast the phosphorylated form does not affect HIV-1 infection. These investigations suggested that the ability of SAMHD1 to block HIV-1 infection can be modulated. To this end, we are currently investigating in human primary cells the regulation of phosphorylation by different cytokines. We recently found that SAMHD1 is S-glutathionylated, and that this post-translational modification is essential for the ability of SAMHD1 to block HIV-1 infection. We are currently investigating the contribution of SAMHD1 S-glutathionylation to restriction. Although we have performed extensive biochemical and cellular characterization of SAMHD1(Brandariz-Nuñez et al., 2013; 2012; Ryoo et al., 2014; St Gelais et al., 2014; Welbourn et al., 2012; White et al., 2013b; 2014), we have not explored the role of SAMHD1 in immunity. Efficient lentiviral infection of macrophages in old world monkeys correlates with a strong adaptive immunity (Schaller et al., 2012).

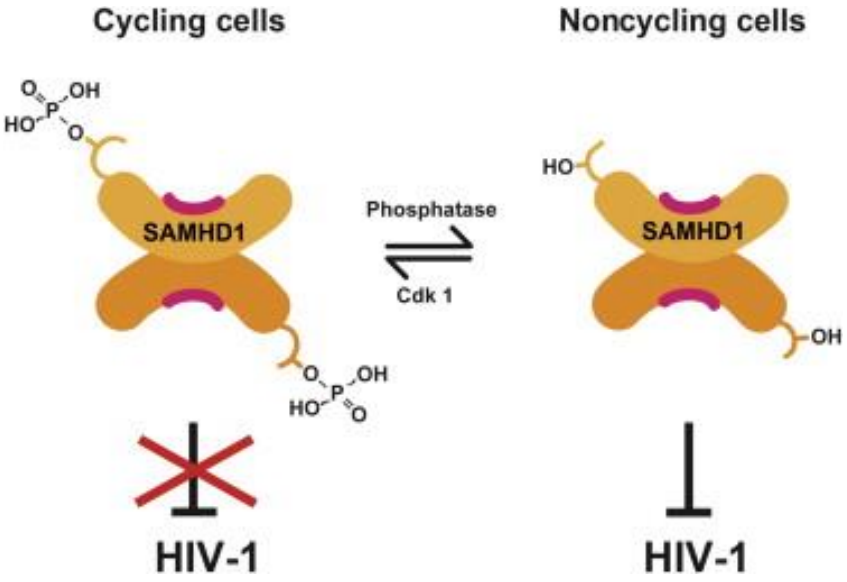


Figure 5. Regulation of SAMHD1 anti-HIV-1 activity by phosphorylation. Our investigations revealed that phosphorylation of SAMHD1 at T592 modulates the ability of this restriction factor to block HIV-1 infection of macrophages.

We are currently testing the hypothesis that SAMHD1 is involved in adaptive immunity. To this end, we will study adaptive immunity in the SAMHD1 knockout mice by testing antibody response, and the ability of the mice to prevent the growth of diverse pathogens. We will initially test pathogens that are known to be inhibited by SAMHD1, such as HSV-1/2(Kim et al., 2013) and mycobacterium tuberculosis(our unpublished preliminary studies). These investigations will help us understand the role of SAMHD1 in the immune system.

PUBLICATIONS

(h-index=27, total citation=2618). Total papers = 69

1. Yudina, Z., Roa, A., Johnson, R., Biris, N., Vieira, D., Tsiperson, V., Reszka, N., Taylor, A., Hart, J., Demeler, B., Diaz-Griffero, F*., Ivanov, D*. RING dimerization links Higher-order Assembly of TRIM5α to synthesis of K63-linked polyubiquitin. Cell Reports 12, 1-10, August 4, 2015.

2. Posch W, Steger M, Knackmuss U, Blatzer M, Baldauf HM, Doppler W, White TE, Hörtnagl P, Diaz-Griffero F, Lass-Flörl C, Hackl H, Moris A, Keppler OT, Wilflingseder D. Complement-Opsonized HIV-1 Overcomes Restriction in Dendritic Cells**. PLoS Pathog.** 2015 Jun 29;11(6):e1005005. doi: 10.1371/journal.ppat.1005005. eCollection 2015 Jun. PubMed PMID: 26121641; PubMed Central PMCID: PMC4485899.

3. Schulte B, Buffone C, Opp S, Di Nunzio F, Augusto De Souza Aranha Vieira D, Brandariz-Nuñez A, **Diaz-Griffero F**. Restriction of HIV-1 Requires the N-terminal Region of MxB/Mx2 as a Capsid-Binding Motif but not as a Nuclear Localization Signal. **J Virol.** 2015 Jun 10. pii: JVI.00753-15. [Epub ahead of print] PubMed PMID: 26063425.

4. Malikov V, da Silva ES, Jovasevic V, Bennett G, de Souza Aranha Vieira DA, Schulte B, **Diaz-Griffero F**, Walsh D, Naghavi MH. HIV-1 capsids bind and exploit the kinesin-1 adaptor FEZ1 for inward movement to the nucleus. **Nat Commun.** 2015 Mar 30;6:6660. doi: 10.1038/ncomms7660. PubMed PMID: 25818806; PubMed Central PMCID: PMC4380233.

5. Lelek M, Casartelli N, Pellin D, Rizzi E, Souque P, Severgnini M, Di Serio C, Fricke T, **Diaz-Griffero F**, Zimmer C, Charneau P, Di Nunzio F. Chromatin organization at the nuclear pore favours HIV replication. **Nat Commun.** 2015 Mar 6;6:6483. doi: 10.1038/ncomms7483. PubMed PMID: 25744187; PubMed Central PMCID: PMC4366494.

6. Buffone C, Schulte B, Opp S, **Diaz-Griffero F**. Contribution of MxB oligomerization to HIV-1 capsid binding and restriction. **J Virol.** 2015 Mar;89(6):3285-94. doi: 10.1128/JVI.03730-14. Epub 2015 Jan 7. PubMed PMID: 25568212; PubMed Central PMCID: PMC4337540.

7. Bhattacharya A, Alam SL, Fricke T, Zadrozny K, Sedzicki J, Taylor AB, Demeler B, Pornillos O, Ganser-Pornillos BK, **Diaz-Griffero F**, Ivanov DN, Yeager M. Structural basis of HIV-1 capsid recognition by PF74 and CPSF6. **Proc Natl Acad Sci U S A.** 2014 Dec 30;111(52):18625-30. doi: 10.1073/pnas.1419945112. Epub 2014 Dec 17. PubMed PMID: 25518861; PubMed Central PMCID: PMC4284599.

8. Fricke T, Buffone C, Opp S, Valle-Casuso J, **Diaz-Griffero F**. BI-2 destabilizes HIV-1 cores during infection and Prevents Binding of CPSF6 to the HIV-1 Capsid. **Retrovirology.** 2014 Dec 11;11:120. doi: 10.1186/s12977-014-0120-x. PubMed PMID: 25496772; PubMed Central PMCID: PMC4271331.

9. Lukic Z, Dharan A, Fricke T, **Diaz-Griffero F**, Campbell EM. HIV-1 uncoating is facilitated by dynein and kinesin 1. **J Virol.** 2014 Dec;88(23):13613-25. doi: 10.1128/JVI.02219-14. Epub 2014 Sep 17. PubMed PMID: 25231297; PubMed Central PMCID: PMC4248982.

10. Hollenbaugh JA, Tao S, Lenzi GM, Ryu S, Kim DH, **Diaz-Griffero F**, Schinazi RF, Kim B. dNTP pool modulation dynamics by SAMHD1 protein in monocyte-derived macrophages. **Retrovirology.** 2014 Aug 27;11:63. doi: 10.1186/s12977-014-0063-2. PubMed PMID: 25158827; PubMed Central PMCID: PMC4161909.

11. Fricke T, White TE, Schulte B, de Souza Aranha Vieira DA, Dharan A, Campbell EM, Brandariz-Nuñez A, **Diaz-Griffero F**. MxB binds to the HIV-1 core and prevents the uncoating process of HIV-1. **Retrovirology.** 2014 Aug 14;11:68. doi: 10.1186/PREACCEPT-6453674081373986. PubMed PMID: 25123063; PubMed Central PMCID: PMC4145229.

12. Ryoo J, Choi J, Oh C, Kim S, Seo M, Kim SY, Seo D, Kim J, White TE, Brandariz-Nuñez A, **Diaz-Griffero F**, Yun CH, Hollenbaugh JA, Kim B, Baek D, Ahn K. The ribonuclease activity of SAMHD1 is required for HIV-1 restriction. **Nat Med.** 2014 Aug;20(8):936-41. doi: 10.1038/nm.3626. Epub 2014 Jul 20. PubMed PMID: 25038827; PubMed Central PMCID: PMC4318684.

13. White TE, Brandariz-Nuñez A, Valle-Casuso JC, Knowlton C, Kim B, Sawyer SL, **Diaz-Griffero F**. Effects of human SAMHD1 polymorphisms on HIV-1 susceptibility. **Virology.** 2014 Jul;460-461:34-44. doi: 10.1016/j.virol.2014.04.023. Epub 2014 May 29. PubMed PMID: 25010268; PubMed Central PMCID: PMC4101048.

14. Brojatsch J, Lima H, Kar AK, Jacobson LS, Muehlbauer SM, Chandran K, **Diaz-Griffero F**. A proteolytic cascade controls lysosome rupture and necrotic cell death mediated by lysosome-destabilizing adjuvants. **PLoS One.** 2014 Jun 3;9(6):e95032. doi: 10.1371/journal.pone.0095032. eCollection 2014. PubMed PMID: 24893007; PubMed Central PMCID: PMC4043491.

15. Sastri J, Johnsen L, Smolin N, Imam S, Mukherjee S, Lukic Z, Brandariz-Nuñez A, Robia SL, **Diaz-Griffero F**, Wiethoff C, Campbell EM. Restriction of HIV-1 by rhesus TRIM5α is governed by alpha helices in the Linker2 region. **J Virol.** 2014 Aug;88(16):8911-23. doi: 10.1128/JVI.01134-14. Epub 2014 May 28. PubMed PMID: 24872590; PubMed Central PMCID: PMC4136267.

16. St Gelais C, de Silva S, Hach JC, White TE, **Diaz-Griffero F**, Yount JS, Wu L. Identification of cellular proteins interacting with the retroviral restriction factor SAMHD1. **J Virol.** 2014 May;88(10):5834-44. doi: 10.1128/JVI.00155-14. Epub 2014 Mar 12. Erratum in: J Virol. 2014 Jul;88(13):7689. PubMed PMID: 24623419; PubMed Central PMCID: PMC4019113.

17. Fricke T, White TE, Schulte B, de Souza Aranha Vieira DA, Dharan A, Campbell EM, Brandariz-Nuñez A, **Diaz-Griffero F**. MxB binds to the HIV-1 core and prevents the uncoating process of HIV-1. **Retrovirology.** 2014 Aug 14;11:68. doi: 10.1186/s12977-014-0068-x. PubMed PMID: 25927471.

18. Yang Y, Brandariz-Nuñez A, Fricke T, Ivanov DN, Sarnak Z, **Diaz-Griffero F**. Binding of the rhesus TRIM5α PRYSPRY domain to capsid is necessary but not sufficient for HIV-1 restriction. **Virology.** 2014 Jan 5;448:217-28. doi: 10.1016/j.virol.2013.10.012. Epub 2013 Oct 31. PubMed PMID: 24314652; PubMed Central PMCID: PMC3900861.

19. Brandariz-Nuñez A, Valle-Casuso JC, White TE, Nguyen L, Bhattacharya A, Wang Z, Demeler B, Amie S, Knowlton C, Kim B, Ivanov DN, **Diaz-Griffero F**. Contribution of oligomerization to the anti-HIV-1 properties of SAMHD1. **Retrovirology.** 2013 Nov 12;10:131. doi: 10.1186/1742-4690-10-131. PubMed PMID: 24219908; PubMed Central PMCID: PMC3882887.

20. Yang Y, Luban J, **Diaz-Griffero F**. The fate of HIV-1 capsid: a biochemical assay for HIV-1 uncoating. **Methods Mol Biol.** 2014;1087:29-36. doi: 10.1007/978-1-62703-670-2_3. PubMed PMID: 24158811; PubMed Central PMCID: PMC4317566.

21. Fassler M, Weissberg I, Levy N, **Diaz-Griffero F**, Monsonego A, Friedman A, Taube R. Preferential lentiviral targeting of astrocytes in the central nervous system. **PLoS One.** 2013 Oct 2;8(10):e76092. doi: 10.1371/journal.pone.0076092. eCollection 2013. PubMed PMID: 24098426; PubMed Central PMCID: PMC3788778.

22. Kim ET, White TE, Brandariz-Núñez A, **Diaz-Griffero F**, Weitzman MD. SAMHD1 restricts herpes simplex virus 1 in macrophages by limiting DNA replication. **J Virol.** 2013 Dec;87(23):12949-56. doi: 10.1128/JVI.02291-13. Epub 2013 Sep 25. PubMed PMID: 24067963; PubMed Central PMCID: PMC3838123.

23. Biris N, Tomashevski A, Bhattacharya A, **Diaz-Griffero F**, Ivanov DN. Rhesus monkey TRIM5α SPRY domain recognizes multiple epitopes that span several capsid monomers on the surface of the HIV-1 mature viral core. **J Mol Biol.** 2013 Dec 13;425(24):5032-44. doi:

10.1016/j.jmb.2013.07.025. Epub 2013 Jul 23. PubMed PMID: 23886867; PubMed Central PMCID: PMC4116666.

24. Fricke T, Brandariz-Nuñez A, Wang X, Smith AB 3rd, **Diaz-Griffero F**. Human cytosolic extracts stabilize the HIV-1 core. **J Virol.** 2013 Oct;87(19):10587-97. doi: 10.1128/JVI.01705-13. Epub 2013 Jul 24. PubMed PMID: 23885082; PubMed Central PMCID: PMC3807412.

25. Lima H Jr, Jacobson LS, Goldberg MF, Chandran K, **Diaz-Griffero F**, Lisanti MP, Brojatsch J. Role of lysosome rupture in controlling Nlrp3 signaling and necrotic cell death. **Cell Cycle.** 2013 Jun 15;12(12):1868-78. doi: 10.4161/cc.24903. Epub 2013 May 20. PubMed PMID: 23708522; PubMed Central PMCID: PMC3735701.

26. Fricke T, Valle-Casuso JC, White TE, Brandariz-Nuñez A, Bosche WJ, Reszka N, Gorelick R, **Diaz-Griffero F**. The ability of TNPO3-depleted cells to inhibit HIV-1 infection requires CPSF6. **Retrovirology.** 2013 Apr 26;10:46. doi: 10.1186/1742-4690-10-46. PubMed PMID: 23622145; PubMed Central PMCID: PMC3695788.

27. White TE, Brandariz-Nuñez A, Valle-Casuso JC, Amie S, Nguyen LA, Kim B, Tuzova M, **Diaz-Griffero F**. The retroviral restriction ability of SAMHD1, but not its deoxynucleotide triphosphohydrolase activity, is regulated by phosphorylation. **Cell Host Microbe.** 2013 Apr 17;13(4):441-51. doi: 10.1016/j.chom.2013.03.005. PubMed PMID: 23601106; PubMed Central PMCID: PMC3864637.

28. Di Nunzio F, Fricke T, Miccio A, Valle-Casuso JC, Perez P, Souque P, Rizzi E, Severgnini M, Mavilio F, Charneau P, **Diaz-Griffero F**. Nup153 and Nup98 bind the HIV-1 core and contribute to the early steps of HIV-1 replication. **Virology.** 2013 May 25;440(1):8-18. doi: 10.1016/j.virol.2013.02.008. Epub 2013 Mar 21. PubMed PMID: 23523133; PubMed Central PMCID: PMC3860269.

29. Pham QT, Veillette M, Brandariz-Nuñez A, Pawlica P, Thibert-Lefebvre C, Chandonnet N, **Diaz-Griffero F**, Berthoux L. A novel aminoacid determinant of HIV-1 restriction in the TRIM5α variable 1 region isolated in a random mutagenic screen. **Virus Res.** 2013 May;173(2):306-14. doi: 10.1016/j.virusres.2013.01.013. Epub 2013 Jan 25. PubMed PMID: 23357295; PubMed Central PMCID: PMC4317569.

30. Jacobson LS, Lima H Jr, Goldberg MF, Gocheva V, Tsiperson V, Sutterwala FS, Joyce JA, Gapp BV, Blomen VA, Chandran K, Brummelkamp TR, **Diaz-Griffero F**, Brojatsch J. Cathepsin-mediated necrosis controls the adaptive immune response by Th2 (T helper type 2)-associated adjuvants. **J Biol Chem.** 2013 Mar 15;288(11):7481-91. doi: 10.1074/jbc.M112.400655. Epub 2013 Jan 7. PubMed PMID: 23297415; PubMed Central PMCID: PMC3597789.

31. White TE, Brandariz-Nuñez A, Valle-Casuso JC, Amie S, Nguyen L, Kim B, Brojatsch J, **Diaz-Griffero F**. Contribution of SAM and HD domains to retroviral restriction mediated by human SAMHD1. **Virology.** 2013 Feb 5;436(1):81-90. doi: 10.1016/j.virol.2012.10.029. Epub 2012 Nov 13. PubMed PMID: 23158101; PubMed Central PMCID: PMC3767443.

32. Welbourn S, Miyagi E, White TE, **Diaz-Griffero F**, Strebel K. Identification and characterization of naturally occurring splice variants of SAMHD1. **Retrovirology.** 2012 Oct 23;9:86. doi: 10.1186/1742-4690-9-86. PubMed PMID: 23092512; PubMed Central PMCID: PMC3503569.

33. Brandariz-Nuñez A, Roa A, Valle-Casuso JC, Biris N, Ivanov D, Diaz-Griffero F. Contribution of SUMO-interacting motifs and SUMOylation to the antiretroviral properties of TRIM5α. **Virology.** 2013 Jan 20;435(2):463-71. doi: 10.1016/j.virol.2012.09.042. Epub 2012 Oct 16. PubMed PMID: 23084420; PubMed Central PMCID: PMC3534947.

34. Yang Y, Fricke T, **Diaz-Griffero F**. Inhibition of reverse transcriptase activity increases stability of the HIV-1 core. **J Virol.** 2013 Jan;87(1):683-7. doi: 10.1128/JVI.01228-12. Epub 2012 Oct 17. PubMed PMID: 23077298; PubMed Central PMCID: PMC3536417.

35. Di Nunzio F, Danckaert A, Fricke T, Perez P, Fernandez J, Perret E, Roux P, Shorte S, Charneau P, **Diaz-Griffero F**, Arhel NJ. Human nucleoporins promote HIV-1 docking at the nuclear pore, nuclear import and integration. **PLoS One.** 2012;7(9):e46037. doi: 10.1371/journal.pone.0046037. Epub 2012 Sep 25. Erratum in: PLoS One. 2013;8(12). doi:10.1371/annotation/34f46796-5d53-4a23-b3df-b599b0369eb9. PubMed PMID: 23049930; PubMed Central PMCID: PMC3457934.

36. **Diaz-Griffero F**. The Role of TNPO3 in HIV-1 Replication. **Mol Biol Int.** 2012;2012:868597. doi: 10.1155/2012/868597. Epub 2012 Jul 19. PubMed PMID: 22888429; PubMed Central PMCID: PMC3409535.

37. Biris N, Yang Y, Taylor AB, Tomashevski A, Guo M, Hart PJ, **Diaz-Griffero F**, Ivanov DN. Structure of the rhesus monkey TRIM5α PRYSPRY domain, the HIV capsid recognition module. **Proc Natl Acad Sci U S A.** 2012 Aug 14;109(33):13278-83. doi: 10.1073/pnas.1203536109. Epub 2012 Jul 30. PubMed PMID: 22847415; PubMed Central PMCID: PMC3421187.

38. Brandariz-Nuñez A, Valle-Casuso JC, White TE, Laguette N, Benkirane M, Brojatsch J, **Diaz-Griffero F**. Role of SAMHD1 nuclear localization in restriction of HIV-1 and SIVmac. **Retrovirology.** 2012 Jun 12;9:49. doi: 10.1186/1742-4690-9-49. PubMed PMID: 22691373; PubMed Central PMCID: PMC3410799.

39. Valle-Casuso JC, Di Nunzio F, Yang Y, Reszka N, Lienlaf M, Arhel N, Perez P, Brass AL, **Diaz-Griffero F**. TNPO3 is required for HIV-1 replication after nuclear import but prior to integration and binds the HIV-1 core. **J Virol.** 2012 May;86(10):5931-6. doi: 10.1128/JVI.00451-12. Epub 2012 Mar 7. PubMed PMID: 22398280; PubMed Central PMCID: PMC3347269.

40. Roa A, Hayashi F, Yang Y, Lienlaf M, Zhou J, Shi J, Watanabe S, Kigawa T, Yokoyama S, Aiken C, **Diaz-Griffero F**. RING domain mutations uncouple TRIM5α restriction of HIV-1 from inhibition of reverse transcription and acceleration of uncoating. **J Virol.** 2012 Feb;86(3):1717-27. doi: 10.1128/JVI.05811-11. Epub 2011 Nov 23. PubMed PMID: 22114335; PubMed Central PMCID: PMC3264337.

41. **Diaz-Griffero F**. Caging the beast: TRIM5α binding to the HIV-1 core. **Viruses.** 2011 May;3(5):423-8. doi: 10.3390/v3050423. Epub 2011 Apr 27. PubMed PMID: 21994740; PubMed Central PMCID: PMC3186010.

42. Lienlaf M, Hayashi F, Di Nunzio F, Tochio N, Kigawa T, Yokoyama S, **Diaz-Griffero F**. Contribution of E3-ubiquitin ligase activity to HIV-1 restriction by TRIM5alpha(rh): structure of the RING domain of TRIM5alpha. **J Virol.** 2011 Sep;85(17):8725-37. doi: 10.1128/JVI.00497-11. Epub 2011 Jul 6. PubMed PMID: 21734049; PubMed Central PMCID: PMC3165826.

43. **Diaz-Griffero F**, Gallo DE, Hope TJ, Sodroski J. Trafficking of some old world primate TRIM5α proteins through the nucleus. **Retrovirology.** 2011 May 15;8:38. doi: 10.1186/1742-4690-8-38. PubMed PMID: 21575157; PubMed Central PMCID: PMC3120760.

44. Sastri J, O'Connor C, Danielson CM, McRaven M, Perez P, **Diaz-Griffero F**, Campbell EM. Identification of residues within the L2 region of rhesus TRIM5alpha that are required for retroviral restriction and cytoplasmic body localization. Virology. 2010 Sep 15;405(1):259-66. doi: 10.1016/j.virol.2010.06.015. Epub 2010 Jul 14. PubMed PMID: 20633914; PubMed Central PMCID: PMC2914212.

45. Peña-Cruz V, McDonough SM, **Diaz-Griffero F**, Crum CP, Carrasco RD, Freeman GJ. PD-1 on immature and PD-1 ligands on migratory human Langerhans cells regulate antigen-presenting cell activity. **J Invest Dermatol.** 2010 Sep;130(9):2222-30. doi: 10.1038/jid.2010.127. Epub 2010 May 6. PubMed PMID: 20445553; PubMed Central PMCID: PMC2927196.

46. Gosselin A, Monteiro P, Chomont N, **Diaz-Griffero F**, Said EA, Fonseca S, Wacleche V, El-Far M, Boulassel MR, Routy JP, Sekaly RP, Ancuta P. Peripheral blood CCR4+CCR6+ and CXCR3+CCR6+CD4+ T cells are highly permissive to HIV-1 infection. **J Immunol.** 2010 Feb 1;184(3):1604-16. doi: 10.4049/jimmunol.0903058. Epub 2009 Dec 30. PubMed PMID: 20042588; PubMed Central PMCID: PMC4321756.

47. Mani M, Carrasco DE, Zhang Y, Takada K, Gatt ME, Dutta-Simmons J, Ikeda H, **Diaz-Griffero F**, Pena-Cruz V, Bertagnolli M, Myeroff LL, Markowitz SD, Anderson KC, Carrasco DR. BCL9 promotes tumor progression by conferring enhanced proliferative, metastatic, and angiogenic properties to cancer cells**. Cancer Res.** 2009 Oct 1;69(19):7577-86. doi: 10.1158/0008-5472.CAN-09-0773. Epub 2009 Sep 8. PubMed PMID: 19738061; PubMed Central PMCID: PMC4321734.

48. **Diaz-Griffero F**, Qin XR, Hayashi F, Kigawa T, Finzi A, Sarnak Z, Lienlaf M, Yokoyama S, Sodroski J. A B-box 2 surface patch important for TRIM5alpha self-association, capsid binding avidity, and retrovirus restriction. **J Virol.** 2009 Oct;83(20):10737-51. doi: 10.1128/JVI.01307-09. Epub 2009 Aug 5. PubMed PMID: 19656869; PubMed Central PMCID: PMC2753111.

49. Torimiro JN, Javanbakht H, **Diaz-Griffero F**, Kim J, Carr JK, Carrington M, Sawitzke J, Burke DS, Wolfe ND, Dean M, Sodroski J. A rare null allele potentially encoding a dominant-negative TRIM5alpha protein in Baka pygmies. **Virology.** 2009 Aug 15;391(1):140-7. doi: 10.1016/j.virol.2009.05.038. Epub 2009 Jul 3. PubMed PMID: 19577266; PubMed Central PMCID: PMC2760473.

50. Kar AK, **Diaz-Griffero F**, Li Y, Li X, Sodroski J. Biochemical and biophysical characterization of a chimeric TRIM21-TRIM5alpha protein. **J Virol.** 2008 Dec;82(23):11669-81. doi: 10.1128/JVI.01559-08. Epub 2008 Sep 17. PubMed PMID: 18799572; PubMed Central PMCID: PMC2583655.

51. Taube R, Zhu Q, Xu C, **Diaz-Griffero F**, Sui J, Kamau E, Dwyer M, Aird D, Marasco WA. Lentivirus display: stable expression of human antibodies on the surface of human cells and virus particles. **PLoS One.** 2008 Sep 11;3(9):e3181. doi: 10.1371/journal.pone.0003181. PubMed PMID: 18784843; PubMed Central PMCID: PMC2527531.

52. **Diaz-Griffero F**, Perron M, McGee-Estrada K, Hanna R, Maillard PV, Trono D, Sodroski J. A human TRIM5alpha B30.2/SPRY domain mutant gains the ability to restrict and prematurely uncoat B-tropic murine leukemia virus. **Virology.** 2008 Sep 1;378(2):233-42. doi: 10.1016/j.virol.2008.05.008. Epub 2008 Jun 30. PubMed PMID: 18586294; PubMed Central PMCID: PMC2597210.

53. **Diaz-Griffero F**, Taube R, Muehlbauer SM, Brojatsch J. Efficient production of HIV-1 viral-like particles in mouse cells. **Biochem Biophys Res Commun.** 2008 Apr 11;368(3):463-9. doi: 10.1016/j.bbrc.2007.12.195. Epub 2008 Jan 30. PubMed PMID: 18241668.

54 **Diaz-Griffero F**, Kar A, Lee M, Stremlau M, Poeschla E, Sodroski J. Comparative requirements for the restriction of retrovirus infection by TRIM5alpha and TRIMCyp. **Virology.** 2007 Dec 20;369(2):400-10. Epub 2007 Oct 24. PubMed PMID: 17920096; PubMed Central PMCID: PMC2153441.

55. **Diaz-Griffero F**, Kar A, Perron M, Xiang SH, Javanbakht H, Li X, Sodroski J. Modulation of retroviral restriction and proteasome inhibitor-resistant turnover by changes in the TRIM5alpha B-box 2 domain. **J Virol.** 2007 Oct;81(19):10362-78. Epub 2007 Jul 11. PubMed PMID: 17626085; PubMed Central PMCID: PMC2045480.

56. Javanbakht H, **Diaz-Griffero F**, Yuan W, Yeung DF, Li X, Song B, Sodroski J. The ability of multimerized cyclophilin A to restrict retrovirus infection. **Virology.** 2007 Oct 10;367(1):19-29. Epub 2007 Jun 15. PubMed PMID: 17574642; PubMed Central PMCID: PMC2065756.

57. Li X, Gold B, O'hUigin C, **Diaz-Griffero F**, Song B, Si Z, Li Y, Yuan W, Stremlau M, Mische C, Javanbakht H, Scally M, Winkler C, Dean M, Sodroski J. Unique features of TRIM5alpha among closely related human TRIM family members. **Virology.** 2007 Apr 10;360(2):419-33. Epub 2006 Dec 6. PubMed PMID: 17156811.

58. Díaz-Griffero F, Espinoza Cancino C, Medina Arévalo C, Arce-Johnson P. Expression of the crucifer-infecting TMV-Cg movement protein in tobacco plants complements in trans a TMV-U1 trafficking-deficient mutant. **Biol Res.** 2006;39(2):269-79. Epub 2006 Jul 25. PubMed PMID: 16874402.

59. Javanbakht H, Yuan W, Yeung DF, Song B, **Diaz-Griffero F**, Li Y, Li X, Stremlau M, Sodroski J. Characterization of TRIM5alpha trimerization and its contribution to human immunodeficiency virus capsid binding. **Virology.** 2006 Sep 15;353(1):234-46. Epub 2006 Jun 30. PubMed PMID: 16808955.

60. **Diaz-Griffero F**, Vandegraaff N, Li Y, McGee-Estrada K, Stremlau M, Welikala S, Si Z, Engelman A, Sodroski J. Requirements for capsid-binding and an effector function in TRIMCyp-mediated restriction of HIV-1. **Virology.** 2006 Aug 1;351(2):404-19. Epub 2006 May 2. PubMed PMID: 16650449.

61. Stremlau M, Perron M, Lee M, Li Y, Song B, Javanbakht H, **Diaz-Griffero F**, Anderson DJ, Sundquist WI, Sodroski J. Specific recognition and accelerated uncoating of retroviral capsids by

the TRIM5alpha restriction factor. **Proc Natl Acad Sci U S A.** 2006 Apr 4;103(14):5514-9. Epub 2006 Mar 15. PubMed PMID: 16540544; PubMed Central PMCID: PMC1459386.

62. **Diaz-Griffero F**, Li X, Javanbakht H, Song B, Welikala S, Stremlau M, Sodroski J. Rapid turnover and polyubiquitylation of the retroviral restriction factor TRIM5. **Virology.** 2006 Jun 5;349(2):300-15. Epub 2006 Feb 10. PubMed PMID: 16472833.

63. Mische CC, Javanbakht H, Song B, **Diaz-Griffero F**, Stremlau M, Strack B, Si Z, Sodroski J. Retroviral restriction factor TRIM5alpha is a trimer. **J Virol.** 2005 Nov;79(22):14446-50. PubMed PMID: 16254380; PubMed Central PMCID: PMC1280198.

64. Song B, **Diaz-Griffero F**, Park DH, Rogers T, Stremlau M, Sodroski J. TRIM5alpha association with cytoplasmic bodies is not required for antiretroviral activity. **Virology.** 2005 Dec 20;343(2):201-11. Epub 2005 Sep 22. PubMed PMID: 16183097.

65. **Diaz-Griffero F**, Jackson AP, Brojatsch J. Cellular uptake of avian leukosis virus subgroup B is mediated by clathrin. **Virology.** 2005 Jun 20;337(1):45-54. PubMed PMID: 15914219.

66. Javanbakht H, **Diaz-Griffero F**, Stremlau M, Si Z, Sodroski J. The contribution of RING and B-box 2 domains to retroviral restriction mediated by monkey TRIM5alpha. **J Biol Chem.** 2005 Jul 22;280(29):26933-40. Epub 2005 May 15. PubMed PMID: 15897199.

67. **Diaz-Griffero F**, Hoschander SA, Brojatsch J. Bystander killing during avian leukosis virus subgroup B infection requires TVB(S3) signaling. **J Virol.** 2003 Dec;77(23):12552-61. PubMed PMID: 14610178; PubMed Central PMCID: PMC262550.

68 **Diaz-Griffero F**, Hoschander SA, Brojatsch J. Endocytosis is a critical step in entry of subgroup B avian leukosis viruses. **J Virol.** 2002 Dec;76(24):12866-76. PubMed PMID: 12438612; PubMed Central PMCID: PMC136682.

69. Chi Y, **Diaz-Griffero F**, Wang C, Young JA, Brojatsch J. An NF-kappa B-dependent survival pathway protects against cell death induced by TVB receptors for avian leukosis viruses. **J Virol.** 2002 Jun;76(11):5581-7. PubMed PMID: 11991986; PubMed Central PMCID: PMC137008.

DR. BETSY HEROLD

Dr. Herold's lab focuses on identifying molecular mechanisms that contribute to the HIV-HSV syndemic and translating these findings into the clinic with the goal of developing novel therapeutic or prevention modalities. In collaboration with William Jacobs, PhD, the lab developed a paradigm shifting live attenuated HSV-2 vaccine candidate by deleting the immundominant envelope glycoprotein D, which has been the primary component of subunit vaccines. Subunit vaccines induce high titer neutralizing antibodies but failed to proect against HSV in clinical trials. In contrast, the gD deletion virus vaccine provided 100% protection against vaginal, skin, and neurological infections with a panel of clinical isolates of HSV-1 and HSV-2 in mice and guinea pigs, completely prevented the establishment of HSV latency, and elicited high titer antibodies that are rapidly transported into mucosal sites and facilitate antibody-dependent cell mediated cytotoxicity (ADCC) and antibody-dependent phagocytosis. Collaboratively, the Herold and Jacobs labs are now actively engaged in studies to define why deletion of gD skews the immune response towards non-neutralizing FcR activating antibody responses and conversely, why gD elicits neutralizing antibody responses. They are also actively engaged in advancing this vaccine into the clinic and using the deletion virus as a vector to engineer vaccines against other pathogens including HIV.

A second major goal of the lab is identifying the signaling pathways that HSV-2 usurps for entry and immune evasion: Genital herpes infections are a major global health problem and a substantial co-factor for HIV infection. Development of new approaches to prevent and ultimately eradicate HSV requires an understanding of the molecular and cellular events critical for the establishment of infection and latency and how the virus evades host immunity. To identify novel targets, the Herold lab has focused on cellular signaling pathways usurped by the virus to promote infection. Current work from the laboratory demonstrates that HSV activates Akt and calcium signaling pathways and these signaling pathways play critical roles in the establishment of infection and in cell-to-cell spread. Notably, HSV triggers the translocation of Akt from the inner to the outer plasma membrane where it interacts with the viral envelope glycoprotein B to activate calcium signaling and promote viral entry. In immune cells, viral induced Akt activation promotes apoptosis. We recently identified several drugs that block HSV-induced Akt signaling and prevent infection in several preclinical models.

The lab is also atively engaged in studies to understand the biology behind the HIV-HSV syndemic. Specifically the lab has found that defensins and secretory leukocyte protease inhibitor (SLPI) inhibit HSV and HIV infection in vitro and contribute to the endogenous antiviral activity of genital tract secretions. However, HSV-2 down-modulates the expression of these antimicrobial peptides, which may facilitate HIV and HSV infection and spread. In addition, HSV-2 induces the expression of proinflammatory cytokines and recruits immune cells to sites of HSV replication to promote HIV acquisition and transmission. HSV also has immunomodulatory effects on immune cells. HSV-2 interferes with dendritic cell (DC) function in vitro by inducing apoptosis, which may allow HSV to escape immune surveillance, but the cytokines and chemokines secreted by DCs in response to HSV promote HIV replication. Many of the in vitro and murine findings were recapitulated in clinical samples comparing genital tract mucosal immunity (genital tract secretions, cytobrushes, and vaginal or skin biopsies) in HIV-infected women who are HSV-2 seropositive or seronegative. Surprisingly, they also found that coinfection with HIV and HSV-2 is associated with significant changes in the phenotype and function of peripheral blood CD4 T cells that may contribute to the HIV reservoirs. Specifically they found that IL-32, which is decreased in HSV-2+ vs HSV-2 seronegative women blocks the effects of latency reversal stimuli being studied for HIV eradication and cure.

Finally the lab is also focused on the development of topical pre-exposure prophylaxis products (PrEP) for HIV and HSV prevention: In collaborative studies, their research team has developed an intravaginal ring that delivers tenofovir disoproxil fumarate (TDF), the more potent prodrug of tenofovir. The greater potency reflects significantly increased tissue and cellular uptake of TDF. The TDF ring prevented 100% of macaques from SHIV infection and retained activity when macaques were pretreated with high dose medroxyprogesterone, which provides a rigorous model of SHIV Infection by increasing susceptibility of NHP to Infection. Notably, a 0.3% TDF gel provided significantly greater protection than 1% tenofovir gel against HSV-2 in wild-type mice and against HIV and HSV-2 in transgenic mice (expressing human CD4, CCR5 and cyclin T1 to render them susceptible to HIV). The lab recently completed a Phase 1 clinical trial in the Bronx. The TDF ring was safe, and the tissue drug levels exceeded those associated with protection against HIV. A more expanded Phase 1 study in sexually active young adults in the US and Kenya is planned.

PUBLICATIONS

1. Nixon, B, Fakioglu, E, Stefanidou, M, Wang, Y, Dutta, M, Goldstein, H and **Herold, BC**. Genital herpes infection of humanized HIV transgenic mice triggers HIV shedding and is associated with greater neurological disease. J. Infec Dis, 2014 Feb 15;209(4):510-22. PMC3903370

2. Cheshenko, N, Trepanier, JB, Gonzalez, PA, Eugenin, EA, Jacobs WR and **Herold, BC**. Herpes simplex virus type 2 glycoprotein H interacts with integrin αvβ3 to facilitate viral entry and calcium signaling in human genital tract epithelial cells. J Virol 2014 June 18. PMC4136333

3. Smith, J.M., Srinivasan, P., Teller, R.S., Lo, Y, Dinh, C.T., Kiser, P.F.and **Herold, B.C**. Tenofovir disoproxil fumarate intravaginal ring protects high dose depot medroxprogesterone acetate treated macaques from multiple SHIV exposures. J Acquir Immune Defic Syndr. 2015 Jan 1;68(1):1-5. PMC4262537

4. Petro, C, Gonzalez, P, Cheshenko, N, Jandl, T, Khajoueinejad, N, Benard, A, Sangupta, M, **Herold, BC*** and Jacobs, WH* (co-senior authors). Herpes simplex type 2 virus deleted in glycoprotein D protects against vaginal, skin and neural disease. Elife 2015 Mar 10; 4. doi: 10.7554/eLife.06054. PMC4352706

5. Taneva E, Crooker K, Park SH, Su JT, Ott A, Cheshenko N, Szleifer I, Kiser PF, Frank B, Mesquita PM, **Herold BC**. Differential mechanisms of tenofovir and tenofovir disoproxil fumarate cellular transport and implications for topical pre-exposure prophylaxis. Antimicrob Agents Chemother. 2015 Dec 28;60(3):1667-75. PMC4775922

6. **Herold BC**, Chen BA, Salata RA, Marzinke MA, Kelly CW, Dezzutti CS, McGowan I, Galaska B, Levy L, Piper JM, Hillier S, Hendrix CW; MTN-011 Study Team. Impact of sex on the pharmacokinetics and pharmacodynamics of 1% tenofovir gel Clin Infect Dis. 2016 Feb 1;62(3):375-82. PMC4706638

7. Nakra, N., Madan, RP, Buckley, N, Huber, AM, Freiermuth, JL, Espinoza, L, Walsh, J, Parikh, UM, Penrose, KJ, Keller, MJ, and **Herold, BC**. Loss of innate host defense following unprotected vaginal sex. J Infec Dis 2016 Mar 1;213(5):840-7. PMC4747617

8. Boukhvalova, M., McKay, J., Mbaye, A., Sanford-Crane, H., Blanco, J., and **Herold, B.C**. Efficacy of HSV-2 glycoprotein D/ASO4 vaccine against genital HSV-2 and HSV-1 infection and disease in the cotton rat *Sigmodon hispidus* model. J Virol 2015 Oct;89(19):9825-40. doi: 10.1128/JVI.01387-15 PMC4577902

9. Keller, MJ, Mesquita P.M., Marzinke, M.A., Teller, R., Espinoza, L., Atrio, J.M. Lo, Y. Frank, B., Srinivasan, S., Fredricks, D.N., Rabe, L., Anderson, P.L., Hendrix, C.W., Kiser, P.F. and **Herold, B.C.** A phase 1 randomized placebo-controlled safety and pharmacokinetic trial of a tenofovir disoproxil fumarate vaginal ring. AIDS 2016 Mar 13;30(5):743-51. PMC4767579

10. Iversen MB, Reinert LS, Thomsen MK, Bagdonaite I, Nandakumar R, Cheshenko N, Prabakaran T, Vakhrushev SY, Krzyzowska M, Kratholm SK, Ruiz-Perez F, Petersen SV, Goriely S, Bibby BM, Eriksson K, Ruland J, Thomsen AR, **Herold BC**, Wandall HH, Frische S, Holm CK, Paludan SR. An innate antiviral pathway acting before interferons at epithelial surfaces. Nat Immunol 2016 Feb;17(2):150-8. PMC pending

Petro, C.D., Weinrick. B., Khajoueinejad, N., Burn, C., Seller, R., Jacobs, WR. and Herold, B.C. HSV-2 ΔgD Elicits FcγR-Effector Antibodies that Protect Against Clinical Isolates. The Journal of Clinical Investigation Insight. 2016, August.

WILLIAM R. JACOBS JR., Ph.D.

Strategies to Sterilize Infections of the Latent Pathogens *Mycobacterium tuberculosis* and Herpes Simplex Viruses

The Jacobs lab focuses on diseases that affect the developing world. Tuberculosis, caused by *M. tuberculosis*, causes 1.5 million deaths every year and over 9 million cases are reported each year. In fact, the World Health Organization (WHO) reports that 1 in 3 people on planet Earth have latent *M. tuberculosis* infections . Herpes viruses, caused by herpes simplex virus 1 (HSV-1) or herpes simplex virus 2 (HSV-2), cause significant latent infection around the globe, but can cause death or deformities in newborn children and severe infections or infections in the population. Although one pathogen is caused by a bacterium and the other by a virus, they share the common property that they can cause lifelong infections in humans. The Jacobs Lab use genetic approaches to elucidate the mechanisms by which these two pathogens cause latent infections. A major focus of the work is to develop novel vaccine candidates by engineering specific defects in the causative pathogenic agents. For *M. tuberculosis* the Jacobs Lab developed the first tools to move DNA into *M. tuberculosis.* The key to this success was using viruses that affect *M. tuberculosis*. By genetically engineering a mycobacterial virus, Dr. Jacobs made a shuttle phasmid, a chimeric vector that replicates in *E. coli* as a causent and in mycobacteria as a phage. These shuttle phasmids led to the development of the first transformation systems, efficient transposon delivery systems, and reporter mycobacteriophages that could readily asses drug susceptibilities in clinical isolates of *M. tuberculosis*. An effort to understand how M. tuberculosis evades killing by the immune system or bactericidal antibiotics is a major effort of the Jacobs group. For herpes viruses Dr. Jacobs has collaborated with Dr. Betsy Herold and engineered a deletion mutant of HSV-2 deleted for the gene encoding the major immunodomiant antigen and entry protein named gD. Surprisingly immunization of mice or guinea pigs with HSV-2 ΔgD provides sterilizing immunity against HSV-2 or HSV-1. Notably, protection from this deletion mutant is mediated by a special class of antibodies that mediate antibody dependent cell-mediated cytotoxicity (ADCC). Current efforts in the Jacobs lab are focused on elucidating the mechanism of the astonishing immunity and attempting to achieve a more efficacious TB vaccine.

See http://www.aecom.yu.edu/tbresearch/ for lab updates.

Recent Publications:

1. **Foreman, T., Mehra S., LoBato, D., Malek, A., Alvarez,X., Golden, N., Bucşan, A., Didier, P., Doyle, L., Russell-Lodrigue, K., Roya, C., Blanchard, JL., Kuroda, A., Lackner, A., Chan, J., Khader, S., Jacobs, Jr. WR., and Kaushal, D.**(2016) "CD4+ T cell-independent mechanisms suppress reactivation of latent tuberculosis in a macaque model of HIV co-infection" *PNAS*. (Accepted).
2. **Petro CD, Weinrick B, Khajoueinejad N, Burn C, Sellers R, Jacobs WR Jr, Herold BC.** (2016). "HSV-2 ΔgD elicits FcγR-effector antibodies that protect against clinical isolates." *JCI Insight* (12). pii: e88529. PMID: 27536733

3. **Halloum I, Carrère-Kremer S, Blaise M, Viljoen A, Bernut A, Le Moigne V, Vilchèze C, Guérardel Y, Lutfalla G, Herrmann JL, Jacobs WR Jr, Kremer L.** (2016). "Deletion of a dehydratase important for intracellular growth and cording renders rough Mycobacterium abscessus avirulent." *Proc Natl Acad Sci U S A.* 113(29):E4228-37. PMID: 27385830
4. **Olsen A, Chen Y, Ji Q, Zhu G, De Silva AD, Vilchèze C, Weisbrod T, Li W, Xu J, Larsen M, Zhang J, Porcelli SA, Jacobs WR Jr, Chan J.** (2016). "Targeting Mycobacterium tuberculosis Tumor Necrosis Factor Alpha-Downregulating Genes for the Development of Antituberculous Vaccines." *MBio.* 7(3). pii: e01023-15. PMID: 27247233.
5. **Tufariello JM, Chapman JR, Kerantzas CA, Wong KW, Vilchèze C, Jones CM, Cole LE, Tinaztepe E, Thompson V, Fenyö D, Niederweis M, Ueberheide B, Philips JA, Jacobs WR Jr.** (2015). Separable Roles for Mycobacterium tuberculosis ESX-3 Effectors in Iron Acquisition and Virulence. *PNAS.* 113(3):E348-57. PMCID: PMC4725510
6. **Berney, M., L. Berney-Meyer, K. W. Wong, B. Chen, M. Chen, J. Kim, J. Wang, D. Harris, J. Parkhill, J. Chan, F. Wang, and W. R. Jacobs, Jr.** (2015). Essential roles of methionine and S-adenosylmethionine in the autarkic lifestyle of Mycobacterium tuberculosis. *Proc Natl Acad Sci U S A* 112 (32):10008-13. PMCID:PMC4538671.
7. **Tufariello JM., Kerantzas CA., Vilchèze C., Calder RB., Nordberg EK., Fischer JA., Hartman TE., Yang E., Driscoll T., Cole LE., Sebra R., Maqbool SB., Wattam AR., Jacobs WR Jr.** (2015). The Complete Genome Sequence of the Emerging Pathogen *Mycobacterium haemophilum* Explains Its Unique Culture Requirements. *MBio.* 6(6)e01313-15. PMID: 26578674
8. **Petro, C., González, P.A., Cheshenko, N., Jandl, T., Khajjoueinejad, N., Bénard, A., Sengupta, M., Herold, B.C., Jacobs, W.R., Jr.** (2015) Herpes simplex type 2 virus deleted in glycoprotein D protects against vaginal, skin and neural disease. *eLife*. PMCID PMC4352706.
9. **Pope, W.H., Bowman, C.A., Russell, D.A., Jacobs-Sera, D., Asai, D.J., Cresawn, S.G., Jacobs, W.R., Jr., Hendrix, R.W., Lawrence, J.G., Hatfull, G.F**. (2015) Whole genome comparison of a large collection of mycobacteriophages revails a continuum of phage genetic diversity. *eLife* PMCID PMC 4408529.
10. **Hartman, T., Weinrick, B., Vilcheze, C., Berney, M., Tufariello, J.M., G. M. Cook., and W. R. Jacobs, Jr.** (2014) Succinate dehydrogenase is the regulator of respiration in *Mycobacterium tuberculosis*. *PloS Pathogens*. 10(11):e1004510. PMCID PMC 4161257.
11. **Panas, M.W., Jain, P., Yang, H., Mitra, S., Biswas, D., Wattam, A.R., Letvin, N.L., Jacobs, W.R.,Jr.** (2014) Noncanonical SMC protein in *Mycobacterium smegmatis* restricts maintenance of *Mycobacterium fortuitum plasmids*. *Proc Natl Acad Sci.* **111** (37)**:**13264-71. PMCID PMC 4169951.
12. **Vilcheze, C., Molle, V., Carrere-Kremer, S., Leiba, J., Mourey, L., Shenai, S., Baronian, G., Tufariello, J., Hartman, T., Veyron-churlet, R., Trivelli, X., Tiwari, S., Weinrick, B., Alland, D., Guerardel, Y., Jacobs, W.R. Jr., Kremer, L.,** (2014) Phosphorylation Signaling by Ser/Thr Kinases Regulates Virulence and Acid-Fast

Staining in *Mycobacterium tuberculosis*. *PLoS Pathogen.* **10** (5)**:**e1004115. PMCID PMC4014462.

13. **Tufariello, J.M., Malek, A.A., Vilcheze, C., Cole,L.E., Ratner, H.K. Gonzalez, P.A., Jain, P., Hatfull, G.F., Larsen, M.H., Jacobs, W..R. (2014)** Enhanced Specialezed Transduction Using Recombineering in *Mycobacterium tuberculosis**.*** *mBio* 5:(3) e01179-14 PMCID PMC4045075.
14. **Jain, P., Hsu, T., Arai, M., Biermann, K. E., Thaler, D. S., Nguyen, A., Gonzalez, P., Tufariello, J.M., Kriakov, J., Chen, B., Larsen, M. H., Jacobs Jr, W. R., Jr.** (2014) Specialized Transduction Designed for Precise High-throughput Unmarked Deletions in *Mycobacterium tuberculosis. mBio* **5** (3)**:**e01245-14. PMCID PMC4049104.
15. **Vilchèze, C., Hartman, T., Weinrick, B., and Jacobs, W.R.** (2013). *Mycobacterium tuberculosis* is extraordinarily sensitive to killing by a vitamin C-induced Fenton reaction. *Nat. Commun* 4:1881. PMCID PMC 3698613.

11. DR. WILLIAM R. JACOBS, JR.

DR. GANJAM V. KALPANA

Molecular Genetic Analysis of Tumor Suppressor INI1/hSNF5 in HIV-1 Replication and Cancer

INI1/hSNF5 is a component of the chromatin remodeling SWI/SNF complex. It is an interacting partner for HIV-1 integrase (IN) and also a tumor suppressor biallelically mutated in rhabdoid tumors, a rare but highly aggressive pediatric malignancy. The two major areas of focus in the laboratory are: (i) understanding the role of INI1 in HIV-1 replication and exploring its potential as a drug target for intervention of AIDS; and (ii) understanding the mechanism of tumor suppression by INI1/hSNF5 and developing novel and effective therapeutic strategies for rhabdoid tumors.

INI1 in HIV-1 replication: We have found that INI1/hSNF5 directly binds and recruits components of Sin3a-histone deacetylase (HDAC) complex into the HIV-1 virions and this HDAC1 complex appears to be required for viral infectivity. We are currently isolating and characterizing IN and INI1 mutants defective for binding to HDAC1 complex and testing their effect on HIV-1 replication. We have found that HIV-1 harboring IN mutants defective or binding to INI1 are severely compromised for replication. Furthermore, we have found that INi1 mutants defective for binding to HDAC1 complex dominant negatively inhibit HIV-1 but not SIV replication. These studies are likely to open up a new paradigm for role of INI1 in HIV-1 replication and may provide novel strategies to inhibit viral replication.

Mechanism of Tumor suppression by INI1/hSNF5: By using a series of genetic systems developed in our laboratory and by isolating cancer-associated mutations of INI1, and a wealth of protein-protein interaction defective mutants of INI1, we are dissecting the exact mechanism of INI1-medaited G0/G1 cell cycle arrest, mitotic arrest, and senescence and tumor suppression. Furthermore, characterizing the INI1-associated HDAC1 complex has revealed an unanticipated role of INI1 in interferon signaling and tumor suppression.

Development of targeted therapies for rhabdoid tumors based on INI1 function: One of the goals of our laboratory is to develop molecularly targeted therapies based on the understanding of genesis of rhabdoid tumors. Majority of rhabdoid tumors have biallelic inactivation of *INI1* gene. Our previous studies demonstrated that Cyclin D1 is a direct downstream target of INI1 mediated repression and that rhabdoid tumors are exquisitely dependent on Cyclin D1 for genesis and survival. Our preclinical studies have provided proof of principle for our hypothesis that targeting Cyclin/cdk axis is an effective means of inhibiting rhabdoid tumors *in vitro* and *in vivo*. The current goal is to develop novel strategies to facilitate clinical translation of laboratory findings to establish an effective therapy for these tumors. For this purpose, we are using non-invasive imaging technology such as microPET to monitor the therapeutic efficacy in primary mouse tumor models, developing novel drugs to target these tumors and investigating the interaction between *Cyclin D1*, the cdk pathway and *Ini1* in mouse models.

Identification of downstream pathways regulated by *INI1* has been instrumental in novel biomarkers and therapeutic targets for these tumors. *Aurora A* is repressed by *INI1* and it is de-repressed in rhabdoid tumors due to loss of *INI1*. We have found that Aurora A is a novel therapeutic target as siRNA-mediated depletion of this gene resulted in potent mitotic catastrophe and cell death in rhabdoid tumors.

PUBLICATIONS:

Smith ME, Cimica V, Chinni S, Challagulla K, Mani S**, Kalpana GV.** Rhabdoid tumor growth is inhibited by flavopiridol. *Clin Cancer Res.* 2008 Jan 15;14(2):523-32.

Das BC, Smith ME**, Kalpana GV.** Design, synthesis of novel peptidomimetic derivatives of 4-HPR for rhabdoid tumors. Bioorg Med Chem Lett. 2008 Jul 15;18(14):4177-80. Epub 2008 May 29.

Das S, Cano J**, Kalpana GV.** Multimerization and DNA Binding Properties of INI1/hSNF5 and Its Functional Significance. J Biol Chem. 2009 Jul 24;284(30):19903-14. Epub 2009 Apr 27.

Sorin M, Cano J, Das S, Mathew S, Wu X, Davies KP, Shi X, Cheng SW, Ott D**, Kalpana GV.** Recruitment of a SAP18-HDAC1 complex into HIV-1 virions and its requirement for viral replication. *PLoS Pathog*. 2009 Jun;5(6).

Kalpana, G. V. and M. Smith (2009) "Development of targeted therapies for rhabdoid tumors based on the functions of *INI1/hSNF5* tumor suppressor" (*invited chapter for the monograph on Molecularly targeted Therapies for pediatric tumors Edited by Bob Arceci and Peter Houghton*).

Smith ME, Cimica V, Chinni S, Jana S, Koba W, Yang Z, Fine E, Zagzag D, Montagna C**, Kalpana GV.** Therapeutically targeting Cyclin D1 in primary tumors arising from loss of *Ini1.* Proc Natl Acad Sci U S A. 2011 Jan 4;108(1):319-24. Epub 2010 Dec 20.

Jennifer Cano and **Ganjam V. Kalpana.** Inhibition of early stages of HIV-1 assembly by INI1/hSNF5 transdominant negative mutant S6. J Virol. 2011 Mar; 85(5):2254-65. Epub 2010 Dec 15.

SeungJae Lee, Velasco Cimica, David Zagzag and **Ganjam V. Kalpana.** Aurora A Is a Repressed Effector Target of the Chromatin Remodeling Protein INI1/hSNF5 Required for Rhabdoid Tumor Cell Survival. Cancer Res. 2011 May 1;71(9):3225-35. Epub 2011 Apr 26.

DR. LIBUSHA KELLY
http://www.kellylab.org/

Microbial populations are dynamic, transactional social networks of paramount importance to human health. The collection of microbes living in and on our bodies (the 'microbiome') is influenced by top-down and bottom-up regulation and these processes are poorly understood. For example, the availability of resources produced by other microbes ('public goods') can inhibit or support the growth of particular microbes (bottom-up). Administering a drug or viral predation can remodel microbial community structure (top-down). Our research program focuses on how microbial populations in the human body respond to perturbations such as diet, disease, and drugs, and how these responses are linked to health outcomes. The overall goal of our work is to empower patients to improve their health via targeted control of their microbiome. Towards this end we study how microbial communities are influenced by top-down and bottom-up regulation with a focus on three main biological questions: 1) How does microbial metabolism of drugs influence treatment outcomes in patients? Here we develop pretherapy analysis protocols to identify patients with high-risk microbiomes; we propose novel prebiotic approaches to influence microbial drug metabolism; and we predict new microbiome/drug interactions. 2) How do interactions between bacteria and archaea at the bottom of the microbial food chain influence access to dietary substrates? We endeavor to increase access to beneficial dietary compounds and we study how aging influences food processing in the gut. 3) How do virus/host interactions influence population-level metabolism in the gut? We predict viral influences on microbiome health and function and we identify new viruses that infect microbial populations in the human gut. Our approaches borrow from many fields and include metabolomics, high throughput genomics, information theory, synthetic chemistry, flow sorting, and imaging. Taken together, my research program forms the foundation of a new field of targeted microbiome manipulation for personalized health care.

Complete Publication List:
https://scholar.google.com/citations?user=sg7-rm4AAAAJ&hl=en

Selected Publications and Products:
† Indicates that authors contributed equally to the publication

Xenobiotic metabolism and the human microbiome

Leah Guthrie, Sanchit Gupta, Johanna Daily, **Libusha Kelly**. "Human microbiome signatures of differential colorectal cancer drug metabolism". Accepted, Nature Biofilms and Microbiomes.

Ian A. Downs, Olga Aroniadis, **Libusha Kelly**, and Lawrence J. Brandt. "Post-infection irritable bowel syndrome: the links between gastroenteritis, inflammation, the microbiome, and functional disease". In press, Journal of Clinical Gastroenterology.

Wallace BD, Roberts AB, Pollet RM, Ingle JD, Biernat KA, Pellock SJ, Venkatesh MK, Guthrie L, O'Neal SK, Robinson SJ, Dollinger M, Figueroa E, McShane SR, Cohen RD, Jin J, Frye SV, Zamboni WC, Pepe-Ranney C, Mani S, **Kelly L**, Redinbo MR, "Structure and Inhibition of Microbiome β-Glucuronidases Essential to the Alleviation of Cancer Drug Toxicity." Chem Biol. 2015 Sep 17;22(9):1238-49. doi: 10.1016/j.chembiol.2015.08.005.

Patent submitted to U.S. Patent Office. International Patent Application Number PCT/US2016/52742. Title: Endogenous microbial hyperswarmers protect against intestinal inflammation. **Libusha Kelly** and Sridhar Mani.

Phage and bacterial genomics and metagenomics

Kathryn M. Kauffman, Fatima A. Hussain, Joy Yang, Phil Arevalo, Julia M. Brown, William K. Chang, Michael B. Cutler, **Libusha Kelly***, Martin F. Polz*. "Nontailed viruses are major unrecognized killers of bacteria in the ocean." In revision, Nature. (*=co-corresponding author)

Kelly L, Ding H, Huang KH, Osburne MS, Chisholm SW. Genetic diversity in cultured and wild marine cyanomyoviruses reveals phosphorus stress as a strong selective agent. *ISME J. 2013 May 9. doi: 10.1038/ismej.2013.58. [Epub ahead of print]*

Kelly L†, Huang KH†, Ding H, Chisholm SW. ProPortal: A resource for integrated systems biology of *Prochlorococcus* and its phage. *Nucleic Acids Res. 2012 Jan;40(Database issue):D632-40.*

Malmstrom RR†, Rodrigue S†, Huang KH**, Kelly L**, Kern SE, Thompson A, Roggensack S, Berube PM, Henn MR, Chisholm SW. Ecology of uncultured *Prochlorococcus* clades revealed through single-cell genomics and biogeographic analysis. *ISME J*. 2012 Aug 16. *[Epub ahead of print].*

Thompson LR†, Zeng Q†, **Kelly L**, Huang KH, Singer AU, Stubbe J, Chisholm SW. Phage auxiliary metabolic genes and the redirection of cyanobacterial host carbon metabolism. *Proc Natl Acad Sci U S A*. 2011 Sep 27;108(39):E757-64.

Li B†, Sher D†, **Kelly L**, Shi Y, Huang K, Knerr PJ, Joewono I, Rusch D, Chisholm SW, van der Donk WA. Catalytic promiscuity in the biosynthesis of cyclic peptide secondary metabolites in planktonic marine cyanobacteria. *Proc Natl Acad Sci* U S A. 2010 Jun 8;107(23):10430-5.

Computational analysis of human genetic variation

Kelly L†, Fukushima H†, Karchin R, Gow JM, Chinn LW, Pieper U, Segal MR, Kroetz DL, Sali A. Functional hotspots revealed by mutational, evolutionary, and

structural characterization of ABC transporters. *Protein Sci. 2010 Nov;19(11):2110-21.*

Kelly L, Karchin R, Sali A. Protein interactions and disease phenotypes in the ABC transporter superfamily. *Pac Symp Biocomput*. 2007:51-63.

Karchin R, Diekhans M, **Kelly L**, Thomas DJ, Pieper U, Eswar N, Haussler D, Sali A. LS-SNP: large-scale annotation of coding non-synonymous SNPs based on multiple information sources. *Bioinformatics*. 2005 Jun 15;21(12):2814-20.

Karchin R, **Kelly L**, Sali A. Improving functional annotation of non-synonomous SNPs with information theory. *Pac Symp Biocomput. 2005:397-408.*

Olivier M, Aggarwal A, Allen J, Almendras AA, Bajorek ES, Beasley EM, Brady SD, Bushard JM, Bustos VI, Chu A, Chung TR, De Witte A, Denys ME, Dominguez R, Fang NY, Foster BD, Freudenberg RW, Hadley D, Hamilton LR, Jeffrey TJ, **Kelly L**, Lazzeroni L, Levy MR, Lewis SC, Liu X, Lopez FJ, Louie B, Marquis JP, Martinez RA, Matsuura MK, Misherghi NS, Norton JA, Olshen A, Perkins SM, Perou AJ, Piercy C, Piercy M, Qin F, Reif T, Sheppard K, Shokoohi V, Smick GA, Sun WL, Stewart EA, Fernando J, Tejeda, Tran NM, Trejo T, Vo NT, Yan SC, Zierten DL, Zhao S, Sachidanandam R, Trask BJ, Myers RM, Cox DR. A High-Resolution Radiation Hybrid Map of the Human Genome Draft Sequence. *Science. 2001 Feb 16;291(5507):1298-302.*

Computational sequence and structure analysis of membrane proteins

Kelly L, Pieper U, Eswar N, Hays FA, Li M, Roe-Zurz Z, Kroetz DL, Giacomini KM, Stroud RM, Sali A. A survey of integral α–helical membrane proteins. *J Struct Funct Genomics. 2009 Dec;10(4):269-80.*

Li M, Hays FA, Roe-Zurz Z, Vuong L, **Kelly L**, Ho CM, Robbins RM, Pieper U, O'Connell JD 3rd, Miercke LJ, Giacomini KM, Sali A, Stroud RM. Selecting optimum eukaryotic integral membrane proteins for structure determination by rapid expression and solubilization screening. *J Mol Biol.* 2009 Jan 23;385(3):820-30.

Hays FA, Roe-Zurz Z, Li M, **Kelly L**, Gruswitz F, Sali A, Stroud RM. Ratiocinative screen of eukaryotic integral membrane protein expression and solubilization for structure determination. *J Struct Funct Genomics.* 2009 Mar;10(1):9-16.

DR. GREGOIRE LAUVAU

Immune effector cell differentiation & protective host responses against microbial pathogens and tumors *in vivo*

Microbial pathogens invasion usually triggers potent host immune responses, however efficient protection and pathogen killing require the presence of effector cells and combinations of inflammatory signals that are ill-defined in most infections. Tumor often escape immune responses through a variety of mechanisms including immune suppression.

Our work therefore focuses on precisely defining these events in various settings *in vivo*. Specifically, we investigate (i) the inflammatory signals and related pathways, and innate immune cells that regulate T and innate cell differentiation, and (ii) the cross-talks between memory T cells and innate immune cells. Innate immune cells include monocytes, macrophages, dendritic cells and lymphocytes. We use various models of acute microbial pathogen infections in mice, namely the bacteria *Listeria monocytogenes*, *Streptococcus pneumoniae*, the viruses Vesicular Stomatitis virus and Murine Cytomegalovirus. We study the immune response to the parasite of malaria *Plasmodium* in surrogate mouse models and in human patients. We also investigate the role of poor prognosis mutations found in breast cancer patients on immune cells functions. We take advantage of a range of advanced fluorescent-tracer based methodologies and intravital microscopy to monitor and visualize immune cells *in situ*. We use cell transfer experiments and novel genetically modified mice models in which dynamic cell functions can be monitored and/or in which functional subsets of immune cells can be selectively eliminated. We also make use of the latest cutting edge approaches to analyse immune cell expression and epigenetic programming, as well as high end cytometry by time of flight. Overall, the goal of my laboratory is to improve our fine understanding of the factors that orchestrate antimicrobial and antitumoral host protective immune responses *in vivo*. We believe that our work will contribute to better immune cell-mediated preventive and therapeutic vaccination strategies.

PUBLICATIONS

1. Chorro L., Spaulding E. and **G. Lauvau**. Blockade of LAG-3 but not of PD-1/PD-L1 enhances T cell-mediated clearance of blood stage malaria. ***In revision*.**
2. Spaulding E., Fooksman D., Moore JM., Saidi, Feintuch C., Reizis B., Chorro L., Daily J., and **G. Lauvau**. 2016. STING-Licensed Macrophages Prime Type I IFN Production by Plasmacytoid Dendritic Cells in the Bone Marrow during Severe *Plasmodium yoelii* Malaria. **PLoS Pathogens**, 12(10):e1005975.
3. Subramaniam K.S., Spaulding E., Ivan E., Mutimura E., Kim R.S., Liu X., Dong C., Feintuch C.M., Zhang X., Anastos K., **Lauvau G**. and Daily J.P. 2015. The T cell inhibitory molecule BTNL2 is upregulated in mild *Plasmodium falciparum* infection and is protective during experimental cerebral malaria. **J Infect Dis**, Apr 15. pii: jiv217.
4. Chandrabos C.*, M'Homa Soudja S. *, Weinrick B., Gros M., Frangaj A., Rahmoun M., Jacobs W.R. Jr. and **G. Lauvau**. 2014. The p60 and NamA Autolysins from *Listeria monocytogenes* Contributes to Host Colonization and Induction of Protective Memory. **Cell Microbiol**, 17(2):147-63.
5. Anne L. Ruiz,, Saidi M'Homa Soudja, Cyril Deceneux, Grégoire Lauvau and Julien C. Marie. 2014. $NK1.1^{+}$ $CD8^{+}$ T Cells Escape TGFbeta control and Contribute to Early Microbial Pathogen response. 2014. **Nature Communication**, 5:5150.

6. Soudja M'Homa S., Jacob E., Chandrabos C., M. Veenstra, Palliser D. and **G. Lauvau**. 2014. Memory T cell-derived IFN-γ instructs potent innate cell activation for protective immunity. **Immunity**, 40(6):974-88.
7. Soudja M'Homa S., Ruiz A. L., Marie J. C., and **G. Lauvau**. 2012. Inflammatory monocytes switch on memory $CD8^+$ T and innate NK lymphocytes during microbial pathogens invasion. **Immunity**, 37(3):549-62.
8. Narni-Mancinelli E., Soudja M'Homa S., Crozat K., Dalod M., Gounon P., Geissmann F. and **G. Lauvau**. 2011. Inflammatory monocytes and neutrophils are licensed to kill during Memory responses *in vivo*. **PLoS Pathogens**, Vol 7(12):e1002457.
9. Campisi L., S. Soudja M'Homa, N. Cagnard, J. Cazareth, D. Bassand, A. Lazzari, E. Narni-Mancinelli, F. Brau, N. Glaichenhaus, F. Geissmann and **G. Lauvau**. 2011. Splenic $CD8^+$ Dendritic Cells Undergo Rapid Programming by Cytosolic Bacteria and Inflammation to Induce Protective CD8 Memory. **Eur J Immunol**, 41(6):1594-605.
10. Rahmoun M., M. Gros, L. Campisi, D. Bassand, A. Lazzari, C. Massiera, E. Narni-Mancinelli, P. Gounon, and G. Lauvau. 2011. Priming of protective anti-*Listeria* memory $CD8^+$ T cell requires a functional SecA2-secretion system. **Infection and Immunity**, 79:2396-403.
11. Bajénoff M., Narni-Mancinelli E., Brau F., and **G. Lauvau**. 2010. Visualizing early splenic memory $CD8^+$ T cells reactivation against intracellular bacteria. **PLoS One**. 5:11524.
12. Auffray C.[*], D. K. Fogg[*], E. Narni-Mancinelli[*], B. Senechal, C. Touillet, N. Saederup, J. Leemput, K. Bigot, L. Campisi, M. Abitbol, T. Molina 1, I. Charo, D. A. Hume, A. Cumano, **G. Lauvau** and F. Geissmann. 2009. CX3CR1+ CD115+ CD135+ common macrophage/DC precursor (MDP) and the role of CX3CR1 in their response to inflammation. **J Exp Med**, 206: 595-606.
13. Narni-Mancinelli E., L. Campisi, D. Bassand, J. Cazareth, P. Gounon, N. Glaichenhaus and **G. Lauvau**. 2007. Memory $CD8^+$ T cells mediate antibacterial immunity via CCL3 activation of TNF/ROI^+ phagocytes. **J Exp Med**, 204: 2075-2087.
14. Auffray, C., D. K. Fogg, Meriem Garfa**,** G. Elain**,** O. Join-Lambert, S. Kayal, S. Sarnacki, A. Cumano, **G. Lauvau** and F. Geissmann. 2007. Patrolling blood monocytes that monitor blood vessels and tissues for damage and infection. **Science**, 317, 666-70.
15. Muraille, E.[*], E. Narni-Mancinelli[*], P. Gounon[*], D. Bassand, N. Glaichenhaus, L. L. Lenz, and **G. Lauvau**. 2007. Cytosolic Expression of SecA2 is a Prerequisite for Long-Term Protective Immunity. **Cell Microbiol**, 9, 1445-1454.
16. Muraille, E., R. Giannino, P. Guirnalda, I. Leiner, S. Jung, E. G. Pamer and **G. Lauvau.** 2005. Distinct *in vivo* dendritic cell activation by live versus killed *Listeria monocytogenes.* **Eur J Immunol,** 35:1-9*.*
17. Lauvau, G.*, S. Vijh*, P. Kong, T. Horng, K. Kerksiek, N. Serbina, R.A. Tuma, and E.G. Pamer. 2001. Priming of memory but not effector CD8 T cells by a killed bacterial vaccine. **Science**, 294:1735-1739.

Chapters in Books and Review Articles:

1. Lauvau, G. and S. Goriely. Memory CD8 T cells: Orchestrators and Key Players of Innate Immunity? 2016. **PLoS Pathogens**, 12(9):e1005722.
2. Lauvau, G., M. Boutet, T.M. Williams, M. Shu Shien Chin and L. Chorro**.** Memory $CD8^+$ T Cells: Innate-Like Sensors and Orchestrators of Protection. 2016. **Trends in Immunology**, 37(6):375-85.
3. **Lauvau G.**, Loke P., Holh T.M. Monocyte Mediated Defense Against Pathogenic Microbes. 2015. **Seminars in Immunology**, 27(6):397-409.

4. **Lauvau G.**, S. M'Homa Soudja. 2015. Mechanisms of Memory T Cell Activation and Effective Immunity. **Adv Exp Med Biol,** 850:73-80.
5. **Lauvau G**. 2015. PI3-kinase, the key for bugs to get in? **J Exp Med,** 9;212(2):127.
6. **Lauvau G.**, L. Chorro, E. Spaulding, S. M'Homa Soudja. 2014. Inflammatory Monocyte Effector Mechanisms. **Cellular Immunology**, S0008-8749(14)00125-7.
7. **Lauvau G**., M'Homa Soudja S., Chorro L. 2013. T cell Memory. **Encyclopedia of Medical Immunology**, I.R. Mackay, N.R. Rose (eds.), Springer Science, DOI 10.1007/978-0-387-84828-0.
8. Geissmann F., C. Auffray, R. Palframan, C. Wirrig, A. Ciocca, L. Campisi, E. Narni-Mancinelli, and **G. Lauvau**. 2008**.** Blood monocytes: distinct subsets, how they relate to dendritic cells, and their possible roles in the regulation of T cell responses. **Immunology and Cell Biology**, 86:398-408.

DR. THOMAS S. LEYH

My laboratory is focused on understanding the molecular basis of life. Our interdisciplinary pursuit of this issue has provided a broad experimental platform for our work and has proven a recipe for discovery. For example, my group discovered the link between sulfur biology and GTPase function; a linkage that rests with the enzyme ATP sulfurylase, which allosterically couples the chemical potential of GTP hydrolysis to the synthesis of activated sulfate (*APS,* adenosine 5'-phosphosulfate), an essential sulfur metabolite. Our inquiries in this area have revealed further that this same enzyme forms a complex with its partners in the cysteine biosynthetic pathway, and, remarkably, that new catalytic function emerges from this complex - the hydrolysis of ATP. In this case, it is the energy of ATP hydrolysis that is linked to the synthesis of APS. This finding underscores how cellular components can combine in synergistic ways to create hierarchies of function. Such hierarchies, whose behaviors are rooted in the reduction of entropy, are not well understood, and are of keen interest to us. First principles of chemistry and enzymology suggested that ATP sulfurylases that are not linked to an external energy source, such as ATP or GTP hydrolysis, might transfer APS directly to the active site of the next enzyme in the metabolic pathway, APS kinase. We have shown that in a spectacular display of the interplay of structure and function, certain sulfate activating complexes transfer APS directly between the active-sites of ATP sulfurylase and APS kinase *via* a 75Å-long groove that opens and closes in response to the position of the nucleotide within the groove.

Transfer of the sulfuryl-moiety ($-SO_3^-$) from activated sulfate to biological acceptors is used widely by the cell to regulate metabolism, and the extent to which a particular metabolite is sulfated is determined by the balance of the *in-vivo* activity of the sulfotransferase (which transfers the sulfuryl-group) and sulfatase (which hydrolytically removes it). Compelling, disease-relevant biology pivots on the activities of each of the six known cytosolic sulfotransferase isozymes. Our laboratory has concentrated primarily on estrogen sulfotransferase (EST), which sulfates estrogen and thereby prevents it from binding to and activating the estrogen receptor. Aberrant sulfation of estrogen is tightly, causally linked to cancer in primary estrogen-dependent breast tumors. We have recently determined the first transition-state structure of an enzyme catalyzed sulfuryl-transfer reaction – that of EST. While it is quite gratifying to "see" precisely how the electronic structure of the bonds involved in the transfer reorganizes as enzyme-bound substrate moves between their ground- and transition-states, the structure is also of considerable practical value in that it defines the target for the design and synthesis of sulfuryl-transfer transition-state inhibitors (a perfect transition-state mimic is expected to inhibit with picomolar affinity).

Streptococcus pneumonia, a multiple-drug resistant organism, is estimated to take the lives of 3600 people daily, the majority of whom are children and the elderly. We discovered recently that mevalonate kinase, an essential enzyme in the isoprenoid biosynthetic pathway in *Streptococcus pneumoniae*, is potently allosterically inhibited by diphposphomevalonate (*DPM*), a downstream intermediate in the pathway, and that the human isozyme is not. Genetic and animal studies of this multiple-drug resistant organism have taught us that the mevalonate pathway is essential for the survival of *S. pneumonia* in the lung, and human serum. Consequently, we have undertaken a major research effort to develop novel antibiotics that target these enzymes in gram-positive bacteria. The program, carried out under the auspices of the *NIAID*, brings together an interdisciplinary team of faculty, postdoctoral fellows and graduate students in the areas of high-resolution NMR-spectroscopy, crystallography, synthetic chemistry, and biochemistry to explore fundamental issues of allostery, catalysis, and inhibition in these systems. I am pleased to report that our recent efforts have produced inhibitors that act with nanomolar affinity at each of multiple points in the pathway, and are capable of killing infectious *S. pneumonia* in rich media at ~ 25 □g/ml.

The three projects outlined above comprise the core of our research activities; however, we are also expanding into two new areas. The etiology of how *M. tuberculosis* emerges from dormancy in the lung tubercle is not yet well understood. We are beginning to define the molecular logic of this transition with Prof. John Chan, an expert mycobacteriologist who has isolated a mutation that activates growth by derepressing dormancy. While the protein that harbors this mutation has not yet been assigned molecular function, we now know that it co-purifies with one-equivalent of adenine nucleotide bound to it, that it slowly hydrolyses ATP, and that its sequence identifies it as a possible element of a signaling network – we are currently testing this hypothesis using genetic and biochemical assays. In a final program, we are collaborating with a single molecule spectroscopist, and biological chemist at Columbia University to better understand and control the ribosomal editing functions of the protein-synthetic machinery toward mis-acylated tRNA. Editing protects against disease by recognizing and rejecting misacylated tRNA; controlling editing gives way to regiospecific incorporation of non-natural amino acids into proteins, which will facilitate myriad scientific endeavors including single-molecule exploration of the cellular *milieu*.

PUBLICATIONS

1. Pinto, R., Leotta, L., Shanahan, E. R., West, N. P., **Leyh, T**. **S**., Britton, W., and Triccas, J. A. (2013) Host cell-induced components of the sulfate assimilation pathway are major protective antigens of Mycobacterium tuberculosis, *J Infect Dis 207*, 778-785.
2. Cook, I., Wang, T., Almo, S. C., Kim, J., Falany, C. N., and **Leyh, T. S**. (2013) The gate that governs sulfotransferase selectivity, *Biochemistry 52*, 415-424.
3. Cook, I., Wang, T., Almo, S. C., Kim, J., Falany, C. N., and **Leyh, T. S**. (2013) Testing the sulfotransferase molecular pore hypothesis, *J Biol Chem 288*, 8619-8626.
4. Wang, T., and **Leyh, T. S.** (2012) Three-stage assembly of the cysteine synthase complex from Escherichia coli, *J Biol Chem 287*, 4360-4367.
5. Rohn, K. J., Cook, I. T., **Leyh, T. S.**, Kadlubar, S. A., and Falany, C. N. (2012) Potent inhibition of human sulfotransferase 1A1 by 17alpha-ethinylestradiol: role of 3'-phosphoadenosine 5'-phosphosulfate binding and structural rearrangements in regulating inhibition and activity, *Drug Metab Dispos 40*, 1588-1595.
6. Lefurgy, S. T., and **Leyh, T. S.** (2012) Analytical expressions for the homotropic binding of ligand to protein dimers and trimers, *Anal Biochem 421*, 433-438.
7. Cook, I., Wang, T., Falany, C. N., and **Leyh, T. S.** (2012) A nucleotide-gated molecular pore selects sulfotransferase substrates, *Biochemistry 51*, 5674-5683.
8. Reuther, G., Harris, R., Girvin, M., and **Leyh, T. S.** (2011) Backbone 1H, 13C, 15N NMR assignments of the unliganded and substrate ternary complex forms of mevalonate diphosphate decarboxylase from Streptococcus pneumoniae, *Biomol NMR Assign 5*, 11-14.
9. Tang, Q., and **Leyh, T. S.** (2010) Precise, facile initial rate measurements, *J Phys Chem B 114*, 16131-16136.
10. Sun, M., and **Leyh, T. S.** (2010) The human estrogen sulfotransferase: a half-site reactive enzyme, *Biochemistry 49*, 4779-4785.
11. Lefurgy, S. T., Rodriguez, S. B., Park, C. S., Cahill, S., Silverman, R. B., and **Leyh, T. S.** (2010) Probing ligand-binding pockets of the mevalonate pathway enzymes from Streptococcus pneumoniae, *J Biol Chem 285*, 20654-20663.
12. Kudoh, T., Park, C. S., Lefurgy, S. T., Sun, M., Michels, T., **Leyh, T. S.**, and Silverman, R. B. (2010) Mevalonate analogues as substrates of enzymes in the isoprenoid biosynthetic pathway of Streptococcus pneumoniae, *Bioorg Med Chem 18*, 1124-1134.

13. Cook, I. T., **Leyh, T. S.**, Kadlubar, S. A., and Falany, C. N. (2010) Structural rearrangement of SULT2A1: effects on dehydroepiandrosterone and raloxifene sulfation, *Horm Mol Biol Clin Investig 1*, 81-87.
14. Cook, I. T., **Leyh, T. S.**, Kadlubar, S. A., and Falany, C. N. (2010) Lack of substrate inhibition in a monomeric form of human cytosolic SULT2A1, *Horm Mol Biol Clin Investig 3*, 357-366.
15. Apweiler, R., Armstrong, R., Bairoch, A., Cornish-Bowden, A., Halling, P. J., Hofmeyr, J. H., Kettner, C., **Leyh, T. S.**, Rohwer, J., Schomburg, D., Steinbeck, C., and Tipton, K. (2010) A large-scale protein-function database, *Nat Chem Biol 6*, 785.
16. Effraim, P. R., Wang, J., Englander, M. T., Avins, J., **Leyh, T. S.**, Gonzalez, R. L., Jr., and Cornish, V. W. (2009) Natural amino acids do not require their native tRNAs for efficient selection by the ribosome, *Nat Chem Biol 5*, 947-953.

DR. JOSHUA D. NOSANCHUK

Key Words: fungus, histoplasmosis, cryptococcosis, candidiasis, melanin

- *Histoplasma capsulatum*: Research on this fungus pertains to the development of active and passive immunotherapeutics. Since individuals with severe histoplasmosis [such as AIDS patients with disseminated disease] often lack effective cell-mediated immune responses, induction of an effective humoral response or passive administration of antibody has tremendous therapeutic potentials. My laboratory is the first to identify protective monoclonal antibodies for the treatment of *H. capsulatum* infection. We are studying the mechanisms of antibody efficacy and are testing the recombinant antigens as a potential prophylactic and therapeutic vaccine. Also, we have embarked with the Nathenson and Almo labs to study the impact of co-stimulation on histoplasmosis and have demonstrated that the PD-1/PDL pathway is critically important to disease pathogenesis. Antibodies can interfere with this negative co-stimulation pathway and prevent lethal histoplasmosis.

- *Candida parapsilosis*: This is the newest fungus to the laboratory. The incidence of *C. parapsilosis* infections has exploded in recent years and very little is known about its virulence. We developed the first efficient method for targeted gene deletion for *C. parapsilosis* and are actively pursuing targets to define what makes this fungus pathogenic, with a particular emphasis on secreted hydrolytic proteins and lipid metabolism. We are also targeting virulence associated genes in *C. albicans* and studying their role in pathogenesis.

- *Cryptococcus neoformans*: We are primarily using this pathogen to elucidate the impact of melanin in pathogenic fungi. Melanin is a complex polymer of unknown structure that is prevalent throughout the biological kingdoms. Ongoing investigations are examining mechanisms of melanin synthesis and rearrangement by molecular, physical, and immunological methods. This work is in collaboration with Dr. Arturo Casadevall, Departments of Microbiology & Immunology and Medicine.

Additional areas of special emphasis:

Methamphetamine: This drug has increasingly become a major scourge on our society. Although the behavioral impact of methamphetamine is well understood, there is a dearth of data on the effect of the drug on immune function. We have established that methamphetamine significantly adversely regulates diverse aspects of immunity. We have an ongoing program to further elucidate the mechanisms and impact of this disregulation.

Nitric oxide releasing nanoparticles: We are exploring the therapeutic potential of this novel compound for the treatment of diverse infectious diseases, including bacterial and fungal diseases. This work is in collaboration with Dr. Joel Friedman, Departments of Medicine and Physiology & Biophysics.

PUBLICATIONS

Targeted gene deletion in a clinical *Candida papapsilosis* isolate demonstrates the role of secreted lipase in virulence and pathogenesis. Journal of Clinical Investigation. 2007. 117(10):3049-58

The PD-1/PD-L costimulatory pathway critically affects host resistance to the pathogenic fungus *Histoplasma capsulatum*. Proc Natl Acad Sci U S A. 2008 Feb 19;105(7):2658-63.

Methamphetamine enhances histoplasmosis by immunsuppression of the host. Journal of Infectious Diseases. 2009. 200:131-141.

Monoclonal antibodies to heat shock protein 60 alter the pathogenesis of *Histoplasma capsulatum*. Infection and Immunity. 2009. 77(4):1357-67.

Nitric oxide releasing nanoparticles are therapeutic for *Staphylococcus aureu*s abscesses in a murine model of infection. PLoS ONE. 2009. 4(11): e7804.

Cryptococcus neoformans responds to mannitol by increasing capsule size in vitro and in vivo. Cellular Microbiology, 2010. Jun;12(6):740-53.

DR. LIISE-ANNE PIROFSKI

The focus of the Pirofski laboratory is on antibody and B cell immunity to encapsulated microbes using *Streptococcus pneumoniae* (Pneumococcus) and *Cryptococcus neoformans* (Cryptococcus) as examples. Both of these microbes cause disease in normal and immunocompromised people, particularly those with HIV infection, AIDS, B cell and antibody defects. The laboratory conducts translational studies of the serological, cellular, and molecular response to these microbes in normal and immunocompromised patients and basic scientific studies of microbial pathogenesis and host-microbe interaction. The goals of this research are to understand how innate and acquired antibody and B cell immunity to these microbes confers resistance to disease and to translate this knowledge into novel approaches to treatment and prevention of pneumococcal and cryptococcal disease.

PUBLICATIONS

Yano M, Coleman JR, Gohil S, Manix C, **Pirofski L**. Antibodies to *Streptococcus pneumoniae* capsular polysaccharide enhance quorum sensing. MBio. Nov 1;2(5). pii: e00176-11. doi: 10.1128/mBio.00176-11, 2011.

Weber S. Tian H, van RN, **Pirofski L**. A serotype 3 pneumococcal capsular polysaccharide-specific monoclonal antibody requires FcgammaRIII and macrophages to mediate protection against pneumococcal pneumonia in mice. Infect Immun. 80:1314-1322, 2012. *Cover article*.

Rohatgi S, **Pirofski L**. Molecular Characterization of the early B cell response to pulmonary *Cryptococcus neoformans* infection. J Immunol. 189:5820-5830, 2012.

Symczak W, Davis MJ, Lundy SK, Dufaud C, Olsewski M, **Pirofski L**. X-linked immunodeficient mice exhibit enhanced susceptibility to *Cryptococcus neoformans* infection. mBio. Jul 2;4(4). pii: e00265-13. doi: 10.1128/mBio.00265-13, 2013.

Rohatgi S, Gohil S, Kuniholm M, Schultz H, Dufaud C, Armour K, Badri S, Mailliard R, **Pirofski L**. Fc gamma receptor 3A polymorphism and risk for HIV-associated cryptococcal disease. mBio. Aug 27;4(5). pii: e00573-13. doi: 10.1128/mBio.00573-13, 2013.

Khan MN, Coleman JR, Vernatter J, Varshney AK, Dufaud C, **Pirofski L**. An ahemolytic pneumolysin of *Streptococcus pneumoniae* manipulates human innate and CD4+ T cell responses and reduces resistance to colonization in mice in a serotype-independent manner. J Infect Dis, 210: 1658-1669. 2014.

DR. STEVEN PORCELLI

Improving T cell responses for vaccination and disease prevention

Our laboratory studies the control of acquired immune responses by T cells, which we view as the master regulators and key effectors of host defense and immune tolerance. In broad terms, our research can be divided into two interrelated areas. The first is to understand the role of regulatory T cells, with particular emphasis on the activities of a specialized T cell subset known as CD1d-restricted NKT cells. These T cells have the highly unusual property of responding to specific glycolipid antigens, and we are studying ways to control their regulatory and effector functions in various mouse models of disease. A second second major research area is the study of T cell responses against pathogenic microorganisms, especially *Mycobacterium tuberculosis.* We have recently made significant progress in understanding how mycobacteria block effective host T cell responses, and we are now working to incorporate our findings into the rational and intelligent design of a new tuberculosis vaccine. In the short term, we hope to broaden our understanding of how organisms like *M. tuberculosis* successfully evade eradication by the immune system. Our major long term goal is to create a genetically or chemically modified live attenuated *M. tuberculosis* strain that will be safe and effective as a vaccine against tuberculosis. The laboratory has also recently developed interests in novel cancer vaccines, tumor-specific immunotherapies and immunity against emerging viral threats such as Ebola and Zika viruses, and is pursuing several projects in these areas.

PUBLICATIONS

Im JS, Arora P, Bricard G, Molano A, Venkataswamy MM, Baine I, Jerud ES, Goldberg MF, Yu KOA, Ndonye RM, Howell AR, Yuan W, Cresswell P, Chang YT, Illarionov PA, Besra GS and **Porcelli SA**. Kinetics and cellular site of glycolipid loading control the outcome of Natural Killer T cell activation. Immunity 30:888-98 (2009).

Sweeney KA, Dao DN, Hsu T, Ramachandra L, Henao-Tamayo M, Ordway D, Jain P, Chen B, Chen M, Kim J, Harding CV, Orme I, Chan J, **Porcelli SA**, Jacobs WR Jr. A recombinant *Mycobacterium smegmatis* that elicits bactericidal immunity against *M. tuberculosis*. Nature Med. 17:1261-8 (2011).

Singh M, Quispe-Tintaya W, Chandra D, Jahangir A, Venkataswamy MM, Ng TW, Sharma S, Carreño LJ, **Porcelli SA***, Gravekamp C*. (*Joint corresponding authors). Direct incorporation of the NKT cell activator α-galactosylceramide improves efficacy and safety of a recombinant Listeria monocytogenes breast cancer vaccine. Br J Cancer. 111:1945-54 (2014).

Arora P, Baena A, Yu KO, Saini NK, Kharkwal SS, Goldberg MF, Kunnath-Velayudhan S, Carreño LJ, Venkataswamy MM, Kim J, Lazar-Molnar E, Lauvau G, Chang YT, Liu Z, Bittman R, Al-Shamkhani A, Cox LR, Jervis PJ, Veerapen N, Besra GS, **Porcelli SA**. A single subset of dendritic cells controls the cytokine bias of natural killer T cell responses to diverse glycolipid antigens. Immunity. 40:105-16 (2014).

Saini NK, Baena A, Ng TW, Venkataswamy MM, Kennedy SC, Kunnath-Velayudhan S, Carreño L, Xu J, Chan J, Larsen MH, Jacobs Jr. **WR, Porcelli SA**. Suppression of autophagy and antigen presentation by Mycobacterium tuberculosis PE_PGRS47.
Nat Microbiol (9):16133 (2016). PMID: 27562263

DR. VINAYAKA PRASAD

Research in our laboratory is focused on three areas of HIV/AIDS: HIV associated neurocognitive disorders (HAND), HIV replication mechanisms and RNA aptamers targeted to HIV.

HIV associated Neurocognitive Disorders (HAND): The severe form of HAND, the HIV associated dementia (HAD), is common among clade-B HIV-infected individuals in the US, but less common among individuals infected with clade-C HIV-1 in India, suggesting clade-specific differences in neuropathogenicity. Understanding clade-specific determinants of neuropathogenesis may shed light on the disease mechanism and help develop targeted drugs for HAD. We previously demonstrated that due to a C31S polymorphism, clade C Tat lacks the chemokine function of Clade B Tat that plays a crucial role in an increased brain infiltration of monocytic phagocytes in HAD. We studied neuropathogenesis induced by two HIV-1 clades B and C using SCID mouse HIV encephalitis (SCID-HIVE) model and reported that while the introduction of clade B HIV-1_{ADA} into SCID mouse brain recapitulates the key features of human HAD disease, mice exposed to similar inputs of $HIV_{Indie\text{-}C1}$ (clade C) made fewer memory errors than those exposed to HIV-1_{ADA} (clade B). HIV-1_{ADA} also caused greater astrogliosis and loss of neuronal network integrity.

Work from many groups has shown that clade C HIV-1 in Southern Africa can induce HAD at much higher incidence than in India. We hypothesized that such variation is due to polymorphism in the neuropathogenesis determinants in Tat or gp120, the two major neurotoxicity determinants of HAND. With respect to Tat, we observed that the percentage of HIV isolates with dicysteine motif in Tat is 2-3% on the Indian subcontinent while in the Southern African countries, they ranged from 19-26%. These data broadly correlate with the HAD frequencies reported from India, South Africa and Botswana (3-4%, 25% and 38% respectively). This finding has been corroborated using a Zambian HIV-1C isolate that displays a C31 residue and thus an intact dicysteine motif. Our in vitro and SCID-HIVE results clearly indicate that Tat dicysteine motif determines neurovirulence. If confirmed in population studies, it may be possible to predict neurocognitive outcomes of individuals infected with HIV-1C by genotyping Tat.

Since Tat is not the only neurovirulence determinant in HIV-1, we examined whether gp120 exhibits intra-clade differences between India and Southern Africa. Our findings indicate that gp120 can also display region-specific differences. For example, the Southern African HIV isolates appear to contain more robust neurovirulence determinants than those in the Indian isolates. Thus, two different viral genes in India appear to show determinants of low neurotoxicity. These results suggest that clinical studies studying the incidence of HAD or HAND to correlate viral genetic differences must examine both Tat and gp120. *Ongoing work in our laboratory is attempting to identify the neurovirulence signatures of gp120 in clade C and clade B virus isolates and exploring the role of exosomes in neurovirulence.*

Anti-HIV RNA aptamers hematopoietic gene therapy: We previously developed and tested the efficacy of novel, anti-HIV-1 RNA aptamers to inhibit HIV-1 replication. Aptamers are sequences isolated by the iterative process of SELEX and are highly specific to their targets. The most efficacious aptamers identified in our laboratory as well as combinations of them could be tested in nonhuman primates (macaques). We will introduce such aptamers into hematopoeitic stem cells, which will then be used in bone marrow transplantation followed by challenge with chimeric, pathogenic SHIVs. We have thoroughly characterized anti-RT aptamers and generated aptamers to HIV-1 Gag MA and NC proteins. Perturbation of HIV-1 Gag and viral RNA interaction using

anti-Gag aptamers has provided new insights showing that preventing Gag-RNA binding causes down-modulation of viral RNA thus inhibiting virus production. Most recently, we developed high affinity aptamers (Kd = 1nM) to HIV-1 protease. *We are currently characterizing the Nef aptamers to understand the specific Nef functions in HIV replication that are affected by each aptamer.*

HIV Replication Mechanisms: We have a long-standing interest in elucidating the mechanistic basis of key steps in HIV-1 replication. In earlier work, we delineated the determinants of polymerase processivity, fidelity of DNA synthesis and strand displacement synthesis by HIV-1 RT. *Our current work is focused on the role of beta chemokines in HIV-1 budding.*

PUBLICATIONS

HIV Neuropathogenesis

Dara, J., Dow, A., Cromwell, E., Sturdevant, C. B, Mallewa, M., Swanstrom, R., Van Rie, A. and Prasad, V. R. (2015) Multivariable analysis to determine if HIV-1 Tat dicysteine 1 motif is associated with neurodevelopmental delay in HIV-infected children in Malawi. *Brain Beh. Funct.* **11**:38. PMCID: PMC4683967.

Rao, V., Neogi U, Eugenin E and Prasad, V. R. (2014) HIV-1 gp120 is a second determinant of decreased neurovirulence of Indian HIV-1C isolates compared to Southern African HIV-1C isolates. *PLoS One* **9**(9):e107074. doi: 10.1371/journal.pone.0107074. eCollection 2014. PMCID: PMC4154767.

Rao, V., Neogi, U., Talboom, J. S., Padilla, L., Rahman, M., Fritz-French, C, Gonzalez-Ramirez, Verma, A., Wood, C., Ruprecht, R. M., Ranga, U., Azim, T., Joska, John, Eugenin, T., Shet, A., Bimonte-Nelson, Tyor, W. R. and Prasad, V. R. (2013) Clade C HIV-1 isolates circulating in Southern Africa exhibit a greater frequency of dicysteine motif-containing Tat variants than those in Southeast Asia and cause increased neurovirulence. Retrovirology 10:61. DOI: 10.1186/1742-4690-10-61.

Rao, V. R., Sas, A., Eugenin, E., Siddappa, N. B., Bimonte-Nelson, H., Berman, J., Ranga, U., Tyor, W. R. and Prasad, V. R. (2008) HIV-1 clade-specific differences in the induction of neuropathogenesis. J. Neurosci. 28:10010-6.

Dash, P. K., Siddappa, N. B., Mangaiarkarasi, A., Anand, K. K., Padmanabhan, R. B, Mahendarkar, A. V., Mahadevan, A., Satishchandra, P., Shankar, S. K., Prasad, V. R., Ranga, U. (2008) Expanded coreceptor use of Human Immunodeficiency Virus Type-1 subtype C molecular isolates from an Indian subject with HIV-1 associated dementia. Retrovirology 5:25

Ranga, U., Shankarappa, R., Siddappa, N. B., Lakshmi, R., Nagendran, R., Mahalingam, M., Mahadevan, A., Jayasuryan, N., Satishchandra, P., Shankar, S.K. and Prasad, V. R. (2004)Tat protein of HIV-1subtype-C viruses is a defective chemokine. J. Virol. 78:2586-2590.

RNA aptamers targeted to HIV-1

Duclair, S., Gautam, A., Ellington, A. and Prasad, V. R. (2014) High affinity RNA aptamers against HIV-1 protease inhibit both in vitro protease activity and late events of viral replication. *Mol. Ther. Nuc. Acids*. **4**:e228. doi: 10.1038/mtna.2015.1.

Ramalingam, D, Duclair, S., Datta, S., Ellington, A.D., Rein, A. and Prasad, V. R. (2010) RNA aptamers directed to HIV-1 Gag bind to the matrix and nucleocapsid domains and inhibit virus production. J. Virol. 85:305-314.

Joshi. P., North, T. W. and Prasad, V. R. (2005) Aptamers Directed to HIV-1 Reverse Transcriptase Display Greater Efficacy over Small Hairpin RNAs Targeted to Viral RNA in Blocking HIV-1 Replication. Mol. Ther. 11:677-686.

HIV-1 Replication Mechanisms

Lwatula, C., Garforth, SJ and Prasad, VR (2012) Lys66 as a determinant of high mismatch extension and misinsertion rates of HIV-1 reverse transcriptase FEBS J 2012 Aug 28. doi:10.1111/j.1742-4568.2012.08807.x.[Epub ahead of print]

Garforth, S. J., Domaoal, R. A., Lwatula, C., Landau, M. J., Meyer, A. J., Anderson, K. S. and Vinayaka Prasad (2010) K65R and K65A substitutions in HIV-1 reverse transcriptase enhance polymerase fidelity by decreasing both dNTP misinsertion and mispaired primer extension efficiencies. J. Mol. Biol. 401:33-44.

Garforth, S.,Parniak, M. A. and Prasad, V. R. (2008) Utilization of deoxynucleoside diphosphates as substrate by a DNA polymerase. Plos One. 3(4):e2074.

Garforth, S., Lee, T. W., Parniak, M. A., Kool, E. T. and Prasad, V. R. (2007) Site-directed mutagenesis in the fingers subdomain of HIV-1 reverse transcriptase reveals a specific role for the b3- b4 hairpin loop in dNTP selection. J. Mol. Biol. 365:38-49

DR. CHAIM PUTTERMAN

Systemic lupus erythematosus (SLE) is a prototypical autoimmune disease that typically affects women in their reproductive years. Involvement of the kidneys, or lupus nephritis, appears in about 50% of lupus patients during the course of their disease. Despite medical treatment, morbidity and mortality from renal disease are common in lupus patients. The overall increase in the incidence of lupus and in the number of deaths from the disease reported in the United States are additional reasons for significant concern.

Anti-double stranded (ds) DNA antibodies are a serologic hallmark of patients with SLE. In recent years it has been increasingly clear that not only are anti-dsDNA antibodies an important diagnostic marker for lupus, but that these autoantibodies are also instrumental in the pathogenesis of lupus nephritis. The mechanisms by which anti-dsDNA antibodies induce renal injury, however, are not completely understood. It has been suggested that anti-dsDNA antibodies bind DNA in the circulation followed by non-specific deposition of these immune complexes in the kidney, or that in-situ immune complexes are formed in the kidney by binding of anti-dsDNA antibodies to nuclear antigens deposited on the glomerular basement membrane. Alternatively, some anti-dsDNA antibodies may cause injury by penetrating into living cells and affecting unidentified metabolic pathways. Finally, we and others have generated evidence that strongly suggests that at least some anti-dsDNA antibodies are pathogenic not by virtue of their affinity for DNA, but rather by direct cross-reactivity with renal antigen.

The long-term goals of the laboratory are to study the antigenic triggers and renal pathogenicity of anti-dsDNA antibodies. We want to understand which antigen(s) can trigger pathogenic anti-dsDNA antibodies, and whether protein antigens can induce a lupus like anti-DNA response. We are determining the cross-reactive kidney antigen bound by anti-DNA antibodies in human lupus and in mouse models of the disease to understand what determines the nephritogenic potential of these antibodies. Understanding the renal pathogenicity of cross-reactive anti-dsDNA antibodies by identifying the target antigen for these antibodies in the kidney would improve our understanding of a key manifestation of lupus, and would facilitate the development of serological tools to better predict the onset and severity of renal involvement in patients with SLE. Furthermore, identification of the triggering and/or target antigen in lupus will allow us to develop novel approaches to the treatment of lupus, by blocking the effects of anti-DNA antibodies on target organs or by specifically tolerizing pathogenic B cells.

PUBLICATIONS

Zhao Z, Weinstein E, Tuzova M, Davidson A, Mundel P, Marambio P, **Putterman C**. Human lupus anti-DNA/anti-α-actinin antibodies bind to mesangial cells and induce renal disease. Arthritis Rheum 52:522-530, 2005.

Li Q, Xie C, Mackay M, Aranow C, **Putterman C**, Mohan C. Glomerular proteome assays: A novelapproach to study glomerular-reactive autoantibodies in lupus. J Clin Invest 115:3428-3439, 2005.

Campbell S, Burkly LC, Gao HX, Berman JW, Su L, Browning B, Zheng T, Schiffer L, Michaelson JS, **Putterman C**. Pro-inflammatory effects of TWEAK/Fn14 interactions in glomerular mesangial cells. J Immunol 176: 1889–1898, 2006.

Qing X, Zavadil J, Crosby M, Gilkeson G, Bottinger E, **Putterman C**. Nephritogenic anti-DNA antibodies regulate gene expression in glomerular mesangial cells: A possible mechanism for renal pathogenicity in lupus nephritis. Arthritis Rheum 54:2198-2210, 2006.

Zhao Z, Deocharan B, Scherrer, PE, Ozelius LJ, **Putterman C.** Target antigen localization and expression as determinants of the renal pathogenicity of anti-DNA antibodies: The role of α-actinin. J Immunol 176:7704-7714, 2006.

Pitashny M, Schwartz N, Qing X, Hojaili B, Aranow C, Mackay M, **Putterman C**. Urinary Lipocalin-2 as a potential biomarker for human lupus nephritis. Arthritis Rheum 56:1894-1903, 2007.

Deocharan B, Zhou Z, Antar K, Siconolfi-Baez L, Angeletti RH, Hardin J, **Putterman C**. α-actinin immunization elicits anti-chromatin autoimmunity in non-autoimmune mice. J Immunol 179:1313-1321, 2007.

Wu T, Xie C, Wang HW, Zhou XJ, Schwartz N, Mackay M, Aranow C, **Putterman C**, Mohan C. Elevated urinary VCAM-1, P-selectin, sTNFR-1, and CXCL16 in multiple lupus strains and human lupus nephritis. J Immunol 179:7166-7175, 2007.

Zhao Z, Burkly LC, Campbell S, Schwartz N, Molano A, Choudhury A, Eisenberg R, Michaleson JS, **Putterman C**. Blocking TWEAK/Fn14 interactions improves nephritis in the chronic graft versus host (cGVH) model of SLE. J Immunol 179:7949-7958, 2007.

Molano A, Illarionov, PA, Besra GS, **Putterman C**, Porcelli SA. Modulation of invariant natural killer T cell cytokine responses by indoleamine 2,3-dioxygenase. Immunology Lett 117:81-90, 2008.

Qing X, Pitashny M, Thomas DB, Barrat FJ, Hogarth M, **Putterman C**. Pathogenic anti-DNA antibodies modulate gene expression in mesangial cells: Involvement of HMGB1 in anti-DNA antibody induced renal injury. Immunology Lett 121:61-73, 2008.

Gao HX, Campbell SR, Burkly LC, Jakubowski A, Jarchum I, Banas B, Saleem M, Mathieson PW, Berman JW, Michaelson JS, **Putterman C**. TNF-like Weak Inducer of Apoptosis (TWEAK) induces potent inflammatory and proliferative effects in human kidney cells. Cytokine 46:24-35, 2009.

Gao HX, Campbell SR, Cui MH, Zong P, Hwang JH, Gulinello M, **Putterman C**. Depression is an early disease manifestation in lupus-prone MRL/lpr mice. J Neuroimmunol 207:45-56, 2009.

Schwartz N, Rubinstein T, Burkly LC, Collins CE, Su L, Hojaili B, Mackay M, Aranow C, Stohl W, Rovin BH, Michaelson JS, **Putterman C**. Urinary TWEAK as a biomarker of lupus nephritis. Arthritis Research Therapy 11:R143, 2009 (doi:10. 1186/ar2816)

Molano A, Lakhani P, Burkly LC, Michaelson JS, **Putterman C**. TWEAK stimulation of resident kidney cells in the pathogenesis of graft versus host induced lupus nephritis. Immunology Letters 125:119-128, 2009.

Rubinstein T, Pitashny M, Levine B, Schwartz N, Schwartzman J, Weinstein E, Pego-Regiosa JM, Lu TYT, Isenberg D, Rahman A, **Putterman C**. Urinary neutrophil gelatinase associated lipocalin (NGAL) as a novel biomarker for disease activity in lupus nephritis. Rheumatology (Oxford) 49:960-971 (doi:10.1093/rheumatology/kep468), 2010.

Gao HX, Sanders E, Tieng AT, **Putterman C**. Sex and autoantibody titers determine the development of neuropsychiatric manifestations in lupus prone mice. Journal of Neuroimmunology 229:112-122, 2010.

DR. XINGXING ZANG

(Immune Checkpoint and Immunotherapy lab: http://www.einstein.yu.edu/zang)

Our laboratory focuses on new pathways and immunptherapies of T cell costimulation and coinhbition. We have recently discovered new members of the T cell costimulatory/coinhibitory B7 family and CD28 family, including B7x, HHLA2 and TMIGD2, and are using a variety of experimental approaches (gene knock-out mice, transgenic mice, monoclonal antibodies, crystal structure, etc) to understand how new B7/CD28 family members regulate T cell activation and tolerance. Current emphasis in the lab is placed in the following areas:

1) Novel drugs development: Translational medicine of T cell costimulation and coinhibition;
2) In vivo functions of new B7/CD28 pathways;
3) Human cancer-associated new B7/CD28 pathways and cancer immunotherapy;
4) New B7/CD28 pathways in autoimmune diseases/metabolic diseases and immunotherapy;
5) Relationship between new B7/CD28 pathways and infection;
6) Functional and structural characterization of new members of the Ig superfamily.

Our goal is to elucidate the mechanisms by which costimulation and coinhibition regulate T cells in peripheral non-lymphoid organs, and to translate the lessons learned in these studies towards developing new therapeutic strategies for immune-mediated diseases such as cancers, autoimmune disorders, metabolic diseases, infectious diseases, and transplantation rejection. Our research has won extensive attention from biopharmaceutical industry including some of the biggest drug companies.

Since 2008 the lab has mentored total 31 trainees of graduate students, postdoctoral fellows, clinical fellows, and visiting scientists. Many of trainees have subsequently developed independent careers in academic universities, medical centers, biopharmaceutical industry, and US government.

PUBLICATIONS

Janakiram M, Shah UA, Liu W, Zhao A, Schoenberg MP, **Zang X**. The third group of the B7-CD28 immune checkpoint family: HHLA2, TMIGD2, B7x and B7-H3. *Immunological Reviews*, 276:26-39, 2017

Ohaegbulam KC, Liu W, Jeon H, Almo SC, **Zang X**. Tumor-expressed immune checkpoint B7x promotes cancer progression and antigen-specific CD8 T cell exhaustion and suppressive innate immune cells. *Oncotarget*, in press

Sankin A, Narasimhulu D, John P, Gartrell B, Schoenberg M, **Zang X**. The expanding repertoire of targets for immune checkpoint inhibition in bladder cancer: what lies beneath the tip of the iceberg, PD-L1. *Urologic Oncology*, in press

Cheng H, Janakiram M, Borczuk A, Lin J, Qiu W, Liu H, Chinai JM, Halmos B, Perez-Soler R, Zang X. HHLA2, a new immune checkpoint member of the B7 family, is widely expressed in human lung cancer and associated with mutation and histology. *Clinical Cancer Research*, 23:825-832, 2017

Zhou TC, Sankin AI, Porcelli SA, Perlin DS, Schoenberg MP, **Zang X**. A review of the PD-1/PD-L1 checkpoint in bladder cancer: from mediator of immune escape to target for treatment. *Urologic Oncology*, 35:14-20, 2017

Picarda E, Ohaegbulam KC, **Zang X**. Molecular pathways: Targeting B7-H3 (CD276) for human cancer immunotherapy. *Clinical Cancer Research*, 22:3425-3431, 2016

Liu W, Almo SC, **Zang X**. Co-stimulate or co-inhibit regulatory T cells, which side to go? *Immunological Investigations*, 45:813-831, 2016

Janakiram M, Pareek V, Cheng H, Narasimhulu D, **Zang X**. Immune checkpoint blockade in human cancer therapy: Lung cancer and hematopoietic malignancy. *Immunotherapy*, 8:809-819, 2016

Koirala P, Roth M, Gill J, Chinai J, Ewart M, Piperdi S, Geller D, Hoang B, Fatakhova K, Ghorpade M, **Zang X***, Gorlick R*. HHLA2, a new immune checkpoint member of the B7 family, is highly expressed in human osteosarcoma and associated with metastases and worse survival. *Scientific Reports*, 6:31154. doi: 10.1038/srep31154, 2016

Yao Y, Ye H, Qi Z, Mo L, Yue Q, Baral A, Hoon DSB, Vera JC, Heiss JD, Chen CC, Hua W, Zhang J, Jin K, Wang Y, **Zang X***, Mao Y*, Zhou L*. B7-H4(B7x)-mediated cross-talk between glioma-initiating cells and macrophages via the IL-6/JAK/STAT3 pathway lead to poor prognosis in glioma patients. *Clinical Cancer Research*, 22:2778-2790, 2016

Assal A, Kaner J, Pendurti G, **Zang X**. Emerging targets in cancer immunotherapy: beyond CTLA-4 and PD-1. *Immunotherapy*, 7:1169-1186, 2015

Chinai JM, Janakiram M, Chen F, Chen W, Kaplan M, **Zang X.** New immunotherapies targeting the PD-1 pathway. *Trends in Pharmacological Sciences*, 36:587-595, 2015

Janakiram M, Chinai JM, Zhao A, Sparano JA, **Zang X**. HHLA2 and TMIGD2: New immunotherapeutic targets of the B7 and CD28 families. *OncoImmunology*, 4: e1026534-1 - e1026534-3, 2015

Ohaegbulam KC, Assal A, Lazar-Molnar E, Yao Y, **Zang X**. Human cancer immunotherapy with antibodies to the PD-1 and PD-L1 pathway. *Trends in Molecular Medicine*, 21:24-33, 2015

Janakiram M, Chinai JM, Fineberg S, Fiser A, Montagna C, Medaverepu R, Castano E, Jeon H, Ohaegbulam KC, Zhao R, Zhao A, Almo SC, Sparano JA, **Zang X**. Expression, clinical significance, and receptor identification of the newest B7 family member HHLA2 protein. *Clinical Cancer Research*, 21:2359-2366, 2015

Jeon H, Vigdorovich V, Garrett-Thomson SC, Janakiram M, Ramagopal UA, Abadi YM, Lee JS, Scandiuzzi L, Ohaegbulam KC, Chinai JM, Zhao R, Yao Y, Mao Y, Sparano JA, Almo SC, **Zang X.** Structure and cancer immunotherapy of the B7 family member B7x. *Cell Reports*, 9:1089-1098, 2014

Scandiuzzi L, Ghosh K, Hofmeyer KA, Abadi YM, Lázár-Molnár E, Lin EY, Liu Q, Jeon H, Almo SC, Chen L, Nathenson SG, **Zang X**. Tissue-expressed B7-H1 (PD-L1) critically controls intestinal inflammation. *Cell Reports*, 6:625-632, 2014

Zang X. The B7 and CD28 families (book chapter). Encyclopedia of Medical Immunology. I.R. Mackay and N.R. Rose, eds. Springer, 174-181, 2014

Zhao R, Chinai JM, Buhl S, Scandiuzzi L, Ray A, Jeon H, Ohaegbulam KC, Ghosh K, Zhao A, Scharff MD, **Zang X**. HHLA2 is a member of the B7 family and inhibits human CD4 and CD8 T cell function. Proc Natl Acad Sci USA, 110: 9879-9884, 2013

Jeon H, Ohaegbulam KC, Abadi YM, **Zang X**. B7x and myeloid derived suppressor cells in the tumor microenvironment: A tale of two cities. OncoImmunology, e24744-1- e24744-3, 2013

Vigdorovich V, Ramagopal U, Lazar-Molnar E, Sylvestre E, Lee JS, Hofmeyer KA, **Zang X**, Nathenson SG, Almo SC. Structure and T cell inhibition properties of B7 family member, B7-H3. Structure, 21:707-717, 2013

Abadi YM, Jeon H, Ohaegbulam KC, Scandiuzzi L, Ghosh K, Hofmeyer KA, Lee JS, Ray A, Gravekamp C, **Zang X**. Host B7x promotes pulmonary metastasis of breast cancer. J Immunol, 190: 3806-3814, 2013

Janakiram M, Abadi YM, Sparano JA, **Zang X**. T cell coinhibition and immunotherapy in human breast cancer. Discov Med, 14:229-236, 2012

Lee JS, Scandiuzzi L, Ray A, Wei J, Hofmeyer KA, Abadi YM, Loke P, Lin J, Yuan J, Serreze DV, Allison JP, **Zang X.** B7x in the periphery abrogates pancreas specific damage mediated by self-reactive effector CD8 T cells. J Immunol, 189: 4165-4174, 2012

Hofmeyer KA, Scandiuzzi L, Ghosh K, Pirofski LA, **Zang X**. Tissue-expressed B7x affects the immune response to and outcome of lethal pulmonary infection. J Immunol, 189:3054-3063, 2012

Scandiuzzi L, Ghosh K, **Zang X.** T cell costimulation and coinhibition: Genetics and disease. Discov Med, 12:119-128, 2011

Hofmeyer KA, Jeon H, **Zang X.** The PD-1/PD-L1 (B7-H1) pathway in chronic infection-induced cytotoxic T lymphocyte exhaustion. J Biomed Biotechnol, 2011:451694, 2011

Wei J, Loke P, **Zang X***, Allison JP**.** Tissue-specific expression of B7x protects from CD4 T cell-mediated autoimmunity. J Exp Med, 208: 1683-1694, 2011

Barach YS, Lee JS, **Zang X**. T cell coinhibition in prostate cancer: new immune evasion pathways and emerging therapeutics. Trends Mol Med, 17: 47-55, 2011

Zang X, Sullivan PS, Soslow RA, Waitz R, Reuter VE, Wilton A, Thaler HT, Arul M, Slovin SF, Wei J, Spriggs DR, Dupont J, Allison JP. Tumor associated endothelial expression of B7-H3 predicts survival in ovarian carcinomas. Mod Pathol, 23: 1104-1112, 2010

Hofmeyer KA, Ray A, **Zang X**. The contrasting role of B7-H3 (Invited Commentary). Proc Natl Acad Sci USA, 105: 10277-10278, 2008

Thompson RH*, **Zang X***, Lohse CM, Leibovich BC, Slovin S, Reuter VE, Blute ML, Russo P, Kwon ED*, Allison JP*. Serum soluble B7x is elevated in renal cell carcinoma patients and is associated with advanced stage. Cancer Res*, 68: 6054-6058,* 2008.

Zang X, Allison JP. The B7 family and cancer therapy: Costimulation and coinhibition. Clin Cancer Res, 13: 5271-5279, 2007

PUBLICATIONS

MICROBIOLOGY & IMMUNOLOGY

STUDENT PUBLICATIONS

2001 – 2017

2001

Raveh, T., **Droguett, G., Horwitz, M.S.,** DePinho, R.A., and Kimchi, A. DAP-kinase Acivates a p19ARF/p53-mediated Apoptotic Checkpoint to Suppress Oncogenic Transformation. Nature Cell Biology 3:1-7 (2001).

Efrat, S., Serrreze, D., **Svetlanov, A.,** Post, C., C.M. Johnson, E.A. Herold, K., and **Horwitz, M.S.** Adenovirus early region 3 (E3) immunomodulatory genes decrease the incidence of autoimmune diabetes in nonobese diabetic (NOD) mice. Diabetes 50:980-984 (2001)

Reddy, B., Gupta, S., Chuzhin, Y., Kalergis, A., Budhai, L., Zhang, M., **Droguett, G. Horwitz, M.S.,** Roy Chowdhury, J., Nathenson, S.G. and Davidson A. The Effect of CD29/B7 Blockage on Alloreactive T and B Cells after liver cell transplantation. Transplantation 71:801-811 (2001).

Khalil, M., Inaba,K., Steinman, R., Ravetch, J. and **Diamond, B.** T cell in peptide induced model of systemic lupus erythematosus. J. Immunol. 166:166-1674 (2001)

Grimaldi, CM., **Michael, DJ.,** and **Diamond, B.** Expansion and activation of a population of autoreactive marginal zone B cells in a model of estrogen-induced lupus. Cutting Edge. J. Immunol. J. Immunol. 167:1886-1890 (2001).

Beger E, **Deocharan B,** Edelman M, Erblich B, Gu Y, **Putterman C.** A peptide DNA surrogate accelerates autoimmune manifestations and nephritis in lupus-prone mice. J Immunol (in revision) (2001)

Deocharan B, **Qing X,** Lichauco J, **Putterman C.** a-Actinin is a cross-reactive target for pathogenic anti-DNA antibodies in renal tissue. (submitted). (2001)

Davidowitz, E.J., Schoenfeld, A.R. and **Burk, R.D.** VHL induces renal cell differentiation and growth arrest through integration of cell-cell and cell-ECM signaling. Mol. Cell. Biol. 21:865-874, 2001.

Schoenfeld, A., **Davidowitz, E.J**. and **Burk, R.D.** Endoplasmic reticulum/cytosolic localization of von Hippel-Lindau (VHL) gene products is mediated by a 64 amino acid region. Int. J. Cancer 91:457-467, 2001.

Devarajan, P., De Leon, M.,Talasazan, F., Schoenfeld, A.R., **Davidowitz, E.J., and Burk, R.D.** The von Hippel-Lindau gene product inhibits renal cell apoptosis via Bcl-2-dependent pathways. J. Biol. Chem. 276:40599-40605, 2001.

Rezende, L. F., Yvonne Kew and **V. R. Prasad** (2001) The effect of increased processivity on overall fidelity and error specificity. *J. Biomed. Sci.* **8**:197-205.

Schwartz, J.C.D., Zhang, X., Federov, A.A., **Nathenson, S.G** and Almo, S.C. Structural Basis for Costimulation by the Human CTLA-4/B7-2 Complex. Nature, 2001, 410:604-608.

Zhang, X.**, Schwartz, J.C.D., Nathenson, S.G.,** and Almo, S.C. Crystallization and Preliminary X-ray Analysis of the Complex Between Human CTLA-4 and B7-2. Acta Crystallographica, 2001, 898-899.

Reddy, B., Gupta, S., Chuzhin, Y., Kalergis, A.M., **Budhai, L.**, Zhang, M.,Droguett, G., **Horwitz,** M.S., Chowdhury, J.R., Nathenson, S.G., and Davidson, A. The Effect of CD28/B7 Blockade on Alloreactive T and B Cells After Liver Cell Transplantation. Transplantation, 2001, 71:810-811.

Khalil, M., Inaba, K., Steinman, R., Ravetch, J. and **Diamond, B.** T cell in a peptide induced model of systemic lupus erythematosus. J. Immunol. 166:166-1674 (2001).

Grimaldi, CM., Michael, DJ., and **Diamond, B.** Expanison and activation of a population of autoreactive marginal zone B cells in a model of estrogen-induced lupus. Cutting Edge J. Immunol. 167:1886-1890 (2001).

Raveh, T., **Droguett, G., Horwitz,** M.S., DePinho, R.A., and Kimchi, A. DAP-kinase Activates a p19ARF/p53-mediated Apoptotic Checkpoint to Suppress Oncogenic Transformation . Nature Cell Biology 3:1-7 2001.

Efrat, S., Serreze, D., **Svetlanov, A.,** Post, C. M. Johnson, E. A. Herold, K., and **Horwitz,** M.S. Adenovirus early region 3 (E3) immunomodulatory genes decrease the incidence of autoimmune diabetes in nonobese diabetic (NOD) mice. Diabetes 50:980-984 (2001).

Reddy, B., Gupta, S., Chuzhin, Y., Kalergis, A., Budhai, L., Zhang, M., **Droguett, G., Horwitz**, **M.S.,** Roy Chowdhury, J., Nathenson, S.G., and Davidson, A. The Effect of CD29/B7 Blockage on Alloreactive T and B Cells after liver cell transplantation. Transplantation 71:801-811 (2001).

2002

Fisher, T. S. and **Prasad, V. R.** (2002) Mutations that confer resistance to template-analog inhibitors of human immunodeficiency virus type 1 (HIV-1) reverse transcriptase also cause severe defects in HIV replication *J. Virol.* 76:4068-4072.

Fisher, T. S. and **Prasad, V. R.** (2002) Substitutions of Phe61 located in the vicinity of template overhang influence polymerase fidelity and nucleoside analog sensitivity of HIV-1 Reverse Transcriptase J. Biol. Chem. 277:22345-22352.

Ostrov, D.A., **Roden M.M., Shi**, W. Palmieri, E., Christianson G.J., Mendoza,L., Villaflor G., Tilley, D., Shastri, N., Grey, H., Almo, S.C., Roopenian, D., and **Nathenson, S.G.** How H13 Histocompatibility Peptides Differing by a Single Methyl Group and Lacking Conventional MHC Binding Anchor Motifs Determine Self-Nonself Discrimination. J. Immunol., 2002, 168: 283-289.

Schwartz, J.-C.D., Zhang, X., **Nathenson, S.G.** and Almo, S.C. Structural Mechanisms of Costimulation. Nature Immunology, 2002, 3: 427-434.

Zhang, X., **Schwartz, J.-C.D**., Almo, S.C. and **Nathenson, S.G.** Expression, Refolding, Purification, Molecular Characterization, Crystallization and Preliminary X-Ray Analysis of the Receptor Binding Domain of Human B7-2. Protein Expression and Purification, 2002, 105-113.

Chu, YP., Taylor, D., Yan, HG., **Diamond, B.**, and Spatz, L. Persistence of partially functional double stranded DNA binding B cells in mice transgenic for the IgM heavy chain of an anti-dsDNA antibody. Intl. Immunol. 14:45-54 (2002).

Harris, S., **Dagtas, AS.,** Chen, H., and **Diamond, B.** Regulating the isotypic and idiotypic profile of an anti-PC antibody response: lessons from peptide mimics. Infection and Immunity (2002).

Grimaldi, CM., Cleary, J., Dagtas, AS., Moussai, D., and **Diamond, B**. Estrogen alters thresholds for B cell apoptosis and activation. J. Clin. Invest. 109:1625-1633 (2002)

Friedman, J.M. and **Horwitz, M.S.** Inhibition of TNF-a Induced NF-kB Activation by the Adenovirus E3-10.4/14.5K Complex. J. Virol. 76:5515-5521 (2002).

Lesokhin, A.M., **Delgado-Lopez, F.** and **Horwitz, M.S.** Inhibition of chemokine transcription by adenovirus early region three (E3) genes. J. Virol. 76:8236-8243 (2002).

2003

Peeva, E., **Michael, D. ,** Cleary, J., Rice, J., Chen, X., and **Diamond,** B. Prolactin modulates the naïve B. Cell repertoire. *J. Clin. Invest. 111:275-283.* (2003).

Wang, C., Khalil, M., Ravetch, J., and **Diamond, B.** The naïve B cell repertoire predisposes to antigen-induced systemic lupus erythematosus. *J. Immunol.* 170:4826-4832 (2003).

Lieberman, S.M., Evans, A.M., Han, B., Takaki, T., Vinnitskaya, Y., Caldwell, J.A., Serreze, D.V. Sshabanowitz, J., Hunt, D.F., **Nathenson, S. G.,** Santamaria, P., DiLorenzo, T. P. Identification of the Novel β Cell Antigen Targeted By a Prevalent Population of Pathogenic CD8+ T Cells in Autoimmine Diabetes, *Proc. Nat'l Acad. Sci. USA, 2003, 100:8384-8388.*

2004

Fessler, S. (Horwitz Lab) Inhibition of TNF Signal Transduction by the Adenovirus Group C RID complex involves downregulation of surface levels of TNFR1 (*in press*). J.Virol. (2004).

Takaki, T., SM. Lieberman, T.M. Holl, B. Han, P. Santamaria, D.V. Serreze, and **T. P. DiLorenzo. 2004.** Requirement for both H-2D^{b} and H-2K^{d} for the induction of diabetes by the promiscuous CD8^{+} T cell clonotype AI4. *J. Immunol.*.

Johnson, DJ, **Fidock*,** DA, Mungthin, M, **Lakshmanan, V., Sidhu, ABS, Bray**, PG, & Ward, SA* (2004). Evidence for a central role fro PfCRT in conferring *Plasmodium falciparum* resistance to diverse antimalarial agents. Molec. Cell . *Co-corresponding authors.

Shi, W., **Ting, L.M.,** Lewandowicz, A., Tyler, P.C., Evans, G.B., Furneaux, R.H., **Kim, K.,** Almo, S.C. and Schramm, V.L., *Plasmodium falciparum* purine nucleoside phosphorylase; Crystal structures, immucillin inhibitors and dual catalytic function. J Biol Chem. 279:18103-6. 2004.

Qing, X., Zavadil, J, Crosby, M. Gilkeson, G., Bottinger, E., **Putterman, C.** Nephritogenic anti DNA antibodies regulate gene expression in glomerular mesangial cells; A possible mechanism for renal pathogenicity in lupus nephritis. In revision, 2004.

2005

Diaz-Griffero F., Muehlbauer S., **Squires R.,** Litmann D. and **Brojatsch J.** (2005) Production of infectious HIV particles in murine cells.

Yu KOA, Im JS, Molano A, Dutronc Y, Illarionov PA, Forestier C, Fujiwara N, Arias I, Miyake S, Yamamura T, Chang Y-T, Besra GS, **Porcelli SA**. Modulation of CD1d-restricted NKT cell responses using N-acyl variants of α−galactosylceramides. Proc. Nat. Acad. Sci. (USA), 102:3383-8 (2005).

2006

Muehlbauer S. and **Brojatsch J.** (2006) Anthrax lethal toxin-mediated cell death and disease progression.

Alileche, A., **Squires, R.**, **Muehlbauer, S.**, Hoschander, A., and **Brojatsch J.** (2006) Mitochondrial impairment mediates cytolysis in anthrax lethal toxin-treated murine macrophages. *Cell Cycle 5(1):c1.*

Wang, F., A. Nakouzi, **M. Alvarez**, O. Zaragoza, R. H. Angeletti, and **A. Casadevall.** 2006. Structural and functional characterization of glycosylation in an immunoglobulin G1 to Cryptococcus neoformans glucuronoxylomannan. Mol Immunol 43:987-98.

Martinez, L. R., R. A. Bryan, C. Apostolidis, A. Morgenstern, **A. Casadevall,** and E. Dadachova. 2006. Antibody-guided alpha radiation effectively damages fungal biofilms. Antimicrob Agents Chemother 50:2132-6. Valderramos, S.G., and D.A. Fidock. 2006. Transporters involved in resistance to antimalarial drugs. *Trends Pharmacol. Sci.*.

Nkrumah LN, **Muhle RA, Moura PA,** Ghosh P, Hatfull G, Jacobs WR Jr, & **Fidock DA** (2006). Efficient site-specific integration in *Plasmodium falciparum* chromosomes mediated by mycobacteriophage Bxb1 integrase. *Nature Methods* 3:615-621.

Sidhu ABS, Uhlemann A-C, **Valderramos SG,** Valderramos J-C, Krishna S & **Fidock DA** (2006). Decreasing *pfmdr1* copy number in *Plasmodium falciparum* malaria heightens susceptibility to mefloquine, lumefantrine, halofantrine, quinine, and artemisinin. *J. Infect. Dis.* 194:528-535.

Lakshmanan V, Bray PG, Verdier-Pinard D, Johnson DJ, Horrocks P, **Muhle RA,** Alakpa GE, Hughes RH, Ward SA, Krogstad DJ, Sidhu ABS & **Fidock DA** (2005). A critical role for PfCRT K76T in *Plasmodium falciparum* verapamil-reversible chloroquine resistance**. *EMBO J.*** 24: 2294-305.

Sidhu ABS, **Valderramos SG** & Fidock DA (2005). *pfmdr1* mutations contribute to quinine resistance and enhance mefloquine and artemisinin sensitivity in *Plasmodium falciparum*. *Mol. Microbiol.* 57: 913-26.

Cao, E., Ramagopal, U., Federov, A., Fedorov, L., Yan, Q., Lary, J., Cole, J., **Nathenson, S.G.**, and Almo, S.C. NTB-A crystal structure: homophilic interactions and signaling by the SLAM family of receptors, Immunity, 2006.

Campbell S, Burkly LC, Gao HX, Berman JW, Su L, Browning B, Zheng T, Schiffer L, Michaelson JS, **Putterman C**. Pro-inflammatory effects of TWEAK/Fn14 interactions in glomerular mesangial cells. J Immunol 176: 1889–1898, 2006.

Qing X, Zavadil J, Crosby M, Gilkeson G, Bottinger E, **Putterman C**. Nephritogenic anti-DNA antibodies regulate gene expression in glomerular mesangial cells: A possible mechanism for renal pathogenicity in lupus nephritis. Arthritis Rheum 54:2198-2210, 2006.

Zhao Z, **Deocharan B**, Scherrer, PE, Ozelius LJ, **Putterman C.** Target antigen localization and expression as determinants of the renal pathogenicity of anti-DNA antibodies: The role of a-actinin. J Immunol 176:7704-7714, 2006. **Deocharan B**, Marambio P, Edelman M, **Putterman C**. Differential effects of interleukin-4 in peptide induced autoimmunity. Clin Immunol 108:80-88, 2003.

Campbell S, Burkly LC, Gao HX, Berman JW, Su L, Browning B, Zheng T, Schiffer L, Michaelson JS, **Putterman C**. Pro-inflammatory effects of TWEAK/Fn14 interactions in glomerular mesangial cells. J Immunol 176: 1889–1898, 2006.

Peeva, E., Gonzalez, J., **Hicks, R.** and **Diamond, B.** Lupus susceptibility Interval *Sle3/5* confers responsiveness to prolactin in C57Bl6 mice. J. Immunol. 177:1401-1405 (2006).

Zhao Z, **Deocharan B**, Scherrer, PE, Ozelius LJ, **Putterman C.** Target antigen localization and expression as determinants of the renal pathogenicity of anti-DNA antibodies: The role of α-actinin. J Immunol 176:7704-7714, 2006.

2007

Alileche A., **Squires R.**, **Muehlbauer S.** and **J. Brojatsch.** (2006) Mitochondrial impairment mediates cytolysis in anthrax lethal toxin-treated murine macrophages. *Cell Cycle* 5:c1.

Alileche A., **Serfass E**., **Muehlbauer S**., Porcelli S. and **J. Brojatsch.** (2005) Anthrax lethal toxin-mediated killing of dendritic cells impairs the adaptive immune response. *PLoS Pathogens* 1:e19.

Akoachere M., **Squires R**., Nour A., Angelov L., **Brojatsch J.** and E. Abel-Santos. (2007) Identification of an *in vivo* inhibitor of *Bacillus anthracis* spore germination. *J. Biol. Chem.* 282:12112-8.

Muehlbauer S., Evering T., Bonuccelli G., **Squires, R.**, Anthony W. Ashton, Porcelli S., Lisanti MP. and **J. Brojatsch.** (2007) Anthrax lethal toxin kills macrophages in a strain-specific manner by apoptosis or caspase-1-mediated necrosis. *Cell Cycle* 6:758-66.

Squires R., **Muehlbauer S**. and **J. Brojatsch.** (2007) Proteasomes control caspase-1-mediated killing of murine macrophage by anthrax lethal toxin. *J. Biol. Chem.* Accepted.

Jarchum, I., J. C. Baker, T. Yamada, T. Takaki, M. P. Marron, D. V. Serreze, and **T. P. DiLorenzo**. 2007. *In vivo* cytotoxicity of insulin-specific $CD8^+$ T cells in HLA-A*0201-transgenic NOD mice. Diabetes

Li Q, Ndonye RM, Illarionov PA, **Yu KO**, **Jerud ES**, Diaz K, Bricard G, **Porcelli SA**, Besra GS, Chang YT, Howell AR. Rapid Identification of Immunostimulatory α-Galactosylceramides Using Synthetic Combinatorial Libraries. J Combin Chem, (2007).

Yu KOA, Im JS, Illarianov PA, Ndonye RM, Howell AR, Besra GS, **Porcelli SA**. Production and characterization of monoclonal antibodies against complexes of the NKT cell ligand α-galactosylceramide bound to mouse CD1d. J Immunol Methods. 323:11-23 (2007).

Hinchey J. Lee S, Manjunatha V, Chen B, Basaraba RJ, Jeon BY, Derrick SC, Chan J, Braunstein M, Orme IM, Morris SL, Jacobs, Jr. WR, **Porcelli SA**. Enhanced Priming of Adaptive Immunity by a Proapoptotic Mutant of Mycobacterium tuberculosis. J. Clin. Invest. 117 :2279-2288 (2007).

Forestier C, Takaki T, Molano A, Im JS, Baine I, **Jerud ES**, Illarionov P, Ndonye R, Howell AR, Santamaria P, Besra GS, DiLorenzo TP, **Porcelli SA**. Improved outcomes in NOD mice treated with a novel Th2 cytokine biasing NKT cell activator. J Immunology, 178:1415-25 (2007).

2008

Adarichev V, Szanto S, Vegvari A, Szabo Z, Kis-Toth K, Mikecz K, Glant TT. Two loci on chromosome 15 control experimentally-induced arthritis through the differential regulation of IL-6 and lymphocyte proliferation *J. Immunol.* 181:1307-1314. 2008

Adarichev VA, Vegvari A, Szabo Z, Kis-Toth K, Mikecz K, Glant TT. Congenic Strains Displaying Similar Clinical Phenotype of Arthritis Represent Different Immunologic Models of Inflammation. *Genes Immun.*. 2008**.**

Strickler, H.D., Palefsky, J.M. and **Burk, R.D.** HPV types present in invasive cervical cancers of HIV-seropositive women. Int. J. Cancer 123:1224-1225, 2008.

Haddad, R., Crum, C., Chen, Z., Krane, J., Posner, M., Li, Y. and **Burk, R.** HPV16 transmission between a couple with HPV-related head and neck cancer. Oral Oncol, 2008.

Lutz, M.S. and **Burk, R.D.** Primary cilium formation requires von Hippel-Lindau gene function in renal derived cells. Cancer Res. 66:6903-6907, 2006. (Highlighted article in July 15, 2006 Cancer Res.)

De Jesus M; Park CG, Su Y; Goldman D; Steinman RM; **Casadevall A** (2008) Spleen deposition of Cryptococcus neoformans capsular Glucuronoxylomannan in rodents occurs in red pulp macrophages and not marginal zone macrophages expressing the C-type lectin SIGN-R1. Med. Mycol , 2008.

Torres M, Fernandes-Fuentes M, Fiser A, **Casadevall A** (2008) Exchaning murine and human constant chains affects the kinetics and thermodynamics of antigen binding and chimeric antibody autoreactivity. PLOS 2:e1310.

Frases S, Nimrichter L, Viana NB, Nakouzi A, **Casadevall (2008)** Capsular and exopolysaccharide fractions manifest physical, chemical, and antigenic differences. Eukaryot. Cell, 2008.

Maglione PJ, Xu J, Casadevall A, **Chan J.** 2008. Fcγ receptors regulate immune activation and susceptibility during *M. tuberculosis* infection. J Immunol 180:3329-38.

Chakravarty SD, Zhu G, Tsai MC, Mohan VP, Marina S, Kirschner DE, Huang L, Flynn J, **Chan J.** 2008. Tumor necrosis factor blockade in chronic murine tuberculosis enhances granulomatous inflammation and disorganizes granulomas in the lungs. Infect Immun 76:916-26.

Ray JCJ, Wang J, **Chan J,** Kirschner DE. 2008. The timing of TNF and IFN-□ signaling affects macrophage activation strategies during *Mycobacterium tuberculosis* infection. J Theor Biol 252:24-8.

Dao DN, Sweeney K, Hsu T, Gurcha, SS, Nascimento IP, Roshevsky D, Besra GS, **Chan J,** Porcelli S, Jacobs WR Jr. 2008. Mycolic acids of trehalose dimycolate modified by MmaA4 repress IL-12p40 production. Plos Pathogens 4(6):e1000081.

Russell E, Xu J, Wang X, **Chan J,** Tufariello IM. 2008. A Double Rpf knockout *Mycobaterium tuberculosis* strain with altered cell wall composition exhibits profound defects in reactivation from chronic tuberculosis and innate immunity phenotypes. Infect Immun, 2008.

Schiffer L, Bethunaickan R, Ramanujam M, Huang W, Schiffer M, Tao H, Madaio MM, Bottinger EP and **A Davidson**. Activated renal macrophages are markers of disease onset and disease remission in lupus nephritis J. Immunol 180: 1938-1947 2008.

Ramanujam M and **A Davidson.** Targeting of the immune system in systemic lupus Erythematosus. Exp. Rev. Mol. Med Jan 21;10:e2 2008.

Ramanujam M and **A Davidson.** BAFF blockade for SLE – will the promise be fulfilled? Immunological Reviews 223:156-74 2008.

Kahn P, Ramanujam M, Bethunaickan R, Huang W, Tao H, Madaio MP, Factor SM and **A Davidson.** Prevention of murine anti-phospholipid syndrome by BAFF blockade. Arthritis and Rheumatism (2008).

Wang, Y. and **Diamond, B**. Receptor revision diminishes the autoreactive B cell response after antigen activation. JCI 118:2896-2907 (2008) PMC2467385

Kim, SJ., Caton, M., Wang, C., Khalil, M., Zhou, ZJ., Hardin, J. and **Diamond, B.** Increased IL-12 inhibits B cells differentiation to germinal center cells and promotes differentiation to short lived plasmablasts. JExp Med (2008)

Chaparro, R. J., A. R. Burton, D. V. Serreze, D. A. A. Vignali, and **T. P. DiLorenzo**. 2008. Rapid identification of MHC class I-restricted antigens relevant to autoimmune diabetes using retrogenic T cells. *J. Immunol. Methods 335:106-115*.

Mukhopadhaya, A., T. Hanafusa, **I. Jarchum**, Y. G. Chen, Y. Iwai, D. V. Serreze, R. M. Steinman, K. V. Tarbell, and **T. P. DiLorenzo**. 2008. Selective delivery of cell antigen to dendritic cells *in vivo* leads to deletion and tolerance of autoreactive $CD8^+$ T cells in NOD mice. *Proc. Natl. Acad. Sci. USA 105:6374-6379.*

Antal, Z**., I. Jarchum**, and **T. P. DiLorenzo**. HLA class I supertypes in type 1 diabetic children in an urban children's hospital. Ann. NY Acad. Sci., 2008.

Jarchum, I., L. Nichol, M. Trucco, P. Santamaria, and **T. P. DiLorenzo**. 2008. Identification of novel IGRP epitopes targeted in type 1 diabetes patients. *Clin. Immunol. 127:359-365*.

Meertens L., C. Bertaux C, L. Cukierman, E. Cormier and **T. Dragic**. (2008) The tight junction proteins claudin-6 and -9 are entry cofactors for the Hepatitis C virus. *J. Virol*. 82: 3555

L. R. Martinez, D. C. Ibom, A. Casadevall, and **B.C. Fries** Characterization of phenotypic switching in Cryptococcus neoformans biofilm Mycopathologia Published online: 21 June 2008.

N. Jain, F. Hasan, and **B.C. Fries** Phenotypic switching in fungi Current Fungal Infection Reports (Volume 2 Issue)

N. Jain, and **B.C. Fries** Phenotypic switching in Cryptococcus neoformans and Cryptococcus gattii Mycopathologia Published online: 21 June 2008.

A. Guerrero and **B.C. Fries** Phenotypic switching in Cryptococcus neoformans contributes to virulence by changing the immunological host response Infect and Immun ,2008.

A. Joseph, J. Zheng, A. Follenzi, T. DiLorenzo, **K. Sango**, J. Hyman, K. Chen, E. Hooijberg, D. A. Vignali, B. D. Walker and **H. Goldstein**. Transformation of nonreactive peripheral blood CD8 T lymphocytes into HIV-specific CTL with potent in vivo inhibitory activity after lentiviral vector delivery of HIV-specific TCR genes. *J. Virology* 2008;82: 3078–3089.

Z. Tallóczy, J. Martinez, D. Joset, Y. Ray, A. Gácser, S. Toussi, N. Mizushima, J. Nosanchuk, **H. Goldstein,** J. Loike, D. Sulzer, and L. Santambrogio. Methamphetamine inhibits antigen processing, presentation, and phagocytosis, *PLoS Pathogens* 2008;4:e28

Z. Tallóczy, J. Martinez, D. Joset, Y. Ray, A. Gácser, S. Toussi, N. Mizushima, J. Nosanchuk, **H. Goldstein,** J. Loike, D. Sulzer, and L. Santambrogio. Methamphetamine inhibits antigen processing, presentation, and phagocytosis, *PLoS Pathogens* 2008;4:e28

H. Goldstein. NIH/NIAID New Humanized Rodent Models 2007 Workshop, *AIDS Research and Therapy,* 2008 ;5:3.

J. Sun, J. H. Zheng, M. Zhao, **Sunhee Lee** and **H. Goldstein**. Increased in vivo activation of microglia and astrocytes in the brains of mice transgenic for an infectious R5 HIV-1 provirus and for CD4-specific expression of human cyclin T1 in response to LPS stimulation. *J.Virology,* 2008;82:5562-5572.

H. Wang, J. Sun, and **H. Goldstein**. HIV-1 infection increases the in vivo capacity of peripheral monocytes to cross the blood brain barrier into the brain and the in vivo sensitivity of the blood brain barrier to disruption by LPS. *J.Virology,* 2008;82:7591-600.

Gravekamp C, Leal B, Denny A, Bahar R, Lampkin S, Castro F, Moore D, and Reddick R. In vivo responses to vaccination with Mage-b, GM-CSF and thioglycollate in a highly metastatic mouse breast tumor model, 4T1. Cancer Immunology and Immunotherapy 2008; 57: 1067-1077.

Kim SH, Castro F, Gonzalez D, Maciag P, Paterson Y, and **Gravekamp C**. Mage-b delivered by recombinant Listeria monocytogenes is highly effective against breast cancer metastases. British Journal of Cancer. 2008..

Gravekamp C, Kim SH, and Castro F. Cancer vaccination: manipulation of immune responses at old age. Mechanisms of Ageing and Development. 2008. (In Press). DOI: 10.1016/j.mad.2008.05.003.

Banaiee, N., January, V., Barthus, C., Lambrick, M., RoDiti, D., Behr, M.A., **Jacobs, W.R., Jr.,** Steyn, L.M. (2008) Evaluation of a Semi-Automated Reporter Phage Assay for Susceptibility Testing of *Mycobacterium tuberculosis* Isolates in South Africa. *Tuberculosis* **88**:64-68.

Ojha, A.K., Baughn, A.D., **Sambandan, D.,** Hsu, T., Trivellil, X., Guerardel, Y., Alahari, A., Kremer, L., **Jacobs, W.R., Jr.,** Hatfull, G.F**.** (2008) Growth of *Mycobacterium tuberculosis* biofilms containing free mycolic acids and harboring drug tolerant bacteria. *Mol. Microbiol.* [Epub]

Dao, D. N., **K. Lawrence**, T. Hsu, I. P. Nascimento, D. Roshevsky, S. S. Gurcha, G. S. Besra, J., Chan, S. A. Porcelli, and **Jacobs, W.R., Jr.** (2008) Repression of IL-12P40 is Controlled by MmaA4 Mediated Modification of Mycolic Acids Associated with Trehalose 6,6' Dimycolate. *PLoS Pathogens* **4**: e1000081.

Vilcheze, C., Av-Gay, Y., Attarian, R., Liu, Z., Hazbon, M.H., Colangeli, R., Chen, B., Liu, W., Alland, D., Sacchettini, J.C., **Jacobs, Jr., W.R.** (2008) Mycothiol biosynthesis is essential for ethionamide susceptibility in *Mycobacterium tuberculosis*.

Smith, ME, Cimica, V., Chinni, S., Challagulla, K., Mani, S. and **Kalpana, GV.** (2008) Inhibition of rhabdoid tumor growth by flavopiridol, a pan-cdk inhibitor. *(Clin Cancer Res* **14**(2):523-32*)*.

Bhaskar C. Das, Melissa E. Smith and **Kalpana, G.V.** (2008) Design and synthesis of 4-HPR derivatives for rhabdoid tumors (2008) (BMCL).

Bhaskar C. Das, Melissa E. Smith and **Kalpana, G.V.** (2008) Design, synthesis and biological activity of novel peptidomimetic derivatives of 4-HPR for rhabdoid tumors (2008) (BMCL).

Kalpana, G. V. and M. Smith (2008) "Development of targeted therapies for rhabdoid tumors based on the functions of Ini1/hSNF5 tumor suppressor" (*invited chapter for the monograph on Molecularly targeted Therapies for pediatric tumors Edited by Bob Arceci and Peter Houghton*).

Das. S. and **Kalpana, G. V.** (2008) Reverse two hybrid system to analyze the protein-protein interactions of HIV-1 viral and cellular proteins. (*invited chapter for the Molecular Microbiology Book series on "HIV-1 protocols" Edited by Prasad and Kalpana*).

Gissot M, Ting LM, Daly TM, Bergman LW, Sinnis P, **Kim K**. High Mobility Group Protein HMGB2 Is a Critical Regulator of *Plasmodium* Oocyst Development. J Biol Chem. 2008 Jun 20;283(25):17030-8. Epub 2008 Apr 8. PMID: 18400754

Binder EM, Lagal V, **Kim K**. The prodomain of *Toxoplasma gondii* GPI-anchored subtilase TgSUB1 mediates its targeting to micronemes. Traffic. 2008 Jun 2. [Epub] PMID: 18532988

Ting LM, Gissot M, Coppi A, Sinnis P, **Kim K**. Attenuated *Plasmodium yoelii* lacking purine nucleoside phosphorylase confer protective immunity. Nature Medicine 2008.

Drumm, J., Bilder, P., Sun, M., **Leyh, T.S.,** Chan J**.** Mycobacterium tuberculosis universal stress protein Rv2623 regulates bacillary growth by ATP-binding: requirement for establishing chronic persistent infection (submitted, *PLoS Pathogen*)

Cheng, H., Chang, C., Yu, A., Zhange, Y. Sun, M., **Leyh,** T. S., Holst, M. J. and McCammon, J. A. Diffusional channeling in the sulfate activating complex: Combined continuum modeling and coarse-grained brownian dynamics studies (submitted, *Biophysical Journal*).

Effraim, P.R., Wang, J., Avins, J., **Leyh, T.S.,** Gonzalez, R.L., Cornish, V.W. Ribosomal selection is independent of tRNA context. (submitted, *Nature Chemical Biology*).

Talloczy, Z, Martinez, J, Joset, D, Ray, Y, Gácser, A, Toussi, Mizushima, N, **Nosanchuk, JD**, Goldstein, H, Loike, J, Sulzer, D, Santambrogio, L. Methamphetamine inhibits antigen processing, presentation, and phagocytosis. PLoS Pathogens. 2008.4(2).e28.

Lázár-Molnár, E, Gácser, A, Freedman, GJ, Almo, SC, Nathenson, SG, **Nosanchuk, JD**. The PD-1/PD-L costimulatory pathway critically affects host resistance to the pathogenic fungus *Histoplasma capsulatum*. Proc Nat Acad Sci USA. 2008. 105(7):2658-2663.

Dadachova, E, Bryan RA, Howell, RC, Schweitzer, AD, Aisen, P, **Nosanchuk, JD**, Casadevall, A. The radioprotective properties of fungal melanin are a function of its chemical composition, stable radical presence, and spatial arrangement. Pigment Cell Melanoma Research. 2008.21(2):192-9.

Albuquerque, PC, Nakayasu, ES, Rodrigues, ML, Frases, S, Casadevall, A, Zancope-Oliveira, RM, Almedia, IC, **Nosanchuk, JD**. Vesicular transport in *Histoplasma capsulatum*: an effective mechanism for trans-cell wall transfer of proteins and lipids in ascomycetes. Cellular Microbiology. 2008. April 17 [epub].

Buissa-Filho R, Puccia R, Marques AF, Pinto FA, Munoz JE, **Nosanchuk JD**, Travassos LR, Taborda CP. Monoclonal antibody against the major diagnostic antigen of *Paracoccidioides brasiliensis* mediates immune protection in infected Balb/c mice challenged intratracheally with the fungus. Infection and Immunity. 2008. Jul;76(7):3321-8.

Li, S, Albuquerque, PC, Lazar-Molnar, E, Wang, X, Santambrogio, L, Gacser, A, **Nosanchuk, JD.** A monoclonal antibody to *Histoplasma capsulatum* alters the intracellular fate of the fungus in murine macrophage. Eukaryotic Cell. 2008. Jul;7(7):1109-17.

Trofa, D, Gacser, A, **Nosanchuk, JD**. *Candida parapsilosis*, an emerging fungal pathogen. Clincial Microbiology Reviews. 2008.

Nosanchuk, JD, Gacser, A. *Histoplasma capsulatum* at the host-pathogen interface. Microbes and Infection. 2008.

Datta K, Lees A, **Pirofski L.** Therapeutic Efficacy of a Conjugate Vaccine containing a Peptide Mimotope of the Cryptococcal Capsular Polysaccharide Glucuronoxylomannan (GXM). Clin Vaccine Immunol. 2008

Dash, P. K., Siddappa, N. B., Mangaiarkarasi, A., Anand, K. K., Padmanabhan, R. B, Mahendarkar, A. V., Mahadevan, A., Satishchandra, P., Shankar, S. K., **Prasad, V. R.**, Ranga, U. (2008) Expanded coreceptor use of Human Immunodeficiency Virus Type-1 subtype C molecular isolates from an Indian subject with HIV-1 associated dementia. *Retrovirology.* **5**:25

Garforth, S., Parniak, M. A. and **Prasad, V. R.** (2008) Utilization of deoxynucleoside diphosphates as substrate by a DNA polymerase. *Plos One*. **3**(4):e2074

Molano A, Illarionov, PA, Besra GS, **Putterman C**, Porcelli SA. Modulation of invariant natural killer T cell cytokine responses by indoleamine 2,3-dioxygenase. Immunology Lett 117:81-90, 2008.

Stohl W, Jacob N, Quinn WJ, Cancro MP, **Gao H**, **Putterman C**, Pricop L, Koss MN. Global T cell dysregulation in non-autoimmune-prone mice promotes rapid development of systemic lupus erythematosus-like autoimmunity in a BAFF-independent manner J Immunol 181:833-841, 2008.

Qing X, Barrat F, Hogarth M, **Putterman C**. Pathogenic anti-DNA antibodies modulate gene expression in mesangial cells: Involvement of HMGB1 in anti-DNA antibody induced renal injury. Immunology Lett, 2008.

Devoti JA, Rosenthal DW, Wu R, Abramson AL, **Steinberg BM**, Bonagura VR. Immune Dysregulation and Tumor-Associated Gene Changes in Recurrent Respiratory Papillomatosis: A Paired Microarray Analysis. Mol Med. 2008 Jul 6.

Hofmeyer KA, Ray A, **Zang X**. The contrasting role of B7-H3 (Invited Commentary). Proc Natl Acad Sci USA, 105: 10277-10278, 2008

Thompson RH*, **Zang X***, Lohse CM, Leibovich BC, Slovin S, Reuter VE, Blute ML, Russo P, Kwon ED*, Allison JP*. Serum soluble B7x is elevated in renal cell carcinoma patients and is associated with advanced stage. Cancer Res.

2009

Serreze, D. V., M. Niens, J. Kulik, and **T. P. DiLorenzo**. Bridging mice to men: using HLA transgenic mice to enhance the future prediction and prevention of autoimmune type 1 diabetes in humans. In: *Mouse Models for Drug Discovery*. M. Wiles and G. Proetzel, eds. Humana Press, Totowa, NJ.

Tamayev R, Zhou D, **D'Adamio L**. The interactome of the Amyloid betaeta Precursor Protein family members is shaped by phosphorylation of their intracellular domains. *Mol Neurodegener.* 2009 Jul 14;4(1):28. [Epub]

Venkataswamy MM, **Baena A**, **Goldberg M**, Bricard G, Im JS, Besra GS, Chan J, **Jacobs Jr. WR and Porcelli SA**. Enhancement of immunogenicity of *M. bovis* BCG by incorporation of NKT cell activating glycolipids. J Immunol. 183:1644-56 (2009).

Pericolini E, Gabrielli E, Cenci E, **De Jesus M**, Bistoni F, **Casadevall A**, Vecchiarelli A (2009). Involvement of glyco-receptors in galactoxylomannan-induced T cell death. J. Immunol.

DeJesus M, Nicola AM, Frases S, Lee IR, Mieses S, **Casadevall A** (2009) Galactoxylomannan-mediated immunological paralysis results from specific B cell depletion in the context of widespread immune system damage. J. Immunol.

Fonseca FL, Nimrichter L, **Cordero RJB**, Frases S, De Siqueira JR, Goldman DL, Andruszkiewicz R, Milewski S, Travassos LR, **Casadevall A,** Rodrigues ML (2009). A role for chitin and chitooligomers in the capsular architecture of Cryptococcus neoformans. Eukaryotic Cell.

Abboud N, DeJesus M, Nakouzi A, Cordero RJB, Puhato M, Fiser A, Rivera J, and **Casadevall A.** (2009) Identification of linear epitopes in bacillus anthracis protective antigen bound by neutralizing antibodies. J.Biol.Chem.

2010

Muehlbauer SM., Lima H., Goldmann D., **Jacobson, LS**., Goldberg MF., Palladino MA., Casadevall A. and **J. Brojatsch.** (2010) Proteasome inhibitors prevent caspase-1-mediated disease in rodents challenged with anthrax lethal toxin. Am. J. Pathol**.** (PMID: 20595632).

Guimaraes AJ, Frases S, **Cordero RJB**, Nimrichter L, **Casadevall A**, Nosanchuk JD (2010). Cryptococcus neoformans responds to mannitol by increasing capsule size in vitro and in vivo. Cell. Microbiol. 12:740-53. PMCID: PMC289187

De Jesus M, Chow S., **Cordero R.J.B.**, Frases S, **Casadevall A** (2010) Galactoxylomannans from Cryptococcus neoformans varieties neoformans and grubii are structurally and antigenically variable. Eukaryotic Cell 9:1018-28.

Janda A, **Casadevall A** (2010) Circular Dichroism Reveals Evidence of Coupling Between Immunoglobulin Constant and Variable Region Secondary Structure. Molecular Immunology 47:1421-5. PMCID: PMC2872143.

Babad, J., A. Geliebter, and **T. P. DiLorenzo**. 2010. T cell autoantigens in the NOD model. *Immunology 131:459-465*.

Sango, K., A. Joseph, M. Patel, K. Osiecki, M. Dutta, and **H. Goldstein.** Evaluation of highly active antiretroviral therapy in a humanized Rag2-/-γc-/- mouse model productively infected with HIV-1. *AIDS Res. Hum. Retro*. 2010;26:735-46.

Katakowski, JA and **Palliser D** (2010) siRNA-based topical microbicides targeting sexually transmitted infections. *Curr Opin Mol Ther*. 12:192-202

Garforth, S. J., Domaoal, R. A., **Lwatula, C**., Landau, M. J., Meyer, A. J., Anderson, K. S. and **Vinayaka Prasad** (2010) K65R and K65A substitutions in HIV-1 reverse transcriptase enhance polymerase fidelity by decreasing both dNTP misinsertion and mispaired primer extension efficiencies. *J. Mol. Biol.* **401**:33-44

Wong AC, Sandesara RG, Mulherkar N, Whelan SP, **Chandran K***. 2010. A forward genetic strategy reveals destabilizing mutations in the ebolavirus glycoprotein that alter its protease dependence during cell entry. J Virol. 84:163-175.

2011

Matsuda S, **Tamayev R**, **D'Adamio L**. Increased AbPP processing in Familial Danish Dementia patients. *J Alzheimers Dis.* In press.

Tamayev R, Matsuda S, Giliberto L, Arancio O, **D'Adamio L.** APP heterozygosity averts memory deficit in knock-in mice expressing the Danish Dementia BRI2 mutant. *EMBO J.* 2011 May 17;30(12):2501-9. doi: 10.1038/emboj.2011.161.

Miller EH, Obernosterer G, Raaben M, Herbert AS, Krishnan A, Ndungo E, Sandesara RG, Carette JE, Kuehne AI, Ruthel G, Dye JM, Whelan SP, Brummelkamp TR, **Chandran K***. Niemann-Pick C1 is a critical intracellular receptor for Ebola and Marburg virus entry. (submitted)

Miller EH, Harrison JS, Radoshitzky SR, Higgins CD, Chi X, Dong L, Kuhn JH, Bavari S, Lai JR, **Chandran K***. 2011. Inhibition of Ebola virus entry by a C-peptide targeted to endosomes. J Biol Chem. 286:15854-15861.

Brooks CF, Francia ME, Gissot M, **Croken MM**, **Kim K***, Striepen B*. *Toxoplasma gondii* sequesters centromeres to a specific nuclear region throughout the cell cycle. Proc Nat Acad Sci U S A. 2011 Mar 1;108(9):3767-72. Epub 2011 Feb 14. PubMed PMID: 21321216; PubMed Central PMCID: PMC3048097. * joint corresponding

Katakowski JA and **Palliser D.** Optimizing siRNA delivery to the genital mucosa. *Discov Med*. **2011** 11:124-32.

Barach YS, Lee JS, **Zang X**. T cell coinhibition in prostate cancer: new immune evasion pathways and emerging therapeutics. Trends Mol Med, 17: 47-55, 2011

Hofmeyer KA, Jeon H, **Zang X**. The PD-1/PD-L1 (B7-H1) pathway in chronic infection-induced cytotoxic T lymphocyte exhaustion. J Biomed Biotechnol, 2011:451694, 2011

2012

Casadevall A, **Janda A** (2012) Immunoglobulin isotype influences affinity and specifity. PNAS 109(31)12272-3.

Antal, Z., J. C. Baker, C. Smith, I. Jarchum, **J. Babad**, G. Mukherjee, Y. Yang, J. Sidney, A. Sette, P. Santamaria, and **T. P. DiLorenzo**. 2012. Beyond HLA-A*0201: New HLA-transgenic nonobese diabetic mouse models of type 1 diabetes identify the insulin C-peptide as a rich source of $CD8^+$ T cell epitopes. *J. Immunol. 188:5766-5775.*

Krupka M, Seydel K, **Feintuch C**, Yee, K, Kim, R, Lin C, Calder R, Petersen C, Taylor T **Daily J**. Mild Plasmodium falciparum malaria following an episode of severe malaria is associated with induction of the interferon pathway in Malawian Children. Infection and Immunity, 2012 Mar;80(3):1150-5.

Venkataswamy MM, **Goldberg MF**, Baena A, Chan J, Jacobs WR Jr, **Porcelli SA**. In vitro culture medium influences the vaccine efficacy of Mycobacterium bovis BCG. Vaccine. 30:1038-49 (2012).

Hofmeyer KA, Scandiuzzi L, Ghosh K, Pirofski LA, **Zang X**. Tissue-expressed B7x affects the immune response and outcome to lethal pulmonary infection. *J Immunol*, 2012.

2013

Mukherjee, G., A. Geliebter, **J. Babad**, P. Santamaria, D. V. Serreze, G. J. Freeman, K. V. Tarbell, A. Sharpe, and **T. P. DiLorenzo**. 2013. DEC-205–mediated antigen targeting to steady-state dendritic cells induces deletion of diabetogenic CD8 T cells independently of PD-1 and PD-L1. *Int. Immunol. (in press)*.

T. Bouklas , and **B. C. Fries***. C. neoformans* constitutes an ideal model organism to unravel the contribution of cellular aging to the virulence of chronic infections. Curr Opin Microbiol. 2013 Apr 27

T. Bouklas, X. Pechuan, D. L. Goldman, B. Edelman, A. Bergman, and **B. C. Fries**. Old *Cryptococcus neoformans* Cells Contribute to Virulence in Chronic Cryptococcosis MBio (in press).

Zhao R, **Chinai JM**, Buhl S, Scandiuzzi L, Ray A, Jeon H, **Ohaegbulam KC**, Ghosh K, Zhao A, Scharff MD, **Zang X**. HHLA2 is a member of the B7 family and inhibits human CD4 and CD8 T cell function. Proc Natl Acad Sci USA, 110: 9879-9884, 2013

Jeon H, **Ohaegbulam KC**, Abadi YM, **Zang X**. B7x and myeloid derived suppressor cells in the tumor microenvironment: A tale of two cities. OncoImmunology, e24744-1- e24744-3, 2013

Abadi YM, Jeon H, **Ohaegbulam KC**, Scandiuzzi L, Ghosh K, Hofmeyer KA, Lee JS, Ray A, Gravekamp C, **Zang X**. Host B7x promotes pulmonary metastasis of breast cancer. J Immunol, 190: 3806-3814, 2013

K. Seay, X. Qi, J. H. Zheng, C. Zhang, K. Chen, M. Dutta, K. Deneroff, C. Ochsenbauer, J. C. Kappes, D. R. Littman and **H. Goldstein**. Mice transgenic for CD4-specific human CD4, CCR5 and Cyclin T1 expression: A new model to study HIV-1 transmission and treatment efficacy. *PLoS ONE*, 2013;8(5):e63537.

2014

Wang J, Dixon SE, Ting LM, Liu TK, Jeffers V, **Croken MM**, Calloway M, Cannella D, Hakimi MA, **Kim K**, Sullivan Jr WJ. Lysine acetyltransferase GCN5b interacts with AP2 factors and is required for *Toxoplasma gondii* proliferation. PLoS Pathog. 2014 Jan;10(1):e1003830. doi: 10.1371/journal.ppat.1003830. Epub 2014 Jan 2. PubMed PMID: 24391497; PubMed Central PMCID: PMC3879359.

Croken MM, Qiu W, White MW, **Kim K**.Gene Set Enrichment Analysis (GSEA) of *Toxoplasma gondii* expression datasets links cell cycle progression and the bradyzoite developmental program. BMC Genomics. 2014 Jun 24;15:515. doi: 10.1186/1471-2164-15-515.PMID: 24962434 PubMed Central PMCID: PMC4092224.

Croken MM, Ma Y, Markillie LM, Taylor RC, Orr G, Weiss LM, **Kim K**. Distinct Strains of *Toxoplasma gondii* Feature Divergent Transcriptomes Regardless of Developmental Stage. PLoS One. 2014 Nov 13;9(11):e111297. doi: 10.1371/journal.pone.0111297. eCollection 2014. PubMed PMID: 25393307.

Jeon H, Vigdorovich V, Garrett-Thomson SC, Janakiram M, Ramagopal UA, Abadi YM, Lee JS, Scandiuzzi L, **Ohaegbulam KC**, **Chinai JM**, Zhao R, Yao Y, Mao Y, Sparano JA, Almo SC, **Zang X.** Structure and cancer immunotherapy of the B7 family member B7x. *Cell Reports*, 9:1089-1098, 2014

Arora P, Baena A, Yu KO, Saini NK, Kharkwal SS, **Goldberg MF**, Kunnath-Velayudhan S, Carreño LJ, Venkataswamy MM, Kim J, Lazar-Molnar E, Lauvau G, Chang YT, Liu Z, Bittman R, Al-Shamkhani A, Cox LR, Jervis PJ, Veerapen N, Besra GS, Porcelli SA. A single subset of dendritic cells controls the cytokine bias of natural killer T cell responses to diverse glycolipid antigens. Immunity. 40:105-16 (2014). PMID: 24412610.

Goldberg MF, Saini NK, Porcelli SA. Evasion of Innate and Adaptive Immunity by Mycobacterium tuberculosis. Microbiol Spectr. 2014 Oct;2(5). doi: 10.1128/microbiolspec.MGM2-0005-2013. Review. PMID: 26104343.

2015

Subramaniam KS, Spaulding E, Ivan E, Mutimura E, Kim RS, Liu X, Dong C, **Feintuch CM**, Zhang X, Anastos K, Lauvau G, **Daily JP.** The T cell inhibitory molecule BTNL2 is upregulated in mild Plasmodium falciparum infection and is protective during experimental cerebral malaria. J Infect Dis. 2015. Oct 15;212(8):1322-31.

Pappa V, Seydel K, Gupta S, **Feintuch CM**, Potchen MJ, Kampondeni S, Goldman-Yassen A, Veenstra M, Lopez L, Kim RS, Berman JW, Taylor T, **Daily JP.** Lipid metabolites of the phospholipase A2 pathway and inflammatory cytokines are associated with brain volume in paediatric cerebral malaria. Malar J. 2015 Dec 21;14(1):513.

Mukherjee, G., R. J. Chaparro, **J. Schloss**, C. Smith, C. D. Bando, and **T. P. DiLorenzo**. 2015. Glucagon-reactive islet-infiltrating CD8 T cells in NOD mice. *Immunology 144:631-640.*

Seay K, Church C, Zheng JH, Deneroff K, Ochsenbauer C, Kappes JC, Liu B, Jeng EK, Wong HC, **Goldstein H**. *In vivo* activation of human NK Cells by treatment with an Interleukin-15 superagonist potently inhibits acute *in vivo* HIV-1 infection in humanized mice. *J. Virology* 2015;89(12):6264-74.

Seay K, Khajoueinejad N, Zheng JH, Kiser P, Ochsenbauer C, Kappes JC, Herold B, **Goldstein H**. The vaginal acquisition and dissemination of HIV-1 infection in a novel transgenic mouse model is facilitated by coinfection with HSV-2 and is inhibited by microbicide treatment. *J. Virology* 2015;89:9559-9570.

Petro, C., González, P.A., Cheshenko, N., Jandl, T., Khajjoueinejad, N., Bénard, A., Sengupta, M., **Herold, B.C., Jacobs, W.R., Jr**. (2015) Herpes simplex type 2 virus deleted in glycoprotein D protects against vaginal, skin and neural disease. *eLife*. PMCID PMC4352706.

Janakiram M, **Chinai JM**, Zhao A, Sparano JA, **Zang X**. HHLA2 and TMIGD2: New immunotherapeutic targets of the B7 and CD28 families. *OncoImmunology*, 4: e1026534-1 - e1026534-3, 2015

Ohaegbulam KC, Assal A, Lazar-Molnar E, Yao Y, **Zang X**. Human cancer immunotherapy with antibodies to the PD-1 and PD-L1 pathway. *Trends in Molecular Medicine*, 21:24-33, 2015

Janakiram M, **Chinai JM**, Fineberg S, Fiser A, Montagna C, Medaverepu R, Castano E, Jeon H, **Ohaegbulam KC**, Zhao R, Zhao A, Almo SC, Sparano JA, **Zang X**. Expression, clinical significance, and receptor identification of the newest B7 family member HHLA2 protein. *Clinical Cancer Research*, 21:2359-2366, 2015

Chinai JM, Janakiram M, Chen F, Chen W, Kaplan M, **Zang X.** New immunotherapies targeting the PD-1 pathway. *Trends in Pharmacological Sciences.*

Arora P, Kharkwal SS, Ng TW, Kunnath-Velayudhan S, Saini NK, **Johndrow CT**, Chang YT, Besra GS, Porcelli SA. Endocytic pH regulates cell surface localization of glycolipid antigen loaded CD1d complexes. Chem Phys Lipids. 2015 Oct;191:75-83. doi: 10.1016/j.chemphyslip.2015.08.010. Epub 2015 Aug 22. PMID: 26306469.

Ng TW, Saavedra-Ávila NA, **Kennedy SC**, Carreño LJ, Porcelli SA. Current efforts and future prospects in the development of live mycobacteria as vaccines. Expert Rev Vaccines. 2015;14(11):1493-507. doi: 10.1586/14760584.2015.1089175. Epub 2015 Sep 14. PMID: 26366616.

2016

Feintuch CM, Saidi A, Seydel K, Chen G, Goldman-Yassen A, Mita N, Kim RS, Frenette PS, Taylor T, **Daily JP**. Activated neutrophils are associated with pediatric cerebral malaria vasculopathy in Malawian children. mBio 2016; 16 February, vol 7. No 1 e01300-15

Babad, J., R. Ali, **J. Schloss**, and **T. P. DiLorenzo**. 2016. An HLA-transgenic mouse model of type 1 diabetes that incorporates the reduced, but not abolished, thymic insulin expression seen in patients. *J. Diabetes Res. 2016:7959060*.

Sidney, J., **J. Schloss**, C. Moore, M. Lindvall, A. Wriston, D. F. Hunt, J. Shabanowitz, **T. P. DiLorenzo**, and A. Sette. 2016. Characterization of the peptide binding specificity of the HLA class I alleles B*38:01 and B*39:06. *Immunogenetics 68:231-236.*

Spaulding E., Fooksman D., **Moore JM.**, Saidi, Feintuch C., Reizis B., Chorro L., Daily J., and **G. Lauvau** 2016. STING-Licensed Macrophages Prime Type I IFN Production by Plasmacytoid Dendritic Cells in the Bone Marrow during Severe *Plasmodium yoelii* Malaria. **PLoS Pathogens**, 12(10):e1005975.

Saini NK, Baena A, Ng TW, Venkataswamy MM, **Kennedy SC**, Kunnath-Velayudhan S, Carreno L, Xu J, Chan J, Larsen MH, Jacobs Jr. WR, **Porcelli SA**. Suppression of autophagy and antigen presentation by Mycobacterium tuberculosis PE_PGRS47. Nat Microbiol, Nat Microbiol (9):16133 (2016). PMID: 27562263.

Olsen A, Chen Y, Ji Q, Zhu G, De Silva AD, Vilchèze C, Weisbrod T, Li W, Xu J, Larsen M, Zhang J, **Porcelli SA, Jacobs WR Jr, Chan J**. Targeting Mycobacterium tuberculosis Tumor Necrosis Factor Alpha-Downregulating Genes for the Development of Antituberculous Vaccines. MBio. 2016 May 31;7(3). pii: e01023-15.

Koirala P, Roth M, Gill J, **Chinai J**, Ewart M, Piperdi S, Geller D, Hoang B, Fatakhova K, Ghorpade M, **Zang X***, Gorlick R*. HHLA2, a new immune checkpoint member of the B7 family, is highly expressed in human osteosarcoma and associated with metastases and worse survival. *Scientific Reports*, in press.

2017

Bardhi A, Wu Y, Chen W, Zhu Z, Zheng JH, Wong H, Jeng E, Jones J, Ochsenbauer C, Kappes JC, Dimitrov DS, Ying T, **Goldstein H**. Potent in vivo NK cell-mediated elimination of HIV-1-Infected cells mobilized by a gp120-bispecific and hexavalent broadly neutralizing fusion protein. ***J. Virology*** 2017: pii: JVI.00937-17. doi: 10.1128/JVI.00937-17. [Epub ahead of print].

Flerin NC, Chen H, Glover TD, Lamothe PA, Zheng JH, Fang JW, Ndhlovu ZM, Newell EW, Davis MM, Walker BD, **Goldstein H**. T-Cell Receptor (TCR) Clonotype-Specific Differences in Inhibitory Activity of HIV-1 Cytotoxic T-Cell Clones Is Not Mediated by TCR Alone. ***J. Virology*** 2017;91:e02412-16. doi: 10.1128/JVI.02412-16.

Thomas T, Seay K, Zheng JH, Zhang C, Ochsenbauer C, Kappes JC, **Goldstein H**. High-Throughput Humanized Mouse Models for Evaluation of HIV-1 Therapeutics and Pathogenesis. *Methods Mol Biol*. 2016;1354:221-35. doi: 10.1007/978-1-4939-3046-3_15

Sankin A, Narasimhulu D, **John P**, Gartrell B, Schoenberg M, **Zang X**. The expanding repertoire of targets for immune checkpoint inhibition in bladder cancer: what lies beneath the tip of the iceberg, PD-L1. *Urologic Oncology*, in press