30TH International Conference on Antiviral Research (ICAR)

May 21-25, 2017
HILTON ATLANTA
Atlanta, GA

hosted by the
INTERNATIONAL
SOCIETY FOR ANTIVIRAL
RESEARCH
(ISAR)

Program AND Abstracts

30TH International Conference on Antiviral Research (ICAR)

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- Gertrude Elion Memorial Award Lecture
- Hepatitis Viruses and Retroviruses
- Poster Session 1

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- Emerging Viruses
- New Member and First Time Attendee Networking
- Career Development Panel

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- William Prusoff Young Investigator Award Lecture
- Respiratory Viruses
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- Medicinal Chemistry
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International Society for Antiviral Research and 30th International Conference on Antiviral Research

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Keynotes & Networking

Keynote Addresses

Sunday, May 21, 2017

Historical Context and Biological Enigma of Rhinovirus C

Ann C. Palmenberg, PhD
University of Wisconsin-Madison

Antivirals at the Interface with Public Health: A Case Study of Polio

Mark A. Pallansch, PhD
Centers for Disease Control and Prevention

Monday, May 22, 2017

Impact of Transmitted HIV Phenotype on Host-Virus Interactions and Disease Progression

Eric Hunter, PhD
Emory University

Wednesday, May 24, 2017

Zika Antiviral and Vaccine Development

Pei-Yong Shi, PhD
University of Texas Medical Branch

Networking Events

Women in Science Roundtable
Sunday, May 21, 2017
12:00 PM – 1:45 PM
CRYSTAL BALLROOM

Opening Reception
Sunday, May 21, 2017
6:30 PM – 8:30 PM
CRYSTAL BALLROOM

New Member and First Time Attendee Networking
Tuesday, May 22, 2017
12:30 PM – 2:00 PM
ROOM 204-205

Career Development Panel
Tuesday, May 22, 2017
2:00 PM – 3:00 PM
ROOM 203

Conference Reception and Banquet
Wednesday, May 23, 2017
7:00 PM – 10:00 PM
WEST BALLROOM

30TH International Conference on Antiviral Research (ICAR)

Schedule at a Glance

Sunday, May 21, 2017

TIME	EVENT	LOCATION
11:00 AM – 5:00 PM	Registration	GRAND BALLROOM PRE-FUNCTION
12:00 PM – 1:45 PM	Women in Science Roundtable	CRYSTAL BALLROOM
2:00 PM – 4:00 PM	Drug Discovery and Development 101	WEST BALLROOM
4:00 PM – 4:30 PM	Coffee Break	GRAND BALLROOM PRE-FUNCTION
4:30 PM – 6:30 PM	Welcome/Keynote Address: Ann C. Palmenberg, PhD and Mark A. Pallansch, PhD	WEST BALLROOM
6:30 PM – 8:30 PM	Opening Reception *Light hors d'oeuvres served*	CRYSTAL BALLROOM

Monday, May 22, 2017

TIME	EVENT	LOCATION
8:00 AM – 5:30 PM	Registration	GRAND BALLROOM PRE-FUNCTION
8:00 AM – 9:00 AM	Continental Breakfast	GRAND BALLROOM PRE-FUNCTION
9:00 AM – 9:55 AM	Keynote Address: Eric Hunter, PhD	WEST BALLROOM
9:55 AM – 12:30 PM	Antiviral Immunity Symposium	WEST BALLROOM
11:00 AM – 11:30 AM	Coffee Break	GRAND BALLROOM PRE-FUNCTION
12:30 PM – 2:00 PM	Lunch	ON YOUR OWN
2:00 PM – 2:45 PM	Gertrude Elion Memorial Award Lecture	WEST BALLROOM
2:50 PM – 5:00 PM	Hepatitis Viruses and Retroviruses	WEST BALLROOM
3:30 PM – 4:00 PM	Coffee Break	GRAND BALLROOM PRE-FUNCTION
5:00 PM – 7:00 PM	Poster Session 1 *Light hors d'oeuvres served*	EAST BALLROOM

Tuesday, May 23, 2017

TIME	EVENT	LOCATION
8:00 AM – 3:00 PM	Registration	GRAND BALLROOM PRE-FUNCTION
8:00 AM – 9:00 AM	Continental Breakfast	GRAND BALLROOM PRE-FUNCTION
9:00 AM – 9:30 AM	Antonin Holý Memorial Award Lecture	WEST BALLROOM
9:30 AM – 12:30 PM	Emerging Viruses	WEST BALLROOM
10:30 AM – 11:00 AM	Coffee Break	GRAND BALLROOM PRE-FUNCTION
12:30 PM – 2:00 PM	New Member and First Time Attendee Networking *Light snacks served*	ROOM 204-205
2:00 PM – 3:00 PM	Career Development Panel *Snacks and beverages served*	ROOM 203

Wednesday, May 24, 2017

TIME	EVENT	LOCATION
7:30 AM – 5:00 PM	Registration	GRAND BALLROOM PRE-FUNCTION
7:30 AM – 8:30 AM	Continental Breakfast	GRAND BALLROOM PRE-FUNCTION
8:30 AM – 9:00 AM	Shotgun Oral Presentations	WEST BALLROOM
9:00 AM – 9:50 AM	Keynote Address: Pei-Yong Shi, PhD	WEST BALLROOM
9:50 AM – 12:30 PM	Emerging Infections Symposium	WEST BALLROOM
10:30 AM – 11:00 AM	Coffee Break	GRAND BALLROOM PRE-FUNCTION
12:30 PM – 1:45 PM	Lunch	ON YOUR OWN
1:45 PM – 2:00 PM	ISAR Business Meeting	WEST BALLROOM
2:00 PM – 2:30 PM	William Prusoff Young Investigator Award Lecture	WEST BALLROOM
2:30 PM – 3:30 PM	Respiratory Viruses	WEST BALLROOM
3:30 PM – 5:30 PM	Poster Session 2 *Light hors d'oeuvres served*	EAST BALLROOM
7:00 PM – 7:30 PM	Closing Reception *Light hors d'oeuvres served*	GRAND BALLROOM PRE-FUNCTION
7:30 PM – 10:00 PM	Closing Banquet *Dinner served*	WEST BALLROOM

Thursday, May 25, 2017

TIME	EVENT	LOCATION
8:00 AM – 12:00 PM	Registration	GRAND BALLROOM PRE-FUNCTION
8:00 AM – 9:00 AM	Continental Breakfast	GRAND BALLROOM PRE-FUNCTION
9:00 AM – 10:30 AM	Medicinal Chemistry	WEST BALLROOM
10:30 AM – 11:00 AM	Coffee Break	GRAND BALLROOM PRE-FUNCTION
11:00 AM – 12:30 PM	DNA Viruses and Respiratory Viruses	WEST BALLROOM
12:30 PM	Conference Concludes	

2017 ISAR Award Winners

Gertrude Elion Memorial Lecture Award

Michael J. Sofia, PhD

Mike Sofia is the principle inventor of sofosbuvir (Sovaldi and Harvoni), the first curative treatment for chronic hepatitis C. He is Chief Scientific Officer of Arbutus Biopharma, a company focused on the discovery and development of curative therapies for hepatitis B. He is listed on more than 50 US patents and on numerous patent applications, and has authored over 100 research papers and 12 book chapters. He holds a professorship at the Baruch S. Blumberg Institute in Doylestown, PA and is an adjunct professor at Drexel University School of Medicine in Philadelphia.

Mike earned a BA degree in chemistry from Cornell University in 1980 and received his PhD in organic chemistry from the University of Illinois, Urbana-Champaign in 1984. He did his postdoctoral training in synthetic organic chemistry as an NIH fellow at Columbia University. During his early career in the pharmaceutical industry, he held the position of Group Director for New Leads Chemistry at Bristol-Myers Squibb, Vice President of Research at Intercardia Research Labs and research positions of increasing responsibility at Eli Lilly and at the Squibb Institute for Medical Research. He then moved to Pharmasset and was senior vice president for chemistry until its acquisition by Gilead in 2012. He then co-founded OnCore Biopharma, which merged with Tekmira in 2015 to form Arbutus.

In addition to his "day job," Mike is a member of the editorial advisory boards of several scientific journals, the Board of the Blumberg Institute and the Board of Trustees of the University of the Sciences in Philadelphia. He was the recipient of Pennsylvania Bio's 2014 Scientific Achievement Award, the 2015 Heroes of Chemistry Award of the American Chemical Society and the 2016 IUPAC-Richter Prize. Together with Charlie Rice and Ralf Bartenschlager, he received the 2016 Lasker-Debakey Award in Clinical Medical Research for his contributions to the discovery of a cure for hepatitis C.

Antonín Holý Memorial Lecture Award

C. K. (David) Chu, PhD

David Chu is a Distinguished Research Professor, Emeritus at the College of Pharmacy of the University of Georgia. He obtained a BS degree in pharmacy from Seoul National University, and after serving as an officer in the Korean Navy, he came to the United States, receiving a MS degree from Idaho State University in 1968 and a PhD in chemistry from the State University of New York at Buffalo in 1975. After working as a postdoctoral fellow in drug discovery at the Memorial Sloan-Kettering Institute of Cancer Research, New York, he stayed on as a research associate for six years before joining the faculty of the University of Georgia in 1982.

David has devoted his 40-year career in medicinal chemistry to the discovery of anticancer and antiviral agents. He has published more than 300 peer-reviewed articles in organic, biochemical and medicinal chemistry, and has edited four textbooks. During his academic career he has discovered a number of clinical candidates for cancer and for viral diseases, and he is listed on 60 US patents. He was one of co-founders of Pharmasset and ATEA Pharmaceuticals. He has trained more than 120 graduate students and postdoctoral fellows and has maintained an active research program in drug design and synthesis since his retirement in 2008. His program in drug discovery has been recognized nationally and internationally.

Among his many honors, David has held an endowed professorship of the University of Georgia Research Foundation, received the Creative Research Medal from the University of Georgia and an NIH Merit Award (2001-2011). He is an elected Fellow of AAAS. For his achievements in nucleoside chemistry and chemotherapy, he received the John A. Montgomery Award from the International Round Table Society in 2014. He was elected a Fellow of the National Academy of Inventors in 2015 and received the Willis Gregory Award from the School of Pharmacy of the SUNY Buffalo in 2017.

William Prusoff Young Investigator Lecture Award

Maaike Everts, PhD

Maaike (pronounced "Micah") Everts is an associate professor in the Division of Infectious Diseases of the Department of Pediatrics, University of Alabama School of Medicine at Birmingham. She was born in Meppel, the Netherlands. After receiving a masters degree in pharmaceutical sciences and a PhD in pharmacokinetics and drug delivery from the University of Groningen, she moved to UAB for postdoctoral training with David Curiel in the Division of Human Gene Therapy, where she pursued her interest in targeted gene delivery for the treatment of cancer, using adenoviral vectors. She joined the UAB Department of Pathology in August 2005, continuing her research on targeted therapies using gene therapy and nanotechnology approaches.

Since 2009, Maaike has been the associate director of the Alabama Drug Discovery Alliance, a collaboration between UAB and Southern Research, with the goal of finding new small-molecule drugs for unmet medical needs in a variety of therapeutic areas. She also assists physician-investigators with the IND application process, and provides quality assurance for the UAB Vector Production Facility, which manufactures novel drugs for Phase I clinical trials. She is also the administrative director for the Antiviral Drug Discovery and Development Center, a multi-institutional consortium headed by Rich Whitley and funded by a U19 grant from NIAID.

Maaike joined ISAR in 2015 and attended the 28th ICAR in Rome. She notes that she was impressed by the collegiality of the attendees and the inter-disciplinary nature of the sessions, which merged biology with medicinal chemistry and other disciplines needed for effective antiviral research. In 2016 she was invited to join the Women in Science committee and to be responsible for organizing the career development panel. Maaike says that attending ICAR "has truly been a joy: members are extremely encouraging of each other and provide mentorship throughout the different stages of their careers."

ICAR
Career Development Panel

Please join us for a panel discussion about career opportunities in antiviral research at the 30th ICAR meeting. In a new format for this event, this year we will host an excellent group of panelists who are recognized experts in various areas of antiviral research and have pursued successful career in academia, government, or industry. As part of a moderated panel discussion, they will be ready to share their experience, answer questions, and provide feedback about career development. They will highlight the similarities and differences between different sectors of antiviral research. At the end of the formal panel discussion, you will have opportunity to informally network with your colleagues and make new contacts.

The event is open to all ICAR attendees. Please sign up during the ICAR conference at the registration desk before 8:30 AM on Tuesday, May 23.

Sunday
May 21, 2017
12:00 PM – 1:45 PM
CRYSTAL BALLROOM

5th Annual WOMEN IN SCIENCE ROUNDTABLE

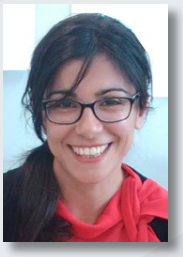
This session is open to all ICAR attendees, both women and men and will address the challenges and opportunities encountered by female scientists while navigating the twists and turns of career progression in todays' environment. This session affords the opportunity for scientists to discuss and exchange ideas on a variety of different topics.

Women in Science Winners

The International Society for Antiviral Research and The Chu Family Foundation Scholarship for Women Scientists

ISAR and The Chu Family Foundation (TCFF) Committee is excited to announce the winners of its 2017 Scholarship for Women Scientists. This scholarship supports the professional development of women with potential for significant contribution in the field of Antiviral Research.

2017 Awardees

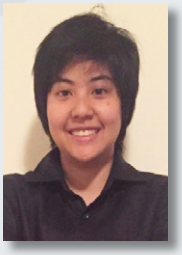


Angela Corona
MONSERRATO, ITALY

Angela obtained her degree in Pharmaceutical Chemistry at University of Cagliari in 2010 and acquired her PhD in "Human and Environmental Biology and Biochemistry School" in 2014. She is a post-doc fellow in the Laboratory of Molecular Virology of the Department of Life and Environmental Sciences, University of Cagliari.

Makda Gebre
UTAH, USA

Makda is currently finishing her Master's degree at the Institute for Antiviral Research at Utah State University. She enjoys the field of Virology and is currently working on a project that investigates differences between Chikungunya strains in disease manifestation and response to antiviral treatment.

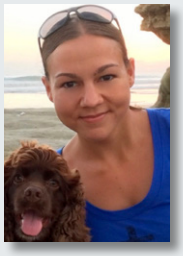


Naphak Modhiran
ANNERLEY, AUSTRALIA

Naphak is currently working as a post-doc at UQ in School of Chemistry and Molecular Bioscience. She received her BSc. (Biology) and MSc. (Microbiology) from Mahidol University, Thailand. She was awarded her PhD in January 2017 at UQ, focusing on dengue viral protein and its implications in pathogenesis.

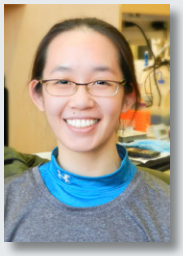
Verónica Quintana
BUENES AIRES, ARGENTINA

Verónica was born in Buenos Aires, Argentina. She studied Chemistry at Buenos Aires University. As an undergraduate student, she started working ad honorem at the Virology Laboratory on antiviral research. She is currently performing her PhD thesis focused on dengue antiviral development with a fellowship of the National Research Council.



Jessica von Recum
CALIFORNIA, USA

Jessica received her PhD in virology from the Humboldt University of Berlin, Germany, in 2014. Afterwards, she moved to San Diego to study host-directed antivirals against influenza viruses in an ex vivo human lung tissue model. She is currently a post-doctoral fellow at UCSD but will move back to Germany soon to continue her research at Philipps-Universität Marburg.



May Wang
MASSACHUSETTS, USA

May graduated from Tufts University with a degree in Biochemistry. Currently she is pursuing her PhD degree in the Harvard Virology program in the lab of Dr. James Cunningham, developing and studying the mechanisms of action of small molecule inhibitors of Lassa virus and Ebola virus entry.

Linlin Zhang
LUBECK, GERMANY

Linlin was born in 1986 in Heze, Shandong province, Eastern China. After school, she studied pharmaceutical engineering at the Qingdao University of Science and Technology (QUST). After obtaining her BSc degree, she moved on to the East China University of Science and Technology (ECUST) and Fudan University, both in Shanghai, where she was awarded an MSc degree (from ECUST). Her specialization was protein refolding. Because of this expertise, Prof. Rolf Hilgenfeld hired her in 2012 to work under him at the University of Lübeck, Germany. In her work towards PhD, she investigated the binding of antiviral compounds to proteases of emerging viruses (corona-, entero-. and flaviviruses) by X-ray crystallography. Following her PhD, she continues to work in the same department as a postdoc.

2017 ICAR Speaker Biographies

Robert W. Buckheit, Jr. PhD

Dr. Robert (Bob) Buckheit is President and Chief Scientific Officer of ImQuest BioSciences and Executive Vice President of ImQuest Pharmaceuticals, companies that he founded in 2004 and 2005. During his 25 years of experience in antiviral research Bob has assisted academic researchers, biotechnology companies, and pharmaceutical companies to develop agents to treat infectious disease based on his expertise in antiviral screening, mechanism of action, resistance and combination assays. Bob's research has resulted in over 180 publications in the peer review literature, especially involving the development of HIV therapeutic and prevention products. Over the past decade he has led efforts resulting in three IND submissions on the pyrimidinedione series of anti-HIV compounds for HIV therapy (IQP-0410) and as topically applied prevention agents (vaginal IQP0528 gel, Vaginal-rectal IQP-0528 DuoGel).Bob was named Entrepreneur of the Year in 2010 by the Maryland Entrepreneur Council in recognition of his leadership and the growth and development of ImQuest BioSciences, and ImQuest BioSciences was named the 2010-2011 Small Business of the Year by the Maryland Technology Council. ImQuest was also named one of Frederick's Best Places to Work in 2014. Bob has been a member of ISAR for over 20 years during his tenures with Southern Research Institute, Therimmune Research Corporation, and ImQuest BioSciences. He has held various positions to support the Society and the annual Conference, including the Membership Committee, the Poster Awards Committee, and the Conference Committee. Bob chaired the Program Committee for 6 years and took an active role in improving the scientific robustness of the annual conference, instituting the Drug Discovery & Development 101, the Keynote Address, Shotgun Poster presentations for young investigators, and the development of key scientific mini-symposia at each conference. In 2010, Bob was also elected to serve on the Board of Directors of ISAR and was elected to the role of President for a six-year term beginning in 2012. Bob serves as Scientific Advisor and consultant for a variety of biotechnology and pharmaceutical companies, and supports the local Frederick community by serving terms on the Board of Directors of the local biotechnology incubator (Frederick Innovative Technology Center, 2010-2014), and the Weinberg Center for the Performing Arts. Bob is a member of the Editorial Board for Antiviral Research, Future Virology, and Expert Opinion on Investigational Drugs, and acts as an ad hoc reviewer for a variety of scientific journal and special emphasis review panels for the National Institutes of Health. Bob also is an adjunct Professor at Hood College in Frederick MD and has successfully mentored many students obtaining their Masters Degrees in the BioSciences Program.

Mariano A. Garcia-Blanco, MD, PhD

Dr. Garcia-Blanco was born in Puerto Rico and lived in his native island until he went to Harvard College for his undergraduate education. Subsequently he received his MD and PhD in Molecular Biophysics and Biochemistry from Yale University, and completed a fellowship with Dr. Phillip Sharp at the Massachusetts Institute of Technology. From 1990 to 2014 Dr. Garcia-Blanco was a faculty member at Duke University, where he was the inaugural Charles D. Watts Professor of Molecular Genetics and Microbiology, Professor of Medicine, and Director of the Duke Center for RNA Biology. Since 2014 Mariano A. Garcia-Blanco, MD, PhD, has been Professor and Chair of the Department of Biochemistry and Molecular

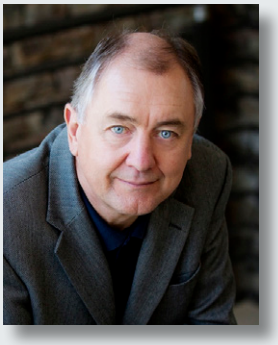
Biology, and the Mildred Hajek Vacek and John Roman Vacek Distinguished Chair in Honor of President Truman G. Blocker, Jr. at The University of Texas Medical Branch in Galveston, Texas, USA. He also is Professor of Emerging Infectious Diseases at Duke-NUS Medical School in Singapore.

Dr. Garcia-Blanco has made seminal and clinically relevant contributions to RNA biology and virology. His studies on mRNA splicing have shed light on the mechanisms of splicing regulation, disease-causing mis-regulation of splicing and a new type of RNA therapy. In the last ten years Dr. Garcia-Blanco has focused his RNA expertise on pathogenic flaviviruses, which threaten both US and global public health and for which there is no approved therapy.

Dr. Garcia-Blanco was elected to the Association of American Physicians (2011), fellow of the American Association for the Advancement of Science (2011), and fellow of the American Academy of Microbiology (2013). He has served on the editorial boards of *Molecular and Cellular Biology, RNA* and *Scientific Reports*. He has served and continues to serve on several boards such as the Council of Scientific Advisers of the International Centre Genetic Engineering and Biotechnology, a body of the United Nations. He is funded by NIH (USA) and NMRC (Singapore) grants and has published over 160 research articles, reviews and book chapters.

Adolfo García-Sastre, PhD

Dr. García-Sastre is a Professor in the Departments of Microbiology and Medicine, and Director of the Global Health and Emerging Pathogens Institute at Icahn School of Medicine Mount Sinai in New York. He is also Principal Investigator for the Center for Research on Influenza Pathogenesis (CRIP), one of five NIAID Centers of Excellence for Influenza Research and Surveillance (CEIRS). For the past 25 years, his research interest has been focused on the molecular biology, virus-host interactions, innate immunity and pathogenesis of influenza viruses and several other RNA viruses, as well as on the development of new vaccines and antivirals. He has more than 480 peer-reviewed publications in these areas of research. He was President of the International Society for Vaccines in 2014-2015.



Eric Hunter, PhD

Eric Hunter is Professor in the Department of Pathology and Laboratory Medicine at Emory University, Atlanta, GA. He is Co-Director of the Emory Center for AIDS Research and a Georgia Research Alliance Eminent Scholar. For the past several years his laboratory has investigated the molecular virological mechanisms underlying HIV transmission among heterosexual couples living in Rwanda (Projet San Francisco) and Zambia (ZEHRP) with an aim toward developing novel vaccine approaches that might prevent these transmission events. Recently this work has expanded to develop an understanding of the roles that the transmitted virus phenotype and host immune responses, in both donor and recipient partners, play in defining the rate and severity of HIV-1 disease progression.

His bibliography includes over 250 articles, reviews and book chapters. He has been the recipient of 4 NIH merit awards for his work on retrovirus molecular biology. After serving as Editor in Chief of the journal *AIDS Research and Human Retroviruses* for 10 years, he currently serves on the Editorial boards of several academic journals, and on the external advisory committees for several academic and commercial institutions, and NIH Institutes.

Rebecca Kaufman, JD

Becky Kaufman is a partner with King & Spalding's Intellectual Property Practice Group. Her practice includes U.S. and international patent prosecution, client counseling, adversarial proceedings and transactional due diligence in the biological and chemical arts. She also handles licensing, strategic collaborations and other agreements involving intellectual property. Ms. Kaufman represents companies, both private and public, investors and research organizations. Her industry expertise includes the life sciences, industrial biotechnology and renewable chemicals. She regularly counsels emerging companies on strategies for developing patent portfolios to protect commercial products.

Before joining the firm, Ms. Kaufman was a principal with Cordova Ventures, an early stage venture capital fund, where she worked closely with Cordova's portfolio companies on patent and business matters. Prior to that, she worked for the Georgia Institute of Technology's Advanced Technology Development Center, where she counseled start-ups on patent and business issues and was involved in the formation of the joint Georgia Tech/Emory University biotechnology incubator, EmTech Bio.

Ms. Kaufman is active in the Southeastern life sciences community. She is a member of the Executive Committee of Southeast BIO(SEBIO), a non-profit organization devoted to the growth of the regional life sciences economy, as well as the immediate past Chairman. SEBIO's forty five member Board of Directors is broadly representative of the region's venture capital, entrepreneurial and technology transfer community. Ms. Kaufman has twice served as Chairman of the SEBIO Investor Forum, the region's premier life sciences venture capital conference. She has served as a member of the Biotechnology Advisory Committee for the Sid Martin Biotechnology Incubator at the University of Florida as well as the Advisory Board for the Emerging Leaders Network (ELN) of Georgia BIO. She is a frequent speaker at industry meetings and has written numerous articles, including many focused on the life sciences industry in the Southeast. Ms. Kaufman was recently named to *Tech Journal South*'s list of the 25 most influential people in the Southeast technology and business world.

Ms. Kaufman received a BS degree in Biology, with honors, from Wake Forest University in 1991. She received her JD degree from the University of Pittsburgh School of Law in 1994. After graduating from law school, she spent more than two years working on a PhD in molecular biology, first at the University of Pittsburgh School of Medicine and later, at the George Washington University School of Medicine.

Jeffrey Murry, PhD

Jeffrey Murry received his PhD in immunology and infectious diseases from Harvard University and is currently a Research Scientist in Discovery Virology at Gilead Sciences. Over the past 7 years his research has investigated the regulation of latent HIV, a critical barrier to achieving a cure. In HIV infected individuals on suppressive antiviral therapy, latent HIV can be found in resting CD4 T cells in a quiescent state that is refractory to treatment. Infected cells that fail to express viral proteins evade antiviral surveillance and persist for decades. Long lived latent HIV reservoirs reseed infection if antiretroviral therapy is interrupted, leading to viral rebound.

Before joining Gilead Sciences, Dr. Murry was a Hewitt Fellow at the Salk Institute in La Jolla, where he worked with Dr. John Young and studied host pathways that regulate latent virus. This work identified novel pathways involved in regulating HIV transcription, such as the sulfonation pathway, as well as several components that regulate HIV through the NF-kB pathway such as cIAP1 and CYLD.

Dr. Murry's work at Gilead has focused on identifying novel strategies for inducing and eliminating latent HIV, relying primarily on the direct assessment of potential therapeutic agents in latent cells isolated from HIV-infected individuals. This work has explored novel chemical matter identified through high throughput screens, targeted approaches and repurposed agents originally developed for other indications. Combinatorial approaches have led to the identification of synergistic combinations that strongly activate HIV transcription without upregulation of markers typically associated with T cell activation. At the same time, his group is examining the effects of these agents on antiviral immunity and evaluating immune modulatory strategies designed to stimulate clearance of infected cells.

Mark A. Pallansch, PhD

Dr. Pallansch is Director, Division of Viral Diseases Division of Viral Diseases, Centers for Disease Control and Prevention in Atlanta, Georgia. He received his BS in Biochemistry (1976) from Virginia Tech and PhD in Biochemistry (1982) from the University of Wisconsin-Madison. He then completed a postdoctoral fellowship (1984) in Virology (Persistent Measles Infection) at the Rockefeller University in New York.

Dr. Pallansch joined the CDC in 1984 as Chief of the Enterovirus Section in the Respiratory and Enteric Viruses Branch until becoming Chief, Polio and Picornavirus Laboratory Branch in 2007. Since assuming his current position in March of 2011, he now leads an exceptional group of more than 250 laboratory and epidemiology scientists, laboratory technicians, data managers and program analysts who are responsible for many aspects of domestic and global viral vaccine preventable diseases, including laboratory support for global efforts at polio eradication and measles elimination, domestic vaccination policy, surveillance and evaluation, reference laboratory testing for respiratory and enteric viral diseases, and research on new viral vaccine development and evaluation. He has also been actively involved in collaborations providing technical expertise and conceptual evaluation of issues for risk assessment and management, including multiple modeling approaches, as well as strategic planning within the polio eradication program.

Dr. Pallansch is the author or co-author of more than 250 publications and book chapters. He has been awarded the Pan American Society for Clinical Virology Ed Nowakowski Senior Memorial Clinical Virology Award (2008) and the Sigma Xi Walter R. Dowdle Award for Achievement in Public Health Science (2008). He has also received numerous awards at the CDC, including the Sheppard Award for Scientific Excellence (1988, 2012 and 2015), U.S. Public Health Service Special Recognition Award (1989), CDC Special Service Award (1991), James H. Nakano Citation (1996, 1999, 2001, 2002, 2003, 2009), Health and Human Services Group Distinguished Service Award (1999), and Stephen R. Preblud Award (2002, 2006). He currently is a member of the Picornavirus Study Group of the International Committee on Taxonomy of Viruses.

Ann C. Palmenberg, PhD

Ann Palmenberg received her BS degree (Chemistry) from St. Lawrence University in 1970, and her PhD from the University of Wisconsin- Madison (Biochemistry, 1975), working in the laboratory of Paul Kaesberg. She received postdoctoral training with Charles Weissmann (Zurich) and Roland Rueckert (Madison). As the PI of a continuously funded (NIH) independent research program at the UW-Madison since 1978, Ann is now a Professor in the Department of Biochemistry and the Institute for Molecular Virology.

Her work on the molecular biology of positive-sense RNA viruses, particularly picornaviruses in the cardiovirus and enterovirus (rhinovirus) genera, has achieved international stature for its breadth and content. She has expertise in computational modeling of protein: protein interactions, bioinformatics (sequence analysis), recombinant protein expression, protein purification, protein detection, enzyme assays (activity and complex formation), and structural biology (crystallography, NMR, cryoEM). She has published extensively on these topics, both for research and technical audiences. She is an expert in RNA biochemistry, RNA dynamics, and RNA evolution. She teaches virology, bioinformatics and molecular modeling at the graduate and undergraduate levels.

Ulrike Protzer, PhD

Ulrike Protzer is an expert virologist with many years of research in hepatitis B virus (HBV) molecular virology, virus-host interaction and immunology. Ulrike studied medicine in Erlangen and Switzerland. She has a broad background in infectious diseases, gastroenterology, hepatology and medical virology obtained during her clinical training, and has passed board exams in Internal Medicine (1996) as well as in Microbiology and Virology in 2005.

Her scientific efforts focus on understanding the interaction between HBV and its host and on translating this knowledge into novel therapeutic approaches. She developed cell culture and mouse models of HBV infection, and is using these to exploit therapeutic vaccines, adoptive T cell therapies and antibodies to reconstitute HBV-specific immunity and finally cure HBV.

Isaac R. Rodriguez-Chavez, PhD, MHS, MS

Dr. Isaac R. Rodriguez-Chavez is a science executive with expertise in Virology (Viral Immunology and Viral Oncology) and Vaccinology. He is a science consultant providing services on project and program management in science and technology, business development, pre-clinical and clinical operations, regulatory affairs, and securing non-dilutive funds for small companies and universities.

Overall, Dr. Rodriguez-Chavez has over 17 years of professional work experience at the federal government of the United States, industry and non-profit research organizations. Past positions he has held include: Vice-President for Research at the Texas Biomedical Research Institute, San Antonio, TX; Director of the AIDS and Immunosuppression Program at the National Institute of Dental and Craniofacial Research, National Institutes of Health (NIH); Senior Clinical Scientist at Schering Plough Corporation; and Scientist and Program Official for HIV Vaccine Programs at the National Institute of Allergy and Infectious Diseases, NIH. He has managed basic, translational and clinical research projects connecting global health with the fields of Infectious Diseases, Vaccinology, Oncology, and Immunology. Dr. Rodriguez-Chavez has led multidisciplinary teams of professionals in international clinical networks that have developed novel prevention and therapeutic interventions. He has managed multi-million dollar funds and resources for programs to improve public health. He has designed, implemented and overseen clinical trials and public health policies in support of HIV/AIDS research and complications. He has also led management for global operations and regulatory affairs of clinical laboratories supporting HIV vaccine clinical trials, creating and implementing Good Clinical Laboratory Practices.

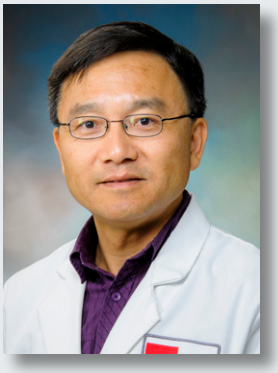
As a senior clinical scientist, Dr. Rodriguez-Chavez has served as a co-Investigator in Phase I-III clinical trials testing new drugs for AIDS malignancies and respiratory diseases. He has published multiple funding opportunity announcements to fund thousands of research projects, influencing HIV/AIDS research globally. He has led multiple teams of editors for scientific publishing in Virology and Viral Oncology. He has published numerous papers and he has been an invited speaker in multiple, international scientific events. He has also served as Associate Editor and Reviewer of scientific journals. He has led major national and international scientific conferences and collaborative projects to improve public health.

David Safronetz, PhD

Dr. David Safronetz received his PhD in Medical Microbiology from the University of Manitoba in 2009. During his graduate work on New World hantaviruses, David developed a keen interest in emerging viral pathogens, particularly those that are rodent-borne. He continued his training within the Laboratory of Virology at Rocky Mountain Laboratories (National Institute of Allergy and Infectious Diseases, National Institutes of Health) where he held the positions of Visiting Fellow followed by Staff Scientist. Throughout this time, David expanded his research to include the study of arenaviruses as well as other zoonotic pathogens. His research primarily focused on developing and characterizing animal models of disease for the purpose of evaluating novel medical countermeasures to treat or prevent human infections. In 2015, Dr. Safronetz joined the Public Health Agency of Canada where he continues to serve as the Chief of Viral Zoonoses at the National Microbiology Laboratories in Winnipeg, Manitoba. His research interests continue to be the ecology, pathogenesis and prevention of emerging high consequence pathogens.

Timothy P. Sheahan, PhD

Throughout his career, Timothy has been working at the host pathogen interface to develop new methods of viral control. After receiving his bachelor's degree in Microbiology from the University of New Hampshire in 1999, he moved to Boston to try to make a career in music. After four years of playing music by night and working as a laboratory technician by day, he realized he enjoyed pipetting more than playing guitar. In 2003, he began graduate school at UNC Chapel Hill in the laboratory of Ralph Baric to pursue his interest in infectious disease. Since SARS coronavirus (SARS-CoV) had recently emerged from wild animals into humans, he focused his research on 1) understanding the molecular mechanisms guiding the emergence of SARS-CoV, 2) determining host factors contributing to disease and 3) evaluating therapeutic antibody and vaccine candidates to prevent future emergence. To expand his virological toolkit, he became a postdoctoral fellow with Charles Rice at the Rockefeller University in 2009. As a postdoc, he was interested in exploring host targeting for antiviral therapy. To this end, he developed near-single-cell approaches to chronicle the host response to hepatitis C virus in primary human hepatocytes revealing the importance of host genetics on the outcome of infection and host factors that could be targeted for antiviral therapy. After his postdoc, he wanted to gain insight into the process of antiviral drug discovery and became an Investigator at GlaxoSmithKline working to develop host targeting small molecules as antivirals. In 2015, he became faculty at UNC Chapel Hill where hes been working to develop broad-spectrum therapeutics against coronavirus with the goal of preventing future pandemic disease.



Pei-Yong Shi, PhD

Dr. Pei-Yong Shi has pioneered flavivirus drug discovery in leading pharmaceutical companies and academia. He has taken both target-based and cell-based antiviral approaches. Dr. Shi has published over 210 peer-reviewed articles and serves as Editor *(ACS Infectious Diseases, Journal of General Virology, and NPJ Vaccine)* and Editorial Board member *(Journal of Virology, Virology, and Antiviral Research)*. In addition to drug discovery, Dr. Shi's group is also highly active in studying flavivirus replication and vaccine development. He is internationally recognized for his scholar and administrative accomplishments at leading research institutions, the public health sector, and the pharmaceutical industry.

Christina Spiropoulou, PhD

Dr. Christina Spiropoulou is the Lead of the Molecular Pathogenesis and Therapeutics Team in the Viral Special Pathogens Branch (VSPB) at the U.S. Centers for Disease Control and Prevention (CDC). VSPB's mission includes the study of the viral hemorrhagic fever (VHF) viruses, a group of approximately 35 highly infectious viruses from 5 different virus families, many of which cause fatal hemorrhagic fever in humans. For the past 20 years, her research interests have focused on applied public health translational research extending from basic molecular biology, virus-host cell interactions, and host immune responses to the development of animal disease models, vaccines, and therapeutic platforms for high-consequence viruses, such as Ebola, Nipah, Crimean-Congo hemorrhagic fever, Rift Valley fever, Lassa, hantaviruses and tick-borne encephalitis viruses. Her current team's projects are focused on scientific questions that have the potential to lead to the development of prototype vaccines and identification of targets for the discovery of antivirals or immunotherapeutics to treat VHFs.

Priscilla L. Yang, PhD

Dr. Yang grew up in Pine Bluff, Arkansas as a child of the Clinton years. Matriculating at Yale College for her undergraduate education, she intended to major in the humanities until work-study employment in a laboratory and an unanticipated affinity for organic chemistry led her to switch majors. She went on to earn combined MS and BS degrees in Molecular Biophysics and Biochemistry in 1993 and subsequently earned her PhD in Bio-organic Chemistry with Dr. Peter G. Schultz in the College of Chemistry at the University of California, Berkeley. As a post-doctoral fellow with Dr. Francis V. Chisari at The Scripps Research Institute in La Jolla, Dr. Yang established the hydrodynamic injection murine model of hepatitis B virus replication that has become widely used as a tool for studying HBV-host interactions and for the evaluation of antivirals. In 2004, Dr. Yang established her own independent research group in the Department of Microbiology and Molecular Genetics at Harvard Medical School. There, her laboratory combines chemical and pharmacological approaches to address fundamental problems in virology.

In recent years, Dr. Yang's research efforts have centered on increasing the number of validated antiviral targets, identifying alternatives to combination therapy for avoiding antiviral resistance, and investigating the function of lipid membranes in RNA virus replication. Her work on antivirals has particularly focused upon dengue virus and other flavivirus pathogens that are major threats to public health and for which there are no approved therapies. Her group has identified the known drug 4-hydroxyphenylretinamide (4-HPR) as an inhibitor with high barrier to resistance targeting RNA replication of dengue, Zika, and other members of the *Flaviviridae* family *in cellulo* and in vivo, developed strategies for rapid identification of host-targeted covalent inhibitors as antivirals with broad-spectrum activity against diverse RNA viruses, investigated polypharmacology as an alternative to combination therapy for suppressing antiviral resistance, and established tools enabling the development of direct-acting antivirals targeting the flavivirus envelope protein.

Dr. Yang is currently Associate Professor in the Department of Microbiology and Immnobiology at Harvard Medical School and Associate Member at the Broad Institute of Harvard and MIT and the Dana-Farber/Harvard Cancer Center. She serves on the Editorial Advisory Boards for ACS Infectious Diseases and Molecular Biosystems and is a member of the NIH Drug Discovery and Mechanisms of Antimicrobial Resistance (DDR) Study Section. As an advocate for diversity in science and at Harvard, she has mentored female graduate students through the Harvard Graduate Women in Engineering and Science in addition to actively recruiting, mentoring and actively championed trainees from underrepresented backgrounds within her own laboratory as well as within the Harvard Graduate Programs in Chemical Biology, Biological and Biomedical Sciences, and Virology and the Harvard Summer Honors Undergraduate Research Program.

Program Schedule

Sunday May 21st, 2017

Interactive Workshop: Drug Discovery and Development 101

Chairs: **José Esté, PhD** and **Robert Buckheit, Jr., PhD**

WEST BALLROOM

2:00 PM – 4:00 PM

2:00 PM **177.** **Trials and Tribulations of Starting a Biotech Business – Entrepreneurship 101**
Robert Buckheit, Jr., PhD
ImQuest BioSciences, Frederick, Maryland, United States of America

2:30 PM **185.** **In Search of a Cure: A Scientist-Entrepreneur's Journey in Biotech**
Michael Sofia, PhD
Arbutus Biopharma, Doylestown, Pennsylvania, United States of America

3:00 PM **178.** **Creating Value by Protecting Intellectual Property**
Rebecca Kaufman, JD
King & Spalding, Atlanta, Georgia, United States of America

3:30 PM **169.** **Top Ten Steps to Start Up a Small Bio-Business and Sources of Funding**
Isaac R. Rodriguez-Chavez, PhD, MHS, MS
Science Consultant, Rockville, Maryland, United States of America

Coffee Break

GRAND BALLROOM PRE-FUNCTION

4:00 PM – 4:30 PM

Welcome and Keynote Addresses

Chair: **Johan Neyts, PhD**

WEST BALLROOM

4:30 PM – 6:30 PM

4:30 PM **Welcome**
José Esté, PhD
President, International Society for Antiviral Research

4:45 PM **165.** **Historical Context and Biological Enigma of Rhinovirus C**
Ann C. Palmenberg, PhD
Institute for Molecular Virology, University of Wisconsin-Madison

5:35 PM **167.** **Antivirals at the Interface with Public Health: A Case Study of Polio**
Mark A. Pallansch, PhD
Centers for Disease Control and Prevention, Atlanta, Georgia, United States of America

Opening Reception

CRYSTAL BALLROOM

6:30 PM – 8:30 PM

Monday May 22nd, 2017

Keynote Address

Chair: **José Esté, PhD**

WEST BALLROOM

9:00 AM – 9:50 AM

9:00 AM **179.** **Impact of Transmitted HIV Phenotype on Host-Virus Interactions and Disease Progression**
Eric Hunter, PhD
Emory University, Atlanta, Georgia, United States of America

Antiviral Immunity Symposium

Chairs: **Rhonda Cardin, PhD** and **Tomas Cihlar, PhD**

WEST BALLROOM

9:55 AM – 12:30 PM

9:55 AM **170.** **Small Molecule Inhibitors of Viral Entry: Pharmacological Mimicry of the Humoral Immune Response to Viral Infection**
Priscilla Yang, PhD
Harvard Medical School, Boston, Massachusetts, United States of America

10:30 AM **180.** **Targeting HIV Reservoirs for Stimulation and Elimination**
Jeff Murry, PhD
Gilead Sciences, Foster City, California, United States of America

11:00 – 11:30 AM **Coffee Break**

11:30 AM **171.** **Catch Me If You Can – Using HBV Immunity for Therapy**
Ulrike Protzer, MD
Institute of Virology, Technical University of Munich / Helmholtz Zentrum München, Munich, Bavaria, Germany

12:00 PM **181.** **Host factors as Potential Targets for Influenza Virus Antivirals**
Adolfo Garcia-Sastre, PhD
Icahn School of Medicine at Mount Sinai, New York, New York, United States of America

Lunch On Your Own

12:30 PM – 2:00 PM

Gertrude Elion Memorial Award Lecture

Chair: **José Esté, PhD**

WEST BALLROOM

2:00 PM – 2:45 PM

2:00 PM **164.** **Viral Hepatitis: The Search for a Cure**

Michael Sofia, PhD

Arbutus Biopharma, Doylestown, Pennsylvania, United States of America

Hepatitis Viruses and Retroviruses

Chair: **Tim Block, PhD** and **Ju-Tao Guo, MD**

WEST BALLROOM

2:50 PM – 5:00 PM

2:50 PM **Elsevier Awards**

Mike Bray, MD

Editor in Chief, Antiviral Research

2:55 PM **150.** **HBV RNaseH Inhibitors: Lack of Sensitivity to Viral Genetic Variation, Synergy with Approved and Experimental Drugs, and *in vivo* Efficacy in FRG Chimeric Mice**

Elena Lomonosova, PhD[1], Kelly Long, MS[2], Qilan Li, PhD[1], Nathan Ponzar, BS[1], Juan Villa, PhD[1], Ryan Murelli, PhD[3], John Bial, PhD[4], John Sagartz, DVM, PhD[2], **John Tavis, PhD**[1]

[1]Saint Louis University School of Medicine, St. Louis, Missouri, United States of America; [2]Seventh Wave Laboratories, St. Louis, Missouri, United States of America; [3]City University of New York, New York, New York, United States of America; [4]Yecuris, Inc., Tualatin, Oregon, United States of America

3:15 PM **137.** **Dual Activation of IFN and Pro-inflammatory Responses by TLR-agonists Leads to a Strong Inhibition of HBV Replication**

Julie Lucifora, PhD[1], Marc Bonnin, PhD[1], Sarah Maadadi, BS[1], Floriane Fusil, PhD[2], Laura Dimier, BS[1], Maud Michelet, MS[1], Océane Floriot, PhD[1], Anna Salvetti, PhD[1], Michel Rivoir, MD, PhD[3], Stephane Daffis, PhD[4], Simon Fletcher, PhD[4], François-Loïc Cosset, PhD[2], Fabien Zoulim, MD, PhD[5], **David Durantel, PhD**[1]

[1]Cancer Research Center of Lyon (CRCL), Inserm U1052, Lyon, France; [2]CIRI, INSERM U1111, Lyon, France; [3]CLB hospital, Inserm U1032, Lyon, France; [4]Gilead Sciences, Foster city, California, United States of America; [5]Hospices Civils de Lyon, CRCL, Inserm U1052, Lyon, France

3:30 – 4:00 PM **Coffee Break**

4:00 PM **139.** **Polo-like-kinase 1 is a Proviral Host-factor for Hepatitis B Virus Replication and a Potential Target for Combined Antiviral Strategies**

Adrien Foca, MS[1], Ahmed Diab, PhD[2], Floriane Fusil, PhD[3], Pascal Jalaguier, PhD[1], Nathalie Isorce, PhD[1], François-Loïc Cosset, PhD[3], Fabien Zoulim, MD, PhD[1], Ourania Andrisani, PhD[2], **David Durantel, PhD**[1]

[1]CRCL, Inserm U1052, Lyon, France; [2]Purdue University, West Lafayette, Indiana, United States of America; [3]CIRI, Inserm U1111, Lyon, France

4:15 PM **157.** **Discovery and Mechanistic Study Of Benzamide Derivatives that Modulate Hepatitis B Virus Capsid Assembly**

Shuo Wu, PhD, Qiong Zhao, PhD, John Kulp, PhD, Timothy Block, PhD, Yanming Du, PhD, Jinhong Chang, PhD, **Ju-Tao Guo, MD**

Baruch S. Blumberg Institute

4:30 PM **135. Activation of STING Mediates Antiviral Effects in a Mouse Model of Chronic Hepatitis B**
Emily Thi, PhD, Luying Pei, BS, Hui Huang, MD, PhD, Xin Ye, PhD, Agnes Jarosz, BS, Joseph Wasney, BS, Xiaowei Teng, PhD, Megan Fowler, BS, Shannon Tang, BS, Laurèn Bailey, PhD, Chris Moore, PhD, Rene Rijnbrand, PhD, Amy Lee, MS, Michael Sofia, PhD
Arbutus Biopharma, Burnaby, British Columbia, Canada

4:45 PM **144. ADAR1 Function Regulates Innate Immune Activation and HIV-1 Susceptibility in Primary Macrophages**
Maria Pujantell, MS, Eva Riveira-Muñoz, PhD, Roger Badia, PhD, Bonaventura Clotet, MD, PhD, José Esté, PhD, Ester Ballana, PhD
AIDS Research Institute – IrsiCaixa, Badalona, Barcelona, Spain

Poster Session 1
EAST BALLROOM
5:00 PM – 7:00 PM

1. Susceptibility of Two RNA Viruses of Public Health Significance to Selected Medicinal Plants of Nigerian Origin
Robert Obi, PhD[1], Juliet Shenge, PhD[2]
[1]Department of Microbiology, Federal University of Technology, Owerri, Imo State, Nigeria; [2]Department of Virology, University of Ibadan, Ibadan, Oyo State, Nigeria

3. Characterization of the Mode Of Antiviral Action of U0126, a MEK Inhibitor, Against Junín Virus
Jesús Brunetti, BS, Verónica Quintana, BS, Luis Scolaro, PhD, Viviana Castilla, PhD
Biochemistry Department, School of Science, University of Buenos Aires, Buenos Aires, Argentina

5. Development of Bipolymer based Novel Nanoparticles in Microsphere System as Vaccine Adjuvant
Saurabh Bhargava, MD, MPH[1], Vishal Bhargava, MD, MPH[2]
[1]Himalayan University, Kanpur, U.P., India; [2]GTB Hospital, Kanpur, U.P., India

7. Synthesis and Antiviral Activity of North and South L-Neplanocin Derivatives
Qi Chen, PhD[1], Amber Davidson, BS[1], Megan Stout, BS[1], Stewart Schneller, PhD[2]
[1]Slippery Rock Univeristy, Slippery Rock, Pennsylvania, United States of America; [2]Auburn University, Auburn, Alabama

9. Selective Inhibition of Hepatitis C Virus Replication by Alpha-zam, a *Nigella Sativa* Seed Formulation
Olufunmilayo Oyero, PhD[1], Masaaki Toyama, PhD[2], Naoki Mitsuhiro, DVM[2], Abdulfattah Onifade, PhD[1], Akemi Hidaka, BS[2], Mika Okamoto, MD, PhD[2], Masanori Baba, MD, PhD[2]
[1]University of Ibadan, Ibadan, Oyo, Nigeria; [2]Kagoshima University, Kagoshima, Kagoshima, Japan

11. CRISPR/Cas9-Mediated Antiviral Immunity Against Geminiviruses
Syed Shan e Ali Zaidi, PhD
Boyce Thompson Institute, Cornell University, Ithaca, NY, Ithaca, New York, United States of America

13. Synthesis of Flexible Purine Analogue Inhibitors of NCp7
Therese Ku, BS, Katherine Seley-Radtke, PhD
University of Maryland, Baltimore County

15. Development of a Rapid in Vitro Assay for Identification of Protein-protein Interactions in Zika virus

Shayli Varasteh Moradi, PhD, Dejan Gagoski, PhD, Wayne A. Johnston, PhD, Kirill Alexandrov, PhD

Institute for molecular biosciences, The University of Queensland, Brisbane

17. HIV-1 Tropism In Patients Living in Grodno Region of Belarus

Natallia Matsiyeuskaya, MD, PhD[1], Irina Tokunova, BS[1], Dmitry Kireev, PhD[2]

[1]State Medical University, Grodno, Belarus; [2]Central Research Institute for Epidemiology, Moscow, Russian Federation

19. Surface Modified Chitosan Nanoparticles for Selective Targeting of Lamivudine to Hepatocyte

Mani Bhargava, MD, MPH[1], Vishal Bhargava, PhD[2], Saurabh Bhargava, MD, MPH[3]

[1]ICFAI University, Knp, India; [2]GTB Hospital, India; [3]Himalayan University, India

21. Evaluation of Cross-strain Neutralizing Potency of Monoclonal Antibodies Against Crimean-Congo Hemorrhagic Fever Virus

Marko Zivcec, PhD, Lisa Guerrero, MS, Cesar Albarino, PhD, Eric Bergeron, PhD, Christina Spiropoulou, PhD

VSPB, DHCPP, NCEZID, CDC, Atlanta, Georgia, United States of America

23. Antibody Coated Liposomes for Transmucosal Vaccination

Sourabh Jain, MD, MPH[1], Saurabh Bhargava, MD, MPH[2]

[1]Bhagyodaya Pharmacy College, Sagar, M.P., India; [2]Himalayan University, Kanpur, u.p., India

25. Disulfiram Can Inhibit MERS and SARS Coronavirus Papain-Like Proteases via Different Modes

Min-Han Lin, BS, Chih-Hua Hsieh, BS, Chi-Yuan Chou, PhD

Department of Life Sciences, National Yang-Ming University, Taipei, Taiwan

27. Solid Lipid Based Nanoparticulate System for Effective Vaccine Delivery

Aakanchha Jain, PhD[1], Piush Khare, PhD[2], Saurabh Bhargava, MD, MPH[3]

[1]DOPS, Sagar, Sagar, M.P., India; [2]United Institute of Pharmacy, Allahabad, U.P., India; [3]Himalayan University, Kanpur, U.P., India

29. NonInvasive Topical Immunization Using Cholera Toxin as Adjuvant for the treatment of Hepatitis B

Gomed Agarwal, MD, MPH, Piush Khare, PhD

[1]United Institute of Pharmacy, Allahabad, India

31. Human Genetic Predisposition to Diseases Caused by Viruses from Flaviviridae Family

Andrey Barkhash, MD, Aida Romaschenko, PhD

Institute of Cytology and Genetics SB RAS, Novosibirsk, Russian Federation

33. A Proof-of-Concept Study for the Prevention of HIV Infection in 'Immune Humanized' Mice Using a Novel Non-Nucleoside Reverse Transcriptase Inhibitor

Ming-Tain Lai, PhD[1], Gary Adamson, PhD[1], Jaume Balsells-Padros, PhD[1], Michael Brehm, PhD[2], Christopher Bungard, PhD[1], Christopher Burgey, PhD[1], Smita Jaiswal, PhD[2], Christopher John, PhD[1], Bonnie Howell, PhD[1], Lee Klein, PhD[1], Maya Lipert, PhD[1], Jeremy Luban, PhD[2], Karsten Menzel, PhD[1], James Perkins, PhD[1], Elena Trepakova, PhD[1], Deping Wang, PhD[1], Steve Ludmerer, PhD[1]

[1]Merck & Co, West Point, Pennsylvania; [2]University of Massachusetts Medical School, Worcester, Massachusetts

35. **Pharmacologic Strategy Against Lassa Virus via Targeting Human Preprotein Convertase Site 1 Protease Upstream Glycoprotein Cleavage Pathway**

Olaposi Omotuyi, PhD[1], Nash Oyekanmi, PhD[2]
[1]Adekunle Ajasin University, Akungba-Akoko, Nigeria; [2]National Biotechnology Development Agency, Abuja, Nigeria

37. **Identification of the Target Regions Responsible for Resistance to Compound A, a Novel Antiviral Agent Against Dengue Virus**

Haruaki Nobori, MS[1], Shinsuke Toba, MS[1], Ryu Yoshida, PhD[1], Yasuko Orba, PhD[2], Hirofumi Sawa, MD, PhD[2], Akihiko Sato, DVM, PhD[1]
[1]SHIONOGI & CO., LTD. Drug discovery & disease research laboratory, Sapporo, Japan; [2]Hokkaido University Research Center for Zoonosis Control, Sapporo, Japan

39. **Systematic *In Vitro* Evaluation of Current and Experimental Hepatitis B Therapeutics: Potential Utility for Combinations Exploiting Multiple and Diverse Mechanisms of Action**

Andrea Cuconati, PhD[1], Andrzej Ardzinski, BS[1], Lauren Bailey, PhD[1], Nagraj Mani, PhD[1], Kim Stever, BS[1], Xiaohe Wang, MD[1], Amy Lee, MS[2], Chris Moore, PhD[1], Rene Rijnbrand, PhD[1], Michael Sofia, PhD[1]
[1]Arbutus Biopharma, Inc., Doylestown, Pennsylvania, United States of America; [2]Arbutus Biopharma Corp., Burnaby, British Columbia, Canada

41. **Synthesis and In-vitro Biological Activity of Novel Imine Derivatives**

Gholamreza Zarini, PhD
Faculty of Natural Science, University of Tabriz, Tabriz, Iran

43. **CD4 Signaling Induced Cytosolic Localization of Topoisomerase II Isoforms**

Sunnam Balakrishna, PhD, **Anand Kondapi, PhD**
University of Hyderabad, Hyderabad, Telangana, India

45. **Targeted Oral Delivery of HIV-1 Drug Combination (3TC + TNF + ATV/r) through Lactoferrin Nanoparticles**

Prashant Kumar, PhD, Yeruva Lakshmi, MS, **Anand Kondapi, PhD**
University of Hyderabad, Hyderabad, Telangana, India

47. **Cirsimaritininhibits Influenza A Virus Replication by Downregulating NF-κB Signal Transduction Pathway**

Haiyan Yan, MS, Huiqiang Wang, MS, Jinqiu Yin, BS, Shuo Wu, PhD, Danqing Song, PhD, Yuhuan Li, PhD
1 Tian Tan Xi Li,Beijing 100050,China

49. **Inhibition of Hepatitis B Virus Replication by N-hydroxyisoquinolinediones and Related Polyoxygenated Heterocycles**

Tiffany Edwardstc, MS[1], Elena Lomonosova, PhD[1], Jenny Patel, MS[1], Qilan Li, PhD[1], Fabrice Bailly, PhD[2], Philippe Cotelle, PhD[2], Erofili Giannakopoulou, MS[3], Grigoris Zoidis, PhD[3], Kelly Long, MS[4], John Sagartz, PhD[4], John Tavis, PhD[1]
[1]Saint Louis University, School of Medicine, St Louis, Missouri, United States of America; [2]University of Lille, Lille, France; [3]University of Athens, Athens, Greece; [4]Seventh Wave Laboratories, St. Louis, Missouri

51. Antiviral Potential of Fluorinated Analogs of Uracil in EBV-associated Cell System

Svitlana Zagorodnya, PhD[1], Krystyna Naumenko, MS[1], Anna Golovan, PhD[1], Galina Baranova, MS[1], Ganna Gudz, PhD[2], Yuriy Shermolovich, PhD[2]
[1]Zabolotny Institute of Microbiology and Virology of the NAS of Ukraine, Kyiv, Ukraine; [2]Institute of Organic Chemistry NAS of Ukraine, Kyiv, Ukraine

53. Development of Small and Specific Herpes Virus Inhibitors Based on Abalone Hemocyanin Proteins

Negar Talaei Zanjani, PhD[1], Monica Miranda-Saksena, PhD[2], Peter Valtchev, PhD[1], Russell J. Diefenbach J. Diefenbach, PhD[3], Jessica Yichen Zhong, PhD[4], Eve Diefenbach Diefenbach, PhD[2], Vincent G. Gomes, PhD[1], Joel P. Mackay, PhD[4], Anthony L. Cunningham, MD, PhD[2], Fariba Dehghani, PhD[1]
[1]School of Chemical and Biomolecular Engineering, The University of Sydney, Australia; [2]Centre for Virus Research, Westmead Millennium Institute for Medical Research, Sydney, Australia; [3]Department of Biomedical Sciences, Macquarie University, Australia; [4]School of Life and Environmental Sciences, The University of Sydney, Australia

55. Nucleoside Inhibitors of Tick-borne Encephalitis Virus: Structure-activity Relationships and Viral Resistance Study

Luděk Eyer, PhD[1], Radim Nencka, PhD[2], Daniel Růžek, PhD[1]
[1]Veterinary Research Institute, Department of Virology, Brno, Czech Republic; [2]Institute of Organic Chemistry and Biochemistry, The Czech Academy of Sciences, Prague, Czech Republic

57. Activity of SAMHD1 in Cycling Cells Permissive to HIV-1 Infection

Maria Pujantell, MS[1], Roger Badia, PhD[1], Javier Torres-Torronteras, PhD[2], Luis Menéndez-Arias, PhD[3], Ramón Martí, PhD[2], Albert Ruzo, PhD[4], Eduardo Pauls, PhD[1], Bonaventura Clotet, MD, PhD[1], Ester Ballana, PhD[1], José Esté, PhD[1], Eva Riveira-Muñoz, PhD[1]
[1]AIDS Research Institute – IrsiCaixa; [2]Vall d'Hebron Institut de Recerca; [3]Centro de Biología Molecular "Severo Ochoa"; [4]The Rockefeller University, New York

59. Butyrate Prodrugs of IHVR-19029 with Enhanced Oral Exposure and Prevention of Gastrointestinal Glucosidase Interaction

Yanming Du, PhD, Jia Guo, PhD, Fang Guo, MD, PhD, Julia Ma, BS, Xuexiang Zhang, MS, Qing Su, PhD, Nicky Hwang, BS, Ju-Tao Guo, MD, Timothy Block, PhD, Jinhong Chang, MD, PhD
Baruch S. Blumberg Institute, Doylestown, Pennsylvania, United States of America

61. Discovery and Characterization of Broad-spectrum Inhibitors of Coronaviruses

Kin Kui LAI, MS, Jun DAI, PhD, Kwok Yung YUEN, MD, Richard Yi Tsun KAO, PhD
Department of Microbiology, Li Ka Shing Faculty of Medicine, The University of Hong Kong, Hong Kong

63. Evaluation of the Innate Immune Modulator Acitretin as a Novel Strategy to Clear HIV Reservoir

Edurne Garcia-Vidal, MS, Maria Pujantell, MS, Roger Badia, PhD, Bonaventura Clotet, MD, PhD, Eva Riveira-Muñoz, PhD, Ester Ballana, PhD, José Esté, PhD
Irsicaixa – AIDS research institute, Badalona, Spain

65. Kinetic, Structural and Thermodynamic Analysis of the H275Y, I223V and S247N Neuraminidase Resistant Mutants of H1N1 2009 Pandemic Influenza Virus

Milan Kožíšek, PhD, Jana Pokorná, PhD, Petr Pachl, PhD, Pavlína Řezáčová, PhD, Aleš Machara, PhD, Jakub Hejdánek, BS, Elena Karlukova, BS, Jan Konvalinka, PhD
Institute of Organic Chemistry and Biochemistry of the Czech Academy of Sciences, Prague, Czech Republic

67. High-Throughput Screening to Identify Inhibitors of BSL-4 Viruses

Mike Flint, PhD, Payel Chatterjee, MS, Stephen Welch, PhD, Michael Lo, PhD, Laura McMullan, PhD, Eric Bergeron, PhD, Cesar Albarino, PhD, Stuart Nichol, PhD, Christina Spiropoulou, PhD
Viral Special Pathogens Branch, Centers for Disease Control and Prevention, Atlanta, Georgia, United States of America

69. Using a Recombinant Crimean-Congo Hemorrhagic Fever Virus Expressing a Fluorescent Protein to Rapidly Evaluate Synergistic Properties of Antiviral Compounds

Stephen Welch, PhD, Florine Scholte, PhD, Mike Flint, PhD, Éric Bergeron, PhD, Christina Spriropoulou, PhD
Centers for Disease Control and Prevention, Atlanta, Georgia, United States of America

71. Anti-Dengue Activity of Traditional Chinese Medicinal Plants

Maqsood Maryam, MS[1], Kian Keong Te, PhD[2], Fai Chu Wong, PhD[3], Tsun Thai Chai, PhD[3], Seng Chiew Gan, PhD[2], Gary Low, PhD[2], Hui Yee Chee, PhD[1]
[1]Faculty of Medicine and Health Sciences, University Putra Malaysia, Serdang, selangor, Malaysia; [2]Faculty of Medicine and Health Sciences, University Tunku Abdul Rahman, Sungai Long, Selangor, Malaysia; [3]Department of Chemical Science, Faculty of Science, University Tunku Abdul Rahman, Kampar, Perak, Malaysia

73. Lupeol and Other Compounds from Natural Sources Exhibited Antiviral Activity Against Enteroviruses 7, 13 and 19

Omonike Ogbole, PhD[1], Abidemi Sunmola, BS[2], Adekunle Adeniji, PhD[3]
[1]Department of Pharmacognosy, Faculty of Pharmacy, University of Ibadan, Ibadan, Nigeria; [2]Department of Pharmaceutical Microbiology, Faculty of Pharmacy, University of Ibadan, Ibadan, Nigeria; [3]Department of Virology, College of Medicine, University of Ibadan, Ibadan, Nigeria

75. Synthesis of γ-modified Nucleoside Triphosphates

Simon Weising, MS[1], Dominique Schols, PhD[2], Jan Balzarini, PhD[2], Chris Meier, PhD[1]
[1]Organic Chemistry, Department of Chemistry, University of Hamburg, Hamburg, Germany; [2]Rega Institute for Medical Research, Katholieke Universiteit Leuven, Leuven, Belgium
[1]University of Cagliari, Cagliari, Italy; [2]University of Magna Graecia

77. Novel Cell Targeting L-ddBCNA Antiviral Inhibits Autophagy in Virus Infected Cells

Rohan Narayan, MS[1], Laura Farleigh, PhD[2], Alina Tscherne, MS[3], Daniela Friese, BS[3], Ed Sayers, PhD[4], Arwyn Jones, PhD[4], **Joachim Bugert, MD, PhD**[3]
[1]Cardiff School of Pharmacy & Pharmaceutical Sciences, Cardiff, Wales, United Kingdom; [2]University of Cambridge, Cambridge, United Kingdom; [3]Institut für Mikrobiologie der Bundeswehr, München, Bavaria, Germany; [4]Cardiff School of Pharmacy & Pharmaceutical Sciences, Cardiff, Wales, United Kingdom

79. Synthesis of Heterocycles Targeting the Inhibition of DNA Glycosylases Involved in Base Excision Repair Pathway

Zahira Tber, PhD[1], Charlotte Rieux, MS[2], Franck Coste, PhD[2], Norbert Garnier, PhD[2], Bertrand Castaing, PhD[2], Vincent ROY, PhD[1], **Luigi Agrofoglio, PhD**[1]
[1]ICOA UMR CNRS 7311 – Universite Orleans, Orleans, France; [2]CBM UPR CNRS 4301, Orleans, France

81. 25-Hydroxycholesterol Inhibition of Lassa Virus Infection through Aberrant GP1 Glycosylation

Punya Shrivastava-Ranjan, PhD, Eric Bergeron, PhD, Ayan Chakrabarti, MS, César Albariño, PhD, Mike Flint, PhD, Stuart Nichol, PhD, Christina Spiropoulou, PhD
Centre for Disease Control and Prevention, Atlanta, Georgia, United States of America

83. **Identification of PIK-III as a Novel Antiviral Against Pathogens Entering Cells by Macropinocytosis**

Olena Shtanko, PhD, Robert Davey, PhD
Texas Biomedical Research Institute, San Antonio, Texas, United States of America

85. **Mechanistic Analysis of Benzoannulene Alphavirus Inhibitors**

Nicole Haese, PhD[1], Kaleem Ahmed, PhD[2], Clayton Morrison, PhD[3], Wes Sanders, PhD[3], Nathaniel Moorman, PhD[3], Nicholas May, BS[4], Vibha Pathak, PhD[2], Corinne Augelli-Szafran, Ph.D[2], Mark Suto, PhD[2], Victor DeFilippis, PhD[1], Thomas Morrison, PhD[4], Mark Heise, PhD[3], Ashish Pathak, PhD[2], Daniel Streblow, PhD[1]
[1]Vaccine & Gene Therapy Institute, Oregon Health & Science Univeristy; [2]Southern Research; [3]University of North Carolina at Chapel Hill; [4]University of Colorado School of Medicine

87. **Probing Membrane Lysis of Individual Virus Particles Induced by an Amphipathic Peptide and Correlations with Antiviral Activity**

Nam-Joon Cho, PhD, Joshua Jackman, PhD
School of Materials Science and Engineering, Nanyang Technological University, Singapore

89. **A Novel Mutation in N1 Neuraminidase Confers Resistance to Multiple Neuraminidase Inhibitors Without Impacting Viral Fitness**

Jin Jung Kwon, BS[1], Won-suk Choi, MS[1], Ju Hwan Jeong, BS[1], Ji Won Han, BS[1], Su Jeong Ahn, BS[1], Hyeok-il Kwon, PhD[1], Eun-Ha Kim, PhD[1], Sun-Woo Yoon, PhD[2], Young Ki Choi, PhD[1], Yun Hee Baek, PhD[1], Min Suk Song, PhD[1]
[1]College of Medicine and Medical Research Institute, Chungbuk National University, Cheongju, Korea, Republic of; [2]Viral Infectious Disease Research Center

91. **Anti-influenza Activity of Plant Flavonoids – di- and tetramethoxy-quercetin Derivatives**

D. Starosyla, PhD[1], M. Platonov, PhD[2], O. Vacylchenko, PhD[2], L. Palchykovska, PhD[2], S. Zagorodnya, PhD[3], S. Dsadiun, PhD[1], S. Rybalko, MD[1], L. Varbanets, PhD[3], V. Atamanyuk, PhD[1]
[1]Gromashevsky L.V. Institute of Epidemiology and Infection Diseases, NAMS of Ukraine, Kyiv, Ukraine; [2]Institute of Molecular Biology and Genetics, NAS of Ukraine, Kyiv, Ukraine; [3]D.K.Zabolotny Institute of Microbiology and Virology NAS of Ukraine, Kyiv, Ukraine

93. **Multiple Effects of Toxins Isolated from *Crotalus durissus terrificus* on the Hepatitis C Virus Life Cycle**

Jacqueline Shimizu, MS[1], Cintia Bittar, PhD[2], Mariana Batista, PhD[2], Guilherme Campos, MS[2], Suely Silva, BS[3], Adélia Cristina Silva, PhD[4], Suely Vilela, PhD[4], Victor Hugo Quintana, PhD[4], Paula Rahal, PhD[2], **Ana Carolina Jardim, PhD**[3]
[1]Institute of Biomedical Science, Federal University of Uberlandia; [2]Sao Paulo State University; [3]Federal University of Uberlandia; [4]University of Sao Paulo

95. **Broad-Spectrum Antiviral Molecules with Biophysical Mechanisms of Action**

Sietske Speerstra, BS[1], Alexey Chistov, BS[2], Gleb Proskurin, BS[2], Vladimir Korshun, PhD[2], Luis Schang, DVM, PhD[3]
[1]Department of Biochemistry, University of Alberta, Edmonton, Alberta, Canada; [2]Shemyakin-Ovchinnikov Institute of Bioorganic Chemistry, Moscow, Russian Federation; [3]Baker Institute, Cornell University & Dept. of Biochemistry, University of Alberta, Ithaca, New York, United States of America

97. Membrane-permeable Nucleoside Triphosphate-prodrugs against Hepatitis C Virus
Matthias Winkler, MS[1], Elena Vedove, MS[2], Chris Meier, PhD[1]
[1]University of Hamburg, Hamburg, Germany; [2]University of Camerino, Camerino, Italy

99. The STING Agonist SB 11285 is a Broad-spectrum Antiviral Agent
Cybele Garcia, PhD[1], José Peña Carcamo, PhD[1], Maria Morell, MS[1], Sandra Cordo, PhD[1], Sreerupa Challa, PhD[2], Shenghua Zhou, PhD[2], Anjaneyulu Sheri, PhD[2], Seetharamaiyer Padmanabhan, PhD[2], Geeta Meher, PhD[2], Diane Schmidt, PhD[2], Niraj Shil, PhD[3], Meleri Jones, PhD[3], Graham Foster, PhD[3], Santanu Bose, PhD[3], Nezam Afdhal, PhD[2], Brent Korba, PhD[5], Radhakrishnan Iyer, PhD[2]
[1]Lab Estrategias Antivirales, Bioquímica y Biología del virus Junín, Univ Buenos Aires, Buenos Aires, Argentina; [2]Spring Bank Pharmaceuticals, Inc., Milford, MA 01757, Massachusetts, United States of America; [3]Washington State University, Pullman, Washington, United States of America; [4]The Liver Unit, Blizard Institute, Barts Health, Queen Mary University of London, London, United Kingdom; [5]Georgetown University Medical Center, Division of Molecular Virology and Immunology, Washington DC, District of Columbia, United States of America

101. Comparison Between Various Strains of Chikungunya in Disease Phenotype and Response to Antiviral Treatment
Makda Gebre, BS, Justin Julander, PhD
[1]Institute for Antiviral Research, Utah State University, Logan, Utah, United States of America

103. Influenza A Inhibitors Based on Copper Binding to a Highly Conserved Histidine
Nathan Gordon, MS[1], Kelly McGuire, BS[2], Spencer Wallentine, BS[2], Gregory Mohl, BS[2], **Mackenzie Hart, BS**[2], Jonathan Lynch, BS[2], Roger Harrison, PhD[2], David Busath, MD[2]
[1]Dept. of PDBio, Provo, Utah, United States of America; [2]Brigham Young University, Provo, Utah, United States of America

105. Role of RSV Polymerase in the Antiviral Effect of Ribavirin
Jerome Deval, PhD, Amy Fung, MS, Jia Meng, PhD, Andreas Jekle, PhD, Guangyi Wang, PhD, Natalia Dyatkina, PhD, Marija Prhavc, PhD, Julian Symons, PhD, Leo Beigelman, PhD
Alios BioPharma

107. Identification of small molecule inhibitors of Ebola virus replication
Priya Luthra, PhD[1], Jue Liang, PhD[2], Colette Pietzch, MS[3], Sudip Khadka, PhD[1], Sampriti De, MS[1], Alexander Bukreyev, PhD[3], Joseph Ready, PhD[2], Christopher Basler, PhD[1]
[1]Georgia State University; [2]UT Southwestern; [3]University of Texas Medical Branch

109. Successful Design of Ribonucleoside Di- and Triphosphate Prodrugs to Improve the Anti-Influenza Virus Activity of T-705 and its Analogue T-1105
Evelien Vanderlinden, PhD[1], Johanna Huchting, PhD[2], Chris Meier, PhD[2], Lieve Naesens, PhD[1]
[1]Rega Institute for Medical Research, Leuven, Belgium; [2]Institute of Organic Chemistry, Hamburg University, Hamburg, Germany

111. Vaccinating Effect of Attenuated Zika Virus Candidates in a Lethal Mouse Model
Justin Julander, PhD[1], Steffen Mueller, PhD[2], Skot Neilson, MS[1], J. Robert Coleman, PhD[2]
[1]Institute for Antiviral Research, Utah State University, Logan, Utah, United States of America; [2]Codagenix Inc., Farmingdale, New York, United States of America

113. Suramin Inhibits Zika and Chikungunya Virus Replication by Interfering with Attachment and Later Steps of the Replication Cycle

Irina Albulescu, MS[1], Kristina Kovacikova, MS[1], Ali Tas, BS[1], Tabitha Hoornweg, PhD[2], Salvatore Ferla, PhD[3], Andrea Brancale, PhD[3], Jolanda Smit, PhD[2], Eric Snijder, PhD[1], **Martijn van Hemert, PhD**[1]

[1]Leiden University Medical Center, Leiden, Netherlands; [2]University Medical Center Groningen, Groningen, Netherlands; [3]Cardiff University, Cardiff, United Kingdom

115. Screening of FDA Approved Compounds Library Targeting the mRNA Capping of *Venezuelan equine encephalitis virus* (VEEV)

Ana S. Ramos, MS[1], Changqing Li, PhD[1], Eydoux Cécilia, PhD[1], Aouadi Wahiba, MS[1], Martin Baptiste, MS[1], Contreras Jean Marie, PhD[2], Morice Christophe, PhD[2], Jung Marie-Louise, PhD[2], Bruno Canard, PhD[1], Guillemot Jean-Claude, PhD[1], Decroly Etienne, PhD[1], Coutard Bruno, PhD[1]

[1]Aix Marseille Université, CNRS, AFMB UMR 7257, Marseille, France; [2]Prestwick Chemical, 67400 ILLKIRCH – Strasbourg – France

117. VIRIP – an Anti-HIV Host Peptide Output Hypothesis

Aitsana A. Maslakova[1], Vera S. Efimova[1], Alexei S. Maslakov[1], Victor E. Spangenberg[2], Mikhail A. Rubtsov[1], Igor V. Orlovsky[3]

[1] Biology Department, Lomonosov MSU, Moscow, Russia; [2] Vavilov Institute of General Genetics, Moscow, Russia; [3] Lomonosov MSU A.N. Belozersky Research Institute of Physical and Chemical Biology, Moscow, Russia

119. Evaluate the Broad-spectrum Protective Efficacy of *Lactobacillus* Species Against Influenza Viruses in Mice

Jung-hoon Kwon, DVM, Seong-Su Yuk, DVM, Sol Jeong, DVM, Jei-hyun Jeong, DVM, Ji-ho Lee, DVM, Jun-beom Kim, DVM, Yu-jin Kim, DVM, Chang-Seon Song, DVM, PhD

College of Veterinary Medicine, Konkuk University, Seoul, Korea

121. Baicalin as an *in vitro* Inhibitor for Chikungunya Virus

Adrian Oo, MS[1], Stephen Higgs, PhD[2], Sazaly AbuBakar, PhD[3], **Keivan Zandi, PhD**[4]

[1]Tropical Infectious Disease Research and Education Center, Faculty of Medicine, UM, Kuala Lumpur, KL, Malaysia; [2]Kansas State University; [3]Tropical Infectious Disease Research and Education Center, UM, Kuala Lumpur, WP, Malaysia; [4]1- TIDREC, UM, Malaysia 2- Emory University, Atlanta, USA, Atlanta, Georgia, United States of America

123. Exploring Polypharmacology for the Design of Broad-spectrum Influenza Antivirals

Jun Wang, PhD

Assistant Professor Department of Pharmacology and Toxicology University of Arizona, Tucson, Arizona, United States of America

125. Sensitivity to a Potent Lassa Antiviral is Modulated by a Virulence Determinant

Sean Amberg, PhD[1], Ikenna Madu, PhD[1], Megan Files, BS[1], Tiffany Huelar, BS[1], Kie-Hoon Jung, PhD[2], Brian Gowen, PhD[2], Shawn Iadonato, PhD[1], Kristin Bedard, PhD[1]

[1]Kineta, Seattle, Washington, United States of America; [2]Utah State University, Logan, Utah, United States of America

127. Evaluation of Ribavirin Against Recombinant Oncolytic Newcastle Disease Virus Replication in vitro

Weijia Wang, MS, Xing Cheng, MS, Udaya Rangaswamy, PhD, Hong Jin, PhD

Medimmune

129. **Human ex vivo Lung Tissue as Model System to Investigate Novel Host-directed Antivirals Against Influenza A Virus**

Jessica von Recum-Knepper, PhD[1], Thorsten Wolff, PhD[2], Silke Stertz, PhD[3], Torsten Steinmetzer, PhD[4], Eva Böttcher-Friebertshäuser, PhD[5], Sumit Chanda, PhD[6]
[1]University of California, San Diego, School of Medicine, La Jolla, California, USA, California, United States of America; [2]Influenza Viruses & Other Respiratory Viruses, Robert Koch Institute, Berlin, Germany; [3]Institute of Medical Virology, University of Zurich, Switzerland; [4]Institute of Pharmaceutical Chemistry, Philipps University Marburg, Germany; [5]Institute of Virology, Philipps University Marburg, Germany; [6]Sanford Burnham Prebys Medical Discovery Institute, La Jolla, USA

Tuesday May 23rd, 2017

Antonin Holý Memorial Award Lecture

Chair: **José Esté, PhD**

WEST BALLROOM

9:00 AM – 9:30 AM

9:05 AM **175.** **Nucleosides: A Rich Source of Antiviral Agents**

Chung K (David) Chu, PhD
University of Georgia, Athens, Georgia, United States of America

Emerging Viruses

Chairs: **Johan Neyts, PhD** and **Leen Delang, PhD**

WEST BALLROOM

9:30 AM – 12:30 PM

9:30 AM **155.** **The Nucleoside Prodrug GS-5734 Inhibits Multiple Coronaviruses and Selects for Resistance Mutations in the RNA-Dependent RNA Polymerase that are Associated with a Decrease in Viral Replication Fitness**

Maria Agostini, BS[1], Erica Andres, BS[1], Xiaotao Lu, BS[1], Amy Sims, PhD[2], Rachel Graham, PhD[2], Timothy Sheahan, PhD[2], Everett Smith, PhD[3], James Case, BS[1], Joy Feng, PhD[4], Robert Jordan, PhD[4], Adrian Ray, PhD[4], Tomas Cihlar, PhD[4], Dustin Siegel, PhD[4], Richard Mackman, PhD[4], Michael Clarke, PhD[4], Ralph Baric, PhD[2], Mark Denison, MD[1]
[1]Vanderbilt University Medical Center, Nashville, Tennessee; [2]University of North Carolina at Chapel Hill, Chapel Hill, North Carolina; [3]The University of the South, Sewanee, Tennessee; [4]Gilead Sciences, Inc., Foster City, California

9:45 AM **154.** **Inhibiting Viral RNA Replication and Enhancing Host Cellular Cytokine Response: Unique Dual Effects of a Benzodiazepine Yellow Fever Virus (YFV) NS4B Inhibitor**

Xuexiang Zhang, MS, Shuo Wu, PhD, Julia Ma, BS, Fang Guo, MD, PhD, Yanming Du, PhD, Timothy Block, PhD, Ju-Tao Guo, MD, **Jinhong Chang, MD, PhD**
Baruch S. Blumberg Institute

10:00 AM **142. Structural and Enzymatic Studies on Zika Virus NS2B-NS3 Protease**
Linlin Zhang, PhD, Yasmin Gül, MS, Jian Lei, PhD, Rolf Hilgenfeld, PhD
Institute of Biochemistry, University of Lübeck, Lübeck, Germany

10:15 AM **111. Vaccinating Effect of Attenuated Zika Virus Candidates in a Lethal Mouse Model**
Justin Julander, PhD[1], Steffen Mueller, PhD[2], Skot Neilson, MS[1], J. Robert Coleman, PhD[2]
[1]Institute for Antiviral Research, Utah State University, Logan, Utah, United States of America; [2]Codagenix Inc., Farmingdale, New York, United States of America

10:25 AM **60. Development of a Model for Enterovirus D68 Infection in Mice**
Bart Tarbet, PhD, Brett Hurst, MS, Joseph Evans, BS
Institute for Antiviral Research, Utah State University, Logan, Utah, United States of America

10:35 – 11:00 AM **Coffee Break**

11:00 AM **140. Recombinant, Fully-Human mAbs Have Potent Therapeutic Activity in Murine Models of Chikungunya Virus Disease**
Marie Mandron, PhD[1], Jonathan Rothblatt, PhD[2], Pierre Cortez, PhD[1], Xavier Marniquet, PhD[1], Heather Hughes, PhD[3], Jooyun Lee, MD[4], Nicole Haese, PhD[5], Skot Neilson, PhD[6], Rebecca Broeckel, PhD[5], Gopal Sapparapu, PhD[7], Anna Park, PhD[3], Julie Bird, PhD[3], Cendrine Lemoine, PhD[1], Catherine Devaud, PhD[8], Anne Caron, PhD[8], Soila Sukupolvi, PhD[9], Julie Fox, PhD[9], Daniel Streblow, PhD[5], Justin Julander, PhD[6], Michael Diamond, MD, PhD[9], James Crowe, MD[7], **Kara Carter, PhD**[2]
[1]Sanofi, Marcy L'Etoile, France; [2]Sanofi, Cambridge, Massachusetts, United States of America; [3]Sanofi, Framingham, Massachusetts, United States of America; [4]Sanofi, Bridgewater, New Jersey, United States of America; [5]Oregon Health Sciences University, Beaverton, Oregon, United States of America; [6]Utah State University, Logan, Utah, United States of America; [7]Vanderbilt University, Nashville, Tennessee, United States of America; [8]Sanofi, Paris, France; [9]Washington University in St Louis, St Louis, Missouri, United States of America

11:15 AM **138. Two Birds with One Stone: Nucleoside Polymerase Inhibitors that Block the Replication of Both Noro- and Rotaviruses, the Two Main Etiological Agents of Viral Diarrhea**
Jana Van Dycke, MS[1], Justine Vandepoele, MS[1], Guido Papa, MS[2], Francesca Arnoldi, PhD[2], Oscar Burrone, PhD[2], Johan Neyts, PhD[1], **Joana Rocha-Pereira, PhD**[1]
[1]Rega Institute for Medical Research, KU Leuven, Leuven, Belgium; [2]International Centre for Genetic Engineering and Biotechnology, Trieste, Italy

11:30 AM **163. Antiviral Activity and Mechanism of Action of Site-1 Protease (S1P) Inhibitor on Crimean-Congo Hemorrhagic Fever Virus**
Éric Bergeron, PhD, Stephen Welch, PhD, Mike Flint, PhD, Stuart Nichol, PhD, Christina Spiropoulou, PhD
Centers for Disease Control and Prevention

11:40 AM **152. Engineering Approaches to Combat Infectious Diseases: An Example of Broad-Spectrum Antiviral Peptides**
Nam-Joon Cho, PhD
School of Materials Science and Engineering, Nanyang Technological University, Singapore

11:50 AM **141. A Novel Agonist of the TRIF Pathway Induces a Cellular State Refractory to Replication of Zika, Chikungunya, and Dengue Viruses**

Kara Pryke, BS[1], Jinu Abraham, PhD[1], Tina Sali, PhD[1], Bryan Gall, PhD[1], Daniel Streblow, PhD[1], Alec Hirsch, PhD[1], Marita Chakhtoura, PhD[2], Elias Haddad, PhD[2], Jessica Smith, PhD[1], **Victor DeFilippis, PhD**[1]

[1]Oregon Health and Science University, Portland, Oregon, United States of America; [2]Drexel University, Philadelphia, Pennsylvania, United States of America

12:00 PM **132. Apilimod, a PIKfyve Inhibitor with Antiviral Activity against Ebola and Marburg Viruses**

Julie Dyall, PhD[1], Elizabeth Nelson, PhD[2], Thomas Hoenen, PhD[3], Alyson Barnes, PhD[2], Huanying Zhou, MS[1], Janie Liang, MS[1], Julia Michelotti, PhD[1], William Dewey, MS[1], Lisa Evans Dewald, PhD[1], Richard Bennett, PhD[1], Patrick Morris, PhD[4], Rajarshi Guha, PhD[4], Carleen Klumpp-Thomas, PhD[4], Crystal McKnight, PhD[4], Yu-Chi Chen, PhD[4], Craig Thomas, PhD[4], Scott Martin, PhD[4], Peter Jahrling, PhD[5], Lisa Hensley, PhD[1], Gene Olinger, PhD[1], Judith White, PhD[2]

[1]Integrated Research Facility, NIAID, NIH, Frederick, Maryland, United States of America; [2]University of Virginia, Charlottesville, Virginia, United States of America; [3]Laboratory of Virology, Division of Intramural Research, NIH, Hamilton, Montana, United States of America; [4]National Center for Advancing Translational Sciences, National Institutes of Health, Bethesda, Maryland, United States of America; [5]Integrated Research Facility and Emerging Viral Pathogens Section, NIAID, NIH, Frederick, Maryland, United States of America

12:10 PM **159. Can Antiviral Drug-resistant Chikungunya Viruses be Transmitted by Mosquitoes?**

Leen Delang, PhD[1], Pei-Shi Yen, MS[2], Marie Vazeille, PhD[2], Anna-Bella Failloux, PhD[2]

[1]Rega Institute for Medical Research, KU Leuven, Leuven, Belgium; [2]URE Arboviruses and Insect vectors, Institut Pasteur, Paris, Paris, France

12:20 PM **134. Identification of Novel Amodiaquine Derivatives as Anti-Ebola Virus Compounds**

Yasuteru Sakurai, PhD[1], Norikazu Sakakibara, PhD[2], Masaaki Toyama, PhD[3], Masanori Baba, MD[3], Robert Davey, PhD[1]

[1]Texas Biomedical Research Institute, San Antonio, Texas, United States of America; [2]Tokushima Bunri University, Sanuki, Tokushima, Japan; [3]Kagoshima University, Kagoshima, Kagoshima, Japan

Lunch On Your Own

12:30 PM – 2:00 PM

New Member and First Time Attendee Networking

ROOM 204/205

12:30 PM – 2:00 PM

Career Development Panel

ROOM 203

2:00 PM – 3:00 PM

Wednesday May 24th, 2017

Shotgun Oral Presentations

Chairs: **Katherine Seley-Radtke, PhD** and **Therese Ku, BS**

WEST BALLROOM

8:30 AM – 9:00 AM

Speakers nominated by the Poster Award Committee

Keynote Address: Pei-Yong Shi, PhD

Chair: **Johan Neyts, PhD**

WEST BALLROOM

9:00 AM – 9:50 AM

9:00 AM **182.** **Zika Antiviral and Vaccine Development**
Pei-Yong Shi, PhD
University of Texas Medical Branch, Galveston, Texas, United States of America

Emerging Infections Symposium

Chairs: **Pei-Yong Shi, PhD** and **Christina Spiropoulou, PhD**

WEST BALLROOM

9:50 AM – 12:30 PM

9:50 AM **173.** **From Basic Science to Promising Antivirals for Hemorrhagic Fever Viruses**
Christina Spiropoulou, PhD
Centers for Disease Control and Prevention, Atlanta, Georgia, United States of America

10:25 AM **184.** **Treatment and Prevention of Emerging Rodent-Borne Viruses**
David Safronetz, PhD
University of Manitoba, Winnipeg, Manitoba, Canada

11:00 – 11:30 AM **Coffee Break**

11:30 AM **174.** **Flaviviral Translation**
Mariano Garcia-Blanco, MD, PhD
University of Texas Medical Branch

12:00 PM **172.** **Broad-spectrum Antivirals to Prevent Emerging Coronavirus Pandemic Disease**
Timothy Sheahan, PhD
University of North Carolina at Chapel Hill, Chapel Hill, North Carolina, United States of America

Lunch On Your Own

12:30 PM – 1:45 PM

ISAR Business Meeting

President: **José Esté, PhD**
Treasurer: **Brian Gowen, PhD**
Secretary: **Graciela Andrei, PhD**
WEST BALLROOM
1:45 PM – 2:00 PM

William Prusoff Young Investigator Award Lecture

Chair: **José Esté, PhD**
WEST BALLROOM
2:00 PM – 2:30 PM

2:00 PM **176.** **Collaborating in Drug Discovery: Challenges and Solutions**
Maaike Everts, PhD[1]
[1]University of Alabama at Birmingham, Birmingham, Alabama, United States of America

Respiratory Viruses

Chairs: **Bart Tarbet, PhD** and **Jia Meng, PhD**
WEST BALLROOM
2:30 PM – 3:30 PM

2:30 PM **156.** **Repurposing Kinase Inhibitors Against Influenza – The Clinically Approved MEK Inhibitor Trametinib Efficiently Blocks IAV Propagation and Limits Hyperexpression of Cytokines**
Tobias Schräder, PhD[1], Sabine Dudek, PhD[1], Christina Ehrhardt, MD, PhD[1], Oliver Planz, PhD[2], Stephan Ludwig, PhD[1]
[1]Institute of Virology (IVM) Westfaelische Wilhelms-University Muenster, Muenster, Germany; [2]Interfaculty Institute for Cell Biology, University of Tuebingen, Germany, Tuebingen

2:45 PM **131.** **Inhibition of Nuclear Export by Verdinexor May Enhance a Broad Therapeutic Window in Mouse Models of Influenza**
Sharon Tamir, PhD[1], Shelton Cochran, BS[1], Patricia Jorquera, PhD[2], Jennifer Pickens, PhD[2], Sharon Shacham, PhD[1], Margaret Lee, PhD[1], Ralph Tripp, PhD[2]
[1]Karyopharm Therapeutics, Newton, Massachusetts, United States of America; [2]Department of Infectious Diseases, University of Georgia, Athens, Georgia, United States of America

3:00 PM **160.** **Development of ALS-8112/ALS-8176 as an Effective Replication Inhibitor of Human Metapneumovirus**
Jia Meng, PhD, Jerome Deval, PhD, Andreas Jekle, PhD, Julian Symons, PhD
Alios BioPharma., Inc, part of the Janssen Pharmaceutical Companies, South San Francisco, California, United States of America

3:13 PM **136.** **Tricyclic Matrinic Derivative DXC-10 Inhibits Influenza A Virus Replication Before the Nucleoprotein Nucleus Entry**
Jinqiu Yin, BS, Haiyan Yan, MS, Huiqiang Wang, MS, Shuo Wu, PhD, Danqing Song, PhD, Yuhuan Li, PhD
Institute of Medicinal Biotechnology, Chinese Academy of Medical Sciences & Peking Union Medical College, Beijing 100050, China

Poster Session 2

EAST BALLROOM

3:30 PM – 5:30 PM

2. Griffithsin, a Novel Inhibitor of Henipavirus Entry and Fusion
Michael Lo, PhD[1], Barry O'Keefe, PhD[2], Anasuya Chattopadhyay, PhD[3], Anne Hotard, PhD[1], Lauren Haugh Krumpe, MS[2], John Rose, PhD[3], Stuart Nichol, PhD[1], Christina Spiropoulou, PhD[1]
[1]Centers for Disease Control and Prevention, Atlanta, Georgia, United States of America; [2]National Cancer Institute, Frederick, Maryland, United States of America; [3]Yale University School of Medicine, New Haven, Connecticut, United States of America

4. Discovery and Development of Novel Non-catalytic Site HIV Integrase Inhibitors
Kyungjin Kim, PhD[1], Uk-Il Kim, MS[1], Hwa-Jung Nam, MS[1], Bon Jin Kim, PhD[2], Ill Young Lee, PhD[2], Chong-Kyo Lee, PhD[2], Jae Hak Kim, MS[2]
[1]ST Pharm.Co.,Ltd.; [2]Korea Research Institute of Chemical Technology

6. Phytochemical Analysis and Antibacterial Activities of Pistacia Atlantica from North-West Iran
Hossein Mostafavi, PhD
University of Tabriz, faculty of Tabriz, Tabriz, Iran, Tabriz, Islamic Republic of Iran

8. Antiviral Activity Of Ursolic Acid In Rotavirus Infections
Maria Julieta Tohme, MS[1], Maria Cecilia Gimenez, MS[1], Andrea Peralta, PhD[2], Maria Isabel Colombo, PhD[3], Laura Ruth Delgui, PhD[4]
[1]IHEM-UNCuyo-CONICET Juan Agustin Maza University, Mendoza, Argentina; [2]INTA-CONICET, Buenos Aires, Argentina; [3]IHEM-UNCuyo-CONICET, Mendoza, Argentina; [4]IHEM-UNCuyo-CONICET Facultad de Ciencias Exactas y Naturales. UNCuyo, Mendoza, Argentina

10. CRISPR/Cas9: An Antiviral Approach to Circumscribe Cotton Leaf Curl Disease
Muhammad Sattar, PhD
King Faisal University, Al-Hafuf, Al-hassa, Saudi Arabia

12. Synthesis of Biogenic Silver Nanoparticle from Methanolic Leaf Extract of *Wrightia tinctoria* and Exploration of its Anticancer and Antiviral Activity
Periyasamy Selvam, PhD[1], Ashish Wadhwani, PhD[2], Sameer Kumar Panda, MS[2]
[1]IRC, Kalasalingam Univerity, Krishnankoil 626126, Tamilnadu, India; [2]Dept Pharm biotechnology, JSS College of Pharmacy, JSS University, Ooty, Tamilnadu, India

14. An Efficient Synthesis of 2′-fluoro-6′-methylene-carbocyclic Adenosine (FMCA) and It's Prodrug FMCAP as an Anti-HBV Agent
Uma Singh, PhD, Ram Mishra, PhD, Varughese Mulamoottil, PhD, Chung Chu, PhD
University of Georgia, Athens, Georgia, United States of America

16. In Vivo Efficacy of Oral Treatment with Pritelivir Against Acyclovir Resistant Herpes Simplex Virus Type 1 or 2 in BALB/c Mice
Debra Quenelle, DVM, PhD[1], Alexander Birkmann, PhD[2], Thomas Goldner, PhD[2], Tamara Pfaff, DVM[2], Holger Zimmermann, PhD[2], Susanne Bonsmann, PhD[2], Deborah Collins, BS[1], Terri Rice, BS[1], Emma Harden, BS[1], Mark Prichard, PhD[1]
[1]The University of Alabama School of Medicine, Birmingham, Alabama, , United States of America; [2]AiCuris Anti-Infective Cures GmbH, Wuppertal, Germany

18. Design and Synthesis of Biogenic Silver Nanoparticles from Ethanoilc Leaf Extract of *Andrographis peniculata* and Exploration of its Anticancer Activity

Periyasamy Selvam, PhD[1], Ashish Wadhwani, PhD[2], Sameer Kumar Panda, MS[2]
[1]*International Research Centre, Kalasalingam University, Krishnankoil 626126, Tamilnadu, India;* [2]*Dept. of Pharmaceutical Biotechnology, JSS College of Pharmacy, JSS University, Ooty, Tamilnadu, India*

20. Evidence that ER α-glucosidase Inhibitors Potentiate Other Broad-spectrum Antivirals Against Multiple Families of Hemorrhagic Fever Viruses in vitro and in vivo

Jinhong Chang, MD, PhD[1], Julia Ma, BS[1], Xuexiang Zhang, MS[1], Travis Warren, PhD[2], Veronica Soloveva, PhD[2], Fang Guo, MD, PhD[1], Qing Su, PhD[1], Shuo Wu, PhD[1], Helen Shen, PhD[3], Eric Solon, PhD[3], Yanming Du, PhD[1], Sina Bavari, PhD[2], Ju-Tao Guo, MD[1], Timothy Block, PhD[1]
[1]*Baruch S. Blumberg Institute;* [2]*United States Army Medical Research Institute of Infectious Diseases;* [3]*QPS, LLC*

22. Combining Hepatitis B Surface Antigen with Tetanus for a Single Oral Vaccine

Mani Agarwal, BS[1], Vishal Bhargava, MD, PhD[2]
[1]*GTB Hospital, Sagar, India;* [2]*KRV Hospitals, Kanpur, U.P., India*

24. Identification Of HSP-90 Inhibitors As Potential Anti-HIV Molecules

Jay Trivedi, MS[1], Afsana Parveen, MS[2], Ashoke Sharon, PhD[2], Debashis Mitra, PhD[1]
[1]*National Centre for Cell Science, Pune, India;* [2]*Birla Institute of Technology, India*

26. Anti-hepatitis B Virus (HBV) Activity of Novel Pyrimidotriazinone Derivatives Through the Inhibition of Viral Nucleocapsid Assembly

Masaaki Toyama, PhD[1], Norikazu Sakakibara, PhD[2], Takayuki Hamasaki, PhD[1], Mika Okamoto, MD, PhD[1], Koichi Watashi, PhD[3], Takaji Wakita, MD, PhD[3], Masanori Baba, MD, PhD[1]
[1]*Kagoshima University, Japan;* [2]*Tokushima Bunri University, Japan;* [3]*National Institute of Infectious Diseases, Japan*

28. Development of Engineered Nanocarrier for Controlled Delivery of a Protease Inhibitor

Saurabh Bhargava, MD, MPH[1], Vishal Bhargava, MD, MPH[2], Aakanchha Jain, PhD[3]
[1]*Himalayan University, Kanpur, India;* [2]*GTB Hospitals, Kanpur, India;* [3]*DOPS, Sagar, India*

30. *In Silico* Discovery of a Protein-Protein Interaction Inhibitor for Influenza Viruses

Gregory Mohl, BS, David Busath, MD
Brigham Young University, Provo, Utah, United States of America

32. Establishment of an Antiviral Assay System and Identification of Severe Fever with Thrombocytopenia Syndrome Virus (SFTSV) Inhibitors

Masanori Baba, MD, PhD[1], Masaaki Toyama, PhD[1], Nobukazu Sakakibara, PhD[2], Mika Okamoto, MD, PhD[1], Masayuki Saijo, MD, PhD[3]
[1]*Kagoshima University, Kagoshima, Japan;* [2]*Tokushima Bunri University, Sanuki, Japan;* [3]*National Institute of Infectious Diseases, Tokyo, Japan*

34. Development and Characterization of an Aerodynamic System for Pulmonary Delivery of Influenza Vaccine

Mani Bhargava, MD, MPH[1], Saurabh Bhargava, MD, MPH[2], Piush Khare, PhD[3]
[1]*ICFAi Univ, Kanpur, India;* [2]*Himalayan University;* [3]*United Institute of Pharmacy*

36. Selective Inhibitor of Nuclear Export (SINE) compounds Reduce RSV Replication in vitro

Cynthia Mathew, PhD[1], Reena Ghildyal, PhD[1], Patricia Jorquera, PhD[2], Sharon Tamir, PhD[3], Jennifer Pickens, PhD[2]

[1]University of Canberra, Canberra, Australia; [2]Department of Infectious Disease, University of Georgia, Athens, Georgia, United States of America; [3]Karyopharm Therapeutics Inc, Newton, MA, USA, 2459, Newton, Massachusetts, United States of America

38. A Novel Kinase Inhibitor is a Pan-Influenza Antiviral with a High Barrier to Resistance

Ryan O'Hanlon, MS, Megan Shaw, PhD

Icahn School of Medicine at Mount Sinai, New York, New York, United States of America

40. 2'-Fluoro-2'-Deoxycytidine Inhibits Arenavirus and Bunyavirus Replication

Donald Smee, PhD, Kie-Hoon Jung, PhD, Jonna Westover, PhD, Brian Gowen, PhD

Institute for Antiviral Research, Utah State University, Logan, Utah, United States of America

42. Anti-Influenza Activity of the N-Benzylaminomethansulphonic Acid

Ruslan Khoma, PhD[1], Alim Ennan, MD, MPH[2], Tetyana Grydina, PhD[3], Karina Radkevich, MS[3], Alla Fedchuk, PhD[4], Viktor Lozitsky, PhD[4]

[1]Odessa I.I. Mechnikov National University, Odesa, Ukraine; [2]Physico-Chemical Institute of Environment and Human' Protection, Odesa, Ukraine; [3]Odesa National Medical University, Odesa, Ukraine; [4]Odessa Research Center for Biological Testing Products and Preparations, Odesa, Ukraine

44. Design, Synthesis and Characterization of Biogenic Chloroquine Silver Nanoparticles as Potential Anti-Hbv And Anticancer Agent

M Chandramohan, MD, PhD[1], P Selvam, PhD[2], D. Sivakumar, MD[1], S.C. Vivekananthan, MD[1], Elanchezhiyan Manickan, MD, PhD[3]

[1]Bharat Ratna Kamarajar Liver Hospital and Research Centre, Madurai., Madurai. Tamilnadu, India; [2]Sir CV Raman-Krishna International Research Centre, Kalasalingam University, Krishnankoil, Tamildau, India; [3]Dept of Microbiology, University of Madras-Taramani Campus, Chennai, Tamilnadu, India

46. Synthesis of Small Molecules for Treating Ebola Virus Infection

Elzbieta Niemiec-Plebanek, PhD[1], Vincent ROY, PhD[1], Maximes Bessieres, PhD[1], Dawid Warszycki, PhD[2], Andrzej Bojarski, PhD[2], Gilles Lalmanach, PhD[3], **Luigi Agrofoglio, PhD**[1]

[1]ICOA UMR CNRS 7311 – Universite Orleans, Orleans, France; [2]Institute Of Pharmacology – Polish Academy of Sciences, Krakow, Poland; [3]INSERM, UMR 1100 – Universite Tours, Tours, France

48. Antiviral Activity of New Fluorinated Thioacyl Derivatives of Amino Acids

Liubov Biliavska, PhD[1], Yulia Pankivska, MS[1], Olga Povnitsa, PhD[1], Svitlana Zagorodnya, PhD[1], Nadya Pikun, PhD[2], Yuriy Shermolovich, MD, MPH[2]

[1]D.K. Zabolotny Institute of Microbiology and Virology of the NASU, Kiev, Ukraine; [2]Institute of Organic Chemistry NAS of Ukraine, Kyiv, Ukraine

50. Study the Influence of Process and Formulation Parameters on Solubility and Dissolution Enhancement of Efavirenz Solid Solutions Prepared by Hot Melt Extrusion Using Box Behnken Factorial Design

Dilipkumar Suryawanshi, PhD

Institute of Chemical Technology, Mumbai, Mumbai, Maharashtra, India

52. 6'-Fluoro-3-deazaneplanocin: Synthesis and Antiviral Properties

Chong Liu, PhD[1], Qi Chen, PhD[2], Steven Cardinale, PhD[3], Terry Bowlin, PhD[3], Stewart Schneller, PhD[1]

[1]Auburn University, Auburn, Alabama, United States of America; [2]Slippery Rock University of Pennsylvania, Slippery Rock, Pennsylvania; [3]Microbiotix, Inc., Worcester, Massachusetts

54. Antiviral Activity of Oroxylin A against Coxsackievirus B3 Alleviates Virus-induced Acute Pancreatic Damage in Mice

Bo-Eun Kwon, MS[1], Hyuk-Hwan Song, PhD[2], Eun-Hye Hong, PhD[1], Hyun-Jeong Ko, PhD[1]

[1]College of Pharmacy, Kangwon National University; [2]Agency for Korea National Food Cluster

56. Formulation of Antiretroviral Drugs into Single and Dual Component Solid Drug Nanoparticles for Improved Oral Bioavailability

Alison Savage, PhD[1], Samantha Chadwick, PhD[1], Darren Moss, PhD[2], Helen Box, PhD[2], Joanne Sharp, PhD[2], Andrew Owen, PhD[2], Steve Rannard, PhD[1]

[1]Department of Chemistry, University of Liverpool, Liverpool, United Kingdom; [2]Department of Molecular and Clinical Pharmacology, University of Liverpool, Liverpool, United Kingdom

58. Development of Nanoparticulate System for Vaccine Delivery

Marut Agarwal, MD, MPH[1], Saurabh Bhargava, MD, MPH[2]

[1]Manav Bharti University, India; [2]Himalayan University, India

60. Development of a Model for Enterovirus D68 Infection in Mice

Bart Tarbet, PhD, Brett Hurst, MS, Joseph Evans, BS

Institute for Antiviral Research, Utah State University, Logan, Utah, United States of America

62. Ribavirin-Imprinted Polymers as Drug Delivery System

Mohamed Ayari, MS, Patrick Favetta, PhD, Vincent Hervin, MS, Vincent Roy, PhD, Luigi Agrofoglio, PhD

ICOA UMR CNRS 7311 – Universite Orleans, Orleans, France

64. Antiadenoviral Activity of New Nanoparticles

Yulia Pankivska, MS[1], Liubov Biliavska, PhD[1], Olga Povnitsa, PhD[1], Svitlana Zagorodnya, PhD[1], Anatoly Dorovskykh, PhD[2], Mykhailo Lokshyn, PhD[3], Valery Lozovski, PhD[4], Volodymyr Lysenko, PhD[3], Valentyn Tertykh, PhD[4]

[1]Zabolotny Institute of Microbiology and Virology of the NASU, Kyiv, Ukraine; [2]LLC Scientific-Production Enterprise «International Medical Center», Kyiv, Ukraine; [3]V. Laskariov Institute of Semiconductor Physics of the NASU, Ukraine; [4]Institute of High Techologies T. Shevchenko National University, Kyiv, Ukraine; [5]Chuiko Institute of Surface Chemistry of the NASU, Kyiv, Ukraine

66. Cardiovascular Mortality of HIV-Infected Patients

Valentina Golyshko, PhD, Victor Snezhitskiy, MD, PhD, **Natallia Matsiyeuskaya, MD, PhD**

Grodno State Medical University, Grodno, Belarus

68. Development of Quinazolinone-Based Inhibitors Against Venezuelan Equine Encephalitis Virus

Nikhil Madadi, PhD[1], Omar Moukha-Chafiq, PhD[1], Saibal Chakraborthy, PhD[1], Daniel Streblow, PhD[2], Nicole Haese, PhD[2], Thomas Morrison, PhD[3], Nicholas May, PhD[3], Mark Heise, PhD[4], Victor DeFilippis, PhD[2], Corinne Augelli-Szafran, PhD[1], Mark Suto, PhD[1], Ashish Pathak, PhD[1]

[1]Southern Research; [2]Oregon Health & Science University; [3]University of Colorado School of Medicine; [4]University of North Carolina at Chapel Hill

70. Benzoannulenes as Inhibitors Against Chikungunya Virus

Syed Ahmed, PhD[1], Vibha Pathak, MS[1], Jaden Cowan, BS[1], Daniel Streblow, PhD[2], Nicole Haese, PhD[2], Nicholas May, PhD[3], Thomas Morrison, PhD[3], Mark Heise, PhD[4], Victor DeFilippis, PhD[2], Corinne Augelli-Szafran, PhD[1], Mark Suto, PhD[1], Ashish Pathak, PhD[1]
[1]Southern Research, Birmingham, Alabama, United States of America; [2]Oregon Health & Science University, Beaverton, Oregon, United States of America; [3]University of Colorado School of Medicine, Aurora, Colorado, United States of America; [4]University of North Carolina at Chapel Hill, Chapel Hill, North Carolina, United States of America

72. Inhibition of Dengue Virus by Novel Inhibitors of RNA-Dependent RNA Polymerase and Protease Activity

Giuseppe La Regina, PhD[1], Valeria Famiglini, PhD[1], Valentina Naccarato, MS[1], Antonio Coluccia, PhD[1], John Hiscott, PhD[2], Jin-Ching Lee, PhD[3]
[1]Sapienza University, Institut Pasteur Italy – P.le A. Moro 5, I-00185 Roma, Italy, Roma, Italy; [2]Institut Pasteur Italy, Viale Regina Elena 291, 00161 Roma, Italy; [3]National Cheng Kung University, Taiwan, Taiwan

74. Antiviral Potentials of *Omidun* And Selected Lactic Acid Bacteria Against Selected Human Enteroviruses (HEV)

Abidemi Sunmola, BS[1], Funmilola Ayeni, PhD[1], Omonike Ogbole, PhD[2], Temitope Faleye, MS[3], Adekunle Adeniji, PhD[3]
[1]Department of Pharmaceutical Microbiology, Faculty of Pharmacy, University of Ibadan, Ibadan, Nigeria; [2]Department of Pharmacognosy, Faculty of Pharmacy, University of Ibadan, Ibadan, Nigeria; [3]W.H. O Polio Laboratory, Department of Virology, University of Ibadan, Ibadan, Nigeria

76. Pyrrole and Pyrazole-3-carbothioamide Derivatives Act as Dual Allosteric Inhibitors of HIV-1 Reverse Transcriptase

Angela Corona, PhD[1], Valentina Onnis, PhD[1], Alessandro Deplano, PhD[1], Monica Demurtas, MS[1], Simona Distinto, PhD[1], Giulia Bianco, PhD[1], Stefano Alcaro, PhD[2], Francesca Esposito, PhD[1], Enzo Tramontano, PhD[1]
[1]University of Cagliari, Cagliari, Italy; [2]University of Magna Graecia

78. Identification of Candidate Immunomodulatory Viral-host Cell Interaction Between Dengue NS5 and Cellular PML Protein

Federico Giovannoni, MS[1], Peter Hemmerich, PhD[2], **Cybele Garcia, PhD**[3]
[1]Lab de Estrategias Antivirales, QB, FCEyN, UBA- Instituto de QB FCEyN-CONICET, Buenos Aires, Argentina; [2]Leibniz Institute on Aging – Fritz-Lipman-Institut, Jena, Germany; [3]Lab de Estrategias Antivirales, QB, FCEyN, UBA- Inst. QB FCEyN (IQUIBICEN)-CONICET., Buenos Aires, Argentina

80. Drug Design and Synthesis of New Indolylarylsulfones as HIV-1 Non-nucleoside Reverse Transcriptase Inhibitors

Valeria Famiglini, PhD[1], Giuseppe La Regina, PhD[1], Antonio Coluccia, PhD[1], Domiziana Masci, PhD[1], Roger Badia, PhD[2], José A. Esté, PhD[2], Emmanuele Crespan, PhD[3], Giovanni Maga, PhD[3]
[1]Sapienza University, Institut Pasteur Italy – P.le A. Moro 5, I-00185 Roma, Italy, Roma, Italy; [2]AIDS Research Institute – IrsiCaixa, Universitat Autonóma de Barcelona, Badalona, Spain; [3]IGM-National Research Council, via Abbiategrasso 207, I-27100 Pavia, Italy, Pavia, Italy

82. Brazilian Natural Compounds and Derivative Synthetic Analogues Efficiently Inhibit Chikv and Zikv Infection

Jacqueline Shimizu, MS[1], Suely Silva, BS[1], Daniel Martins, BS[1], Debora Oliveira, BS[1], Zsofia Igloi, PhD[2], Cintia Bittar, PhD[3], Paula Rahal, PhD[3], Luis Regasini, PhD[3], Andres Merits, PhD[4], Mark Harris, PhD[2], **Ana Carolina Jardim, PhD**[5]
[1]Laboratory of Virology, Federal University of Uberlândia, Uberlândia, MG, Brazil; [2]Institute of Molecular and Cell Biology, University of Leeds, Leeds, UK; [3]Sao Paulo State University – UNESP, São Jose do Rio Preto, SP, Brazil; [4]University of Tartu; [5]Federal University of Uberlandia, Uberlandia, Minas Gerais, Brazil

84. **Cell Line Stably Expressing A ZsGreen Minigenome Enables Drug Screening Using All Known Ebolaviruses**

Markus Kainulainen, PhD, Mike Flint, PhD, Cesar Albarino, PhD, Christina Spiropoulou, PhD
Viral Special Pathogens Branch, Centers for Disease Control and Prevention, Atlanta, Georgia, United States of America
[1]Vaccine & Gene Therapy Institute, Oregon Health & Science Univeristy; [2]Southern Research; [3]University of North Carolina at Chapel Hill; [4]University of Colorado School of Medicine

86. **High Throughput Screen Measuring Marburg Virus VP24 Activation of the Nrf2 Antioxidant Response**

Megan Edwards, PhD, Christopher Basler, PhD
Institute for Biomedical Sciences, Georgia State University, Atlanta, Georgia, United States of America

88. **A Novel Class of Replication Inhibitors of RSV N/P Interaction**

Roberto Manganaro, MS[1], Dirk Jochmans, PhD[2], Johan Neyts, PhD[2], Pleter Leyssen, PhD[2], Andrea Brancale, PhD[1]
[1]Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff, United Kingdom; [2]Rega Institute – University of Leuven, Belgium

90. **Profiling of Neuraminidase Inhibitor Resistance among subtype N4, N5, N6 and N8 Avian Influenza Viruses**

Won-Suk Choi, MS[1], Jin Jung Kwon, BS[1], Ju Hwan Jeong, BS[1], Ji Won Han, BS[1], Su Jeong Ahn, BS[1], Su-Jin Park, BS[1], Young-il Kim, MS[1], Chul-Joong Kim, PhD[2], Young Ki Choi, PhD[1], Yun Hee Baek, PhD[1], Min-Suk Song, PhD[1]
[1]Chungbuk National University, Cheongju, Republic of Korea; [2]Chungnam National University, Dae Jeon, Republic of Korea

92. **Delineate the Dimeric Flip-Flop Mechanism of Coronaviral Main Protease During Catalysis**

Lin Shi, BS, Chi-Yuan Chou, PhD
Department of Life Sciences, National Yang-Ming University, Taipei, Taiwan

94. **A High Content Screen Identifies Cellular microRNAs with Anti-Flavivirus Activity**

Jessica Smith, PhD, Ashleigh Murphy, BS, **Alec Hirsch, PhD**
Vaccine and Gene Therapy Institute, Oregon Health & Science University, Beaverton, Oregon, United States of America

96. **Identification of a Substituted Thienopyrimidine Scaffold with Antiviral Activity Against Zika Virus**

Marcella Bassetto, PhD[1], Juliane Nolte, MS[2], Benno Schreiner, MS[2], Joachim Bugert, MD, PhD[2], Andrea Brancale, PhD[1]
[1]Cardiff University; [2]Institut für Mikrobiologie der Bundeswehr, München

98. **Evaluation of Lamivudine Sustained Release Tablet for Hepatitis B Infection**

Lila Nath, PhD[1]
[1]Department of Pharmaceutical Sciences, Dibrugarh University, Dibrugarh, Dibrugarh, India

100. Advances in Mouse Models of Crimean-Congo Hemorrhagic Fever: Viral Tropism and Neurological Disease in Hu-NSG™-SGM3 Humanized Mice

Jessica Spengler, DVM, PhD[1], M. Kelly Keating, DVM[1], Anita McElroy, MD, PhD[1], Marko Zivcec, PhD[1], JoAnn Coleman-McCray, BS[1], Jessica Harmon, MS[1], Brigid Bollweg, MS[1], Cynthia Goldsmith, MS[1], Eric Bergeron, PhD[1], James Keck, PhD[2], Sherif Zaki, MD [1], Stuart Nichol, PhD[1], Christina Spiropoulou, PhD[1]

[1]Centers for Disease Control and Prevention, Atlanta, Georgia, United States of America; [2]The Jackson Laboratory, Sacramento, California

102. Antiviral Treatment Efficiently Inhibits Chikungunya Virus Replication in the Joints of Mice During the Acute But Not During the Chronic Phase of Infection

Rana Abdelnabi, MS[1], Dirk Jochmans, PhD[1], Erik Verbeken, PhD[2], **Leen Delang, PhD**[1], Johan Neyts, PhD[1]

[1]Rega Institute for Medical Research, KU Leuven, Leuven, Belgium; [2]Department of Pathology, University of Leuven and Leuven University Hospitals, Leuven, Belgium

104. High-Throughput Screening to Identify Dengue Virus Entry Inhibitors

Wenlong Lian, PhD, Priscilla Yang, PhD

MBIB, Harvard Medical School

106. MERS-CoV Infection of Human Monocyte-derived Cells and Antiviral Efficacy of Select FDA-approved Drugs

Yu Cong, MD[1], Brit Hart, MS[2], Robin Gross, BS[1], Huanying Zhou, BS[3], Lisa Hensley, PhD[4], Jahrling Peter, PhD[5], Julie Dyall, PhD[6], Michael Holbrook, PhD[1]

[1]NIAID DCR IRF Battelle, Frederick, Maryland, United States of America; [2]APHL – Association of Public Health Laboratories, Gaithersburg, Maryland, United States of America; [3]NIAID DCR IRF LBERI, Frederick, Maryland, United States of America; [4]NIH NIAID DCR IRF, Frederick, Maryland, United States of America; [5]NIAID DCR IRF, Frederick, Maryland, United States of America; [6]NIAID DCR IRF Tunell, Frederick, Maryland, United States of America

108. Development of an in ovo System for Evaluation of Antivirals Against Zika Virus

Jasper Chan, MD, Kwok-Hung Chan, PhD, Shuofeng Yuan, PhD, Kenn Chik, BS, Zheng Zhu, MS, Kah-Meng Tee, BS, Jessica Tsang, BS, Cyril Yip, PhD, Vincent Poon, MS, Chris Chan, MS, Winger Mak, BS, Anna Zhang, PhD, Kwok-Yung Yuen, MD

Department of Microbiology, The University of Hong Kong, Hong Kong

110. Treatment of Old and New World Arenavirus Infections with Favipiravir

Brian Gowen, PhD[1], Jonna Westover, PhD[1], Eric Sefing, MS[1], Brady Hickerson, BS[1], Kevin Bailey, BS[1], Luci Wandersee, BS[1], Brittney Downs, BS[1], Skot Nielson, MS[1], Yousuke Furuta, PhD[2]

[1]Utah State University; [2]Toyama Chemical Co., Ltd.

112. Investigation of Stem Cell-Derived Alveolar like Macrophages as a Novel RSV Therapeutic

Yuchen Cen, BS[1], Michael Litvack, PhD[2], Wenming Duan, PhD[2], Martin Post, PhD[1], Theo Moraes, MD, PhD[1]

[1]University of Toronto, The Hospital for Sick Children, Toronto, Ontario, Canada; [2]The Hospital for Sick Children, Toronto, Ontario, Canada

114. Discovery of Toll-Like Receptor Potentiators

Joe Baldick, PhD, Betsy Eggers, MS, Robert Bertekap Jr., MS, Kevin Pokornowski, MS, Neil Burford, PhD, Andrew Alt, PhD, **Stephen Mason, PhD**

Bristol-Myers Squibb, Wallingford, Connecticut, United States of America

116. Inactivation of Respiratory Viruses Using Far-infrared Radiant Heater

Chong-Kyo Lee, PhD[1], Chonsaeng Kim, PhD[1], Keunbon Ku, DVM[1], Jin Soo Shin, DVM[1], Hae Soo Kim, BS[1], Gi Ppeum Lee, MS[1], Chun Sik Jeon, MS[2], Hee Jung Lee, BS[2], Jaekyung Hyun, PhD[3]
[1]Korea Research Institute of Chemical Technology, Daejeon, Korea, Republic of; [2]Ecopartners Ltd, Seoul, Korea, Republic of; [3]Korea Basic Science Institute, Cheongju, Korea, Republic of

118. Role of Receptor Tyrosine Kinases and Associated Gangliosides in Influenza Virus Replication

Pieter Vrijens, MS[1], Els Vanstreels, PhD[1], Roberto Ronca, PhD[2], Marco Presta, PhD[2], Sandra Liekens, PhD[1], Lieve Naesens, PhD[1]
[1]Rega Institute for Medical Research, KU Leuven – University of Leuven, Belgium, Leuven, Vlaams-Brabant, Belgium; [2]Experimental Oncology and Immunology, University of Brescia, Italy, Brescia, Italy

120. Synergistic Antiviral Effect of Polymerase and Autophagy Inhibitors on Dengue and Zika Virus Infected Cells *in vitro*

Cecilia Cima, PhD[1], Marcella Bassetto, PhD[1], Daniela Friese, BS[2], Juliane Nolte, BS[2], Gerhard Dobler, MD, PhD[2], Silke Wölfel, MD[2], Andrea Brancale, PhD[1], **Joachim Bugert, MD, PhD**[2]
[1]Cardiff School of Pharmacy & Pharmaceutical Sciences, Cardiff, Wales, United Kingdom; [2]Institut für Mikrobiologie der Bundeswehr, München, Bavaria, Germany

122. Mechanism Study of Baicalin and Baicalein Against Dengue Virus

Pouya Hassandarvish, PhD[1], Justin Jang Hann Chu, PhD[2], Sazaly AbuBakar, PhD[3], **Keivan Zandi, PhD**[4]
[1]Department of Medical Microbiology, Faculty of Medicine, UM, Malaysia; [2]Dept of Microbiology, Yong Loo Lin School of Medicine, National University of Singapore; [3]Tropical Infectious Diseases Research and Education Center, UM, Malaysia; [4]1- Emory University 2-TIDREC, University of Malaya, Atlanta, Georgia, United States of America

124. Pre- and Post-exposure Treatment of Quercetin-3-β-O-D-glucoside Against Ebola Virus Infection

Xiangguo Qiu, MD[1], Andrea Kroeker, PhD[1], Shihua He, PhD[1], Robert Kozak, PhD[1], Jonathan Audet, PhD[1], Majambu Mbikay, PhD[2], Michel Chretirn, MD[2]
[1]Special Pathogens Program, NML/ Public Health Agency of Canada, Winnipeg, Manitoba, Canada; [2]Clinical Research Institute of Montreal

126. Substituted Pyrimidines as Potent Non-nucleoside Reverse Transcriptase Inhibitors

Petr Simon, PhD[1], Lucie Cechova, MS[1], Ondrej Baszczynski, PhD[1], David Saman, PhD[1], George Stepan, PhD[2], Eric Hu, PhD[2], Eric Lansdon, PhD[2], Petr Jansa, PhD[2], Zlatko Janeba, PhD[1]
[1]Institute of Organic Chemistry and Biochemistry, Czech Academy of Sciences, Prague, Czech Republic; [2]Gilead Sciences Inc., 333 Lakeside, Foster City, CA 94404, USA

128. Verdinexor, a Selective Inhibitor of Nuclear Export (SINE) Compound, Exhibits Significant Antiviral Activity Against HIV and SIV

Sharon Tamir, PhD[1], Shelton Cochran, BS[1], Marie Mankowski, BS[2], Priscilla Hogan, BS[2], Trinayan Kashyap, MS[1], Yossi Landesman, PhD[61], Margaret Lee, PhD[1], Sharon Shacham, PhD[1], Roger Ptak, PhD[2]
[1]Karyopharm Therapeutics, Newton, Massachusetts, United States of America; [2]Southern Research Institute, Frederick, Maryland, United States of America

130. **Inhibition of Respiratory Virus Infection by Cholesterol Reducing Agents**
Shringkhala Bajimaya, MS
University of Rochester School of Medicine and Dentistry, Rochester, New York, United States of America

186. **Predicting ADME and PK Properties of Antivirals for Ebola**
Mary A. Lingerfelt[1], Kimberley M Zorn[1], Joel S. Freundlich[2], Manu Anantpadma[3], Gauri Rao[4], John Diep[4], Robert A. Davey[3], Peter B. Madrid[5] and Sean Ekins[1]
[1]Collaborations Pharmaceuticals Inc., Fuquay-Varina, NC 27526; [2]Departments of Pharmacology, Physiology & Neuroscience and Medicine, Center for Emerging and Reemerging Pathogens, Rutgers University, Newark NJ, 07103; [3]Texas Biomedical Research Institute, San Antonio, TX 78227, USA; [4]Eshelman School of Pharmacy, University of North Carolina, Chapel Hill, NC 27599; [5]SRI International, Menlo Park, CA, 94025, USA

Thursday May 25th, 2017

Medicinal Chemistry

Chairs: **Chris Meier, PhD** and **Zlatko Janeba, PhD**

WEST BALLROOM

9:00 AM – 10:30

9:00 AM **143.** **Cyclin G Associated Kinase (GAK) Inhibition as a Strategy for the Discovery of Broad Spectrum Antivirals**
Steven De Jonghe, PhD[1], Stefan Knapp, PhD[2], Piet Herdewijn, PhD[1], Shirit Einav, MD, PhD[3]
[1]Medicinal Chemistry, Rega Institute for Medical Research, KU Leuven, Leuven, Belgium; [2]Goethe-University Frankfurt, Institute of Pharmaceutical Chemistry, Frankfurt-am-Main, Germany; [3]Department of Microbiology and Immunology, Stanford University School of Medicine, Stanford, California, United States of America

9:20 AM **146.** **Lability of the Favipiravir Ribonucleoside and First Mechanistic Details**
Johanna Huchting, PhD, Matthias Winkler, MS, Hiba Nasser, MS, Chris Meier, PhD
Hamburg University, Hamburg, Germany

9:40 AM **133.** **Modified Tetrahydropteridines as Potent Anti-HIV NNRTIs with Improved Resistance Profile and Solubility**
Ondřej Baszczyňski, PhD[1], Petr Šimon, PhD[1], David Šaman, PhD[1], Eric Hu, PhD[2], Eric Lansdon, PhD[2], Petr Jansa, PhD[2], Zlatko Janeba, PhD[1]
[1]IOCB Prague, Prague, Czech Republic; [2]Gilead Sciences Inc., Foster City, California, United States of America

9:52 AM **145.** **Discovery of 2-Pyridinone Aminals: A Prodrug Strategy to Advance a Second Generation of HIV-1 Integrase Strand Transfer Inhibitors**
Izzat Raheem, PhD
Merck, West Point, Pennsylvania

10:04 AM **153.** **Design, Synthesis and Anti-RNA Virus Activity of Novel Fluorocarbocyclic Nucleosides**
Lak Shin Jeong, PhD[1], Jiseong Yoon, MS[1], Young Sup Shin, BS[1], Dnyandev Jarhad, PhD[1], Kristina Kovacikova, PhD[2], Clara Posthuma, MS[2], Eric Snijder, MD, PhD[2], Martijn van Hemert, MD, PhD[2]
[1]Seoul National University, Seoul, Korea, Republic of; [2]Leiden University

10:16 AM **149.** **Inhibitors of Emerging Flaviviruses**
Radim Nencka, PhD[1], Hubert Hřebabecký, PhD[1], Michal Šála, PhD[1], Milan Dejmek, PhD[1], Evzen Boura, PhD[1], Kamil Hercik, PhD[1], Daniel Růžek, PhD[2], Ludeˇk Eyer, PhD[2]
[1]IOCB Prague, Prague, Czech Republic; [2]Veterinary Research Institute, Brno, Czech Republic

10:30 – 11:00 AM **Coffee Break**

DNA Viruses and Respiratory Viruses

Chairs: **Phiroze Sethna, PhD** and **Mike Bray, MD**
WEST BALLROOM
11:00 AM – 12:30 PM

11:00 AM **147.** **Tropolones Powerfully Suppress Herpesvirus Replication: Preliminary Structure-Activity Relationship and Inhibition of Acyclovir-resistant Viruses**
Bindi Patel, BS[1], Aswin Garimallaprabhakaran, PhD[2], Alex Berkowitz, BS[2], Nana Agyemang, MS[2], Andreu Gazquez, BS[1], Peter Ireland, MD[1], Mark Cadiz, BS[1], John Tavis, PhD[1], Ryan Murelli, PhD[2], **Lynda Morrison, PhD**[1]
[1]Saint Louis University School of Medicine, St. Louis, Missouri; [2]The Graduate Center, City University of New York, New York, New York

11:15 AM **148.** **Post-exposure Administration of USC-087 Protects Immunosuppressed Syrian Hamsters Against Lethal Challenge with Human Species C Adenoviruses**
Karoly Toth, DVM[1], Jacqueline Spencer, BS[1], Baoling Ying, MD[1], Ann Tollefson, PhD[1], Caroll Hartline, PhD[2], Jiajun Fan, BS[3], Jinglei Lyu, BS[3], Boris Kashemirov, PhD[3], Mark Prichard, PhD[2], William Wold, PhD[1], Charles McKenna, PhD[3]
[1]Saint Louis University School of Medicine, St. Louis, Missouri, United States of America; [2]University of Alabama at Birmingham, Birmingham, Alabama, United States of America; [3]University of Southern California, Los Angeles, California, United States of America

11:30 AM **162.** **Experience of a Translational Research Platform for the Evaluation of Human Cytomegalovirus (HCMV) Drug-resistance in Belgium**
Graciela Andrei, PhD, Sarah Gillemot, MS, Robert Snoeck, MD, PhD
Rega Institute – KU Leuven, Leuven, Belgium

12:00 PM **151.** **KPT-335, a Selective Inhibitor of Nuclear Export (SINE) Compound, Modulates Respiratory Syncytial Virus (RSV) Matrix Protein Nuclear Trafficking and Immune Responses**
Jennifer Pickens, PhD[1], Sharon Tamar, PhD[2], Margaret Lee, PhD[2], Ralph Tripp, PhD[1]
[1]University of Georgia, Athens, Georgia, United States of America; [2]Karyopharm Therapeutics, Newton, Massachusetts, United States of America

12:13 PM **161.** **Antifungal Azoles that Target an Early Stage of the Parechovirus A3 Life Cycle**
Eric Rhoden, BS[1], Allan Nix, BS[1], William Weldon, PhD[1], Laurence Briesach, MS[2], Rangaraj Selvarangan, PhD[3]
[1]Centers for Disease Control and Prevention; [2]IHRC, contracting agency to the Centers for Disease Control and Prevention; [3]Children's Mercy Hospital

Closing Remarks

1. Antiviral Investigation and Phytochemical Profiling of *Bryophyllum pinnatum* and *Viscum album*

Robert Obi[1,3], Anjorin A.A.[2,3], Oke B.O.[3], Salu O.B.[3], James A.O.[4], Omilabu S.A.[3]

[1].Department of Microbiology, Federal University of Technology, Owerri, Imo State, Nigeria; [2].Department of Microbiology, Lagos State University, Ojo, Lagos State, Nigeria; [3]. Department of Medical Microbiology and Parasitology, College of Medicine, University of Lagos, Nigeria; [4].Department of Biochemistry, College of Medicine, University of Lagos, Nigeria

BACKGROUND: Measles, polio, yellow fever, and herpes are diseases of public health importance worldwide. Despite tremendous progress in human medicine, no drugs exist for the complete treatment of these viral diseases.

AIM: This study was designed to investigate the antiviral potentials of two medicinal plants available locally in South Eastern, Nigeria. Materials and Methods: Fresh leaves of *Bryophyllum pinnatum* and *Viscum album* were collected from Owerri, South Eastern Nigeria. Extraction of the plant materials was done with methanol using the Soxhlet extractor and concentrated using the rotary evaporator.

RESULTS: The toxicity profile shows that the minimum non-toxic concentration (MNTC) of *B. pinnatum* was 0.016 μg μL^{-1} with an IC_{50} of 0.313 μg μL^{-1} while that of *V. album* was 0.063μg μL^{-1} and IC_{50} of 0.063μg μL^{-1}. Result of the antiviral analysis shows that *B. pinnatum* and *V. album* produced activity against measles virus and herpes simplex virus – 1 at the concentrations of 0.016 μg μL^{-1} (IC_{50} 0.004 μg μL^{-1}; IC_{50} 0.004 μg μL^{-1}) and 0.063μg μL^{-1} (IC_{50} 0.031 μg μL^{-1}; IC_{50} 0.039 μg μL^{-1}) respectively, while polio and yellow fever viruses were resistant to both plants extracts at all the concentration tested. Result of the pre- and post- infection antiviral assays shows that the methanol extract of *B. pinnatum* possessed entry/attachment inhibitory against measles (IC_{50} 0.010 μg μL^{-1}) and HSV-1 (IC_{50} 0.004 μg μL^{-1}), while the activity of *V. album* against both viruses (IC_{50} 0.004 μg μL^{-1} for measles and IC_{50} 0.039 μg μL^{-1}, for HSV-1) was in the post infection antiviral assay. Selectivity index calculated for *B. pinnatum* and *V. album* on both viruses show that methanol extract plants was safe for use in antiviral chemotherapy. Result of the phytochemical of both plants revealed the presence of various secondary useful metabolites. Conclusion: This study has shown that the solution to most viral diseases of public health significance could be found in the forest zones of Nigeria.

2. Griffithsin, a Novel Inhibitor of Henipavirus Entry and Fusion.

Michael Lo, PhD[1], Barry O'Keefe, PhD[2], Anasuya Chattopadhyay, PhD[3], Anne Hotard, PhD[1], Lauren Haugh Krumpe, MS[2], John Rose, PhD[3], Stuart Nichol, PhD[1], Christina Spiropoulou, PhD[1]

[1]Centers for Disease Control and Prevention, Atlanta, Georgia, United States of America; [2]National Cancer Institute, Frederick, Maryland, United States of America; [3]Yale University School of Medicine, New Haven, Connecticut, United States of America

Nipah (NiV) and Hendra (HeV) viruses are highly pathogenic members of the genus Henipavirus, and cause fatal encephalitis and respiratory disease in humans. There is currently no approved therapeutic for human use against henipaviruses. Griffithsin (GRFT) is a high-mannose binding lectin that has shown variable broad-spectrum activity against viruses across many virus families, including SARS Coronavirus, HIV-1, and Japanese Encephalitis virus. Here we report the *in vitro* activity of GRFT as well as its related synthetic tandemers against Nipah and Hendra viruses.The 50% effective concentration (EC_{50}) of GRFT against NiV and HeV ranged from 0.5-3 ug/mL. At high concentrations, GRFT was able to reduce virus titers by more than two orders of magnitude. We showed that inhibition of henipaviruses required the natural dimeric form of GRFT, as monomeric GRFT (mG) did not inhibit virus replication. In addition, we observed that trimeric (3mG) and tetrameric (4mG) tandemers of mG had increased potency against henipaviruses compared to

wild-type GRFT. We performed virus entry and fusion assays to determine the mechanism of action of GRFT and 3MG, and observed that while both of them blocked entry at equivalent potencies, 3MG had a greater ability to block NiV glycoprotein-induced cell-to-cell fusion. The antiviral potency of GRFT as shown in this study, along with its previously demonstrated lack of mitogenic activity against human lymphocytes merit further studies to evaluate the potential of GRFT and its tandemers as therapeutics against henipaviruses relevant animal models.

3. Characterization of the Mode of Antiviral Action of U0126, a MEK Inhibitor, Against Junín Virus

Jesús Brunetti, BS, Verónica Quintana, BS, Luis Scolaro, PhD, Viviana Castilla, PhD
Biochemistry Department, School of Science, University of Buenos Aires, Buenos Aires, Argentina

We have previously demonstrated that U0126, a MEK inhibitor, impairs Junín virus (JUNV) multiplication in monkey and human cell cultures. Here we analyzed the mode of antiviral action of U0126 against JUNV multiplication in Vero cells. To determine whether U0126 affects JUNV adsorption, viral particles adsorbed at 4°C in the presence or absence of the inhibitor were quantified by a plaque formation assay (PFU). Another set of cultures infected at 4°C were transferred at 37°C in medium with or without U0126 and at different times post-infection (p.i.) the amount of internalized virus was quantified by PFU, using chlorpromazine, an inhibitor of JUNV internalization, and ammonium chloride, which blocks viral uncoating, as reference drugs. Neither the adsorption nor the kinetics of internalization or uncoating were affected by U0126.

Treatment with U0126 at different times p.i. caused a significant reduction in the synthesis of viral proteins N and GPC (precursor of viral glycoproteins G1 and G2) analyzed at 14h p.i. by western blot assays. Using an acid pH-induced cell fusion assay we also demonstrated that U0126 treatment also affected the expression of glycoprotein G2 at the cell membrane. Immunofluorescence assays revealed that cytoplasmic and membrane expression of G1 was impaired by the ERK inhibitor indicating that U0126 affected GPC synthesis rather than subsequent trafficking of viral glycoproteins to the plasma membrane. Finally, quantification by real-time RT-PCR revealed that treatment with U0126 reduced the synthesis of genomic RNA.

In conclusion, our results indicate that U0126 mainly inhibits JUNV protein and RNA synthesis.

4. Discovery and Development of Novel Non-Catalytic Site HIV Integrase Inhibitors

Kyungjin Kim, PhD[1], Uk-Il Kim, MS[1], Hwa-Jung Nam, MS[1], Bon Jin Kim, PhD[2], Ill Young Lee, PhD[2], Chong-Kyo Lee, PhD[2], Jae Hak Kim, MS[2]
[1]ST Pharm.Co.,Ltd.; [2]Korea Research Institute of Chemical Technology

Integrase (IN) required for the integration of viral DNA into the host genome plays an essential role in human immunodeficiency virus (HIV) replication. The enzyme represents an important target for treatment of HIV infection because there is no human homolog of IN. While several therapeutics targeting at the catalytic site of integrase (CIN) on the market, due to frequent emergence of viral resistance the current treatments for HIV infection are far from being ideal. Consequently, the development of other classes of IN inhibitors through different mechanism of action such as non-catalytic site integrase inhibitors (NCINIs) is highly being requested.

We herein present the development of a novel non-catalytic site integrase inhibitor, STP03-0404, with excellent in vitro antiviral activity against both wild-type and multidrug–resistant HIV strains, which showed good activity against all of Raltegravir-resistant strains in both MT4 cells and PBMCs. STP03-0404 also showed excellent ADME properties and PK profiles. The lead profiling screen results in 68 GPCR and receptors did not provide any inhibition activities. Furthermore, no toxicity issues were found in cytotoxicity studies, hERG assay, Ames test, and the acute toxicity studies conducted in rodent model.
In conclusion, STP03-0404 could be an excellent candidate for the treatment of HIV infection and will gain comparative advantage against viral resistance. The preclinical study of STP03-0404 is on-going and the phase I clinical trial is expected to begin in 2018.

5. Development of Bipolymer Based Novel Nanoparticles in Microsphere System as Vaccine Adjuvant

Saurabh Bhargava, MD, MPH[1], Vishal Bhargava, MD, MPH[2]
[1]*Himalayan University, Kanpur, U.P., India;* [2]*GTB Hospital, Kanpur, U.P., India*

Novel strategies are required for the achievement of safe and effective immunization beyond conventional strategies. Frequent booster dosing can be avoided by development of mucosal/adjuvant vaccine delivery system, which can produce both humoral and cell-mediated responses. The work envisaged uses combined hydrophilic(gelatin nanoparticles,GN) with a hydrophobic polymeric system(PLGA microspheres) which creates a biodegradable system for HBsAg delivery.

GN & PLGA microspheres were prepared by double emulsification method and composite system by phase separation method. Composites were optimized and characterized *in-vitro* for their shape, size by Scanning & Transmission Electron Microscopy, %antigen entrapment and stability. Fluorescence microscopy was carried out to confirm the uptake. *In-vivo* study comprised of estimation of IgG response in serum and sIgA in various body secretions using specific ELISA. The *in-vitro* studies exhibited an initial burst release from gelatin nanoparticles, degradation of antigen from PLGA microspheres & a continuous release from composite system. This supports the hypothesis to formulate single shot vaccine with such system (to mimic booster dosing). The fluorescence studies showed the selective uptake of composites by NALT.

Humoral response generated by single dose of composites was comparative to marketed formulation receiving booster dose. Further, composite system generated effective sIgA antibody which was not elicited by marketed formulation. Thus, it could be concluded from present study that bipolymer based composite system are capable to provide sufficient protein stability and can be a promising candidate for development of single shot vaccine, not only against Hepatitis but against all those diseases that invade host by mucosal surfaces.

6. Phytochemical Analysis and Antibacterial Activities of Pistacia Atlantica from North-West Iran

Hossein Mosatafavi[1],Gholamreza Zarini[2], Sakha Pezhhanfar[1]
[1]*Department of Organic Chemistry & Biochemistry, Faculty of Chemistry, University of Tabriz, Tabriz, Iran,*
[2]*Faculty of Natural Science, University of Tabriz, Tabriz, Iran*

The present study is carried out to analyse the phytochemical & antibacterial potential of Pistacia atlantica extracts. Aqueous ,Ethanol, acetone and chloroform extracts of pistacia atlantica subs kurdica were screened for Alkaloids, Carbohyrates, Flavonoids, tannins, Glycosides, Resins, Steroids and triterpenoids, Tanins, Starch, Inorganic acids, Organic acids, Ascorbic acid, Phenolic compounds, Amino acids, Proteins, coumarins, , antraquinones, Saponins, oil &fat and chalcones.

Detection of biological activitiy of plant extracts against, *Bacillus cereus* ATCC 11778, *Escherichia coli* ATCC 25922, *KLebsiella Pneumonia* ATCC 700603 and *Staphylococcus* ATCC 25952 was done by use of the paper disc diffusion method on Mueller hinton agar (Merck). Chloramphenicol and tetracycline and penicillin were standard reference antibiotics. The zone of inhibition was measured after 24 h at 37 °C. The four extracts of plant show high to moderate antibacterial activity.

KEYWORDS: Pistacia Atlantica, Antibacterial activity, Phytochemical

REFERENCE:

1) Zaika LL (1975). Spices and herbs: their antimicrobial activity and its determination. J food safety , 9: 97 – 118.

2) Sule IO and Agbabiaka TO (2008) .Antibacterial effect of some plant extracts on selected enterobacteriaceae .ethnobotanical leaflets; 12: 1035-1042

7. Synthesis and Antiviral Activity of North and South L-Neplanocin Derivatives

Qi Chen, PhD[1], Amber Davidson, BS[1], Megan Stout, BS[1], Stewart Schneller, PhD[2]
[1]*Slippery Rock Univeristy, Slippery Rock, Pennsylvania, United States of America;* [2]*Auburn University, Auburn, Alabama*

Recent studies have shown that L- like carbocyclic nucleosides, such as L-1', 6'-isoneplanocin analogues, possess broad spectrum antiviral activities, including Ebola, norovirus, vaccinia, HBV, HCMV, measles and Dengue. It is noteworthy that replacing the nitrogen atom to a CH or a CBr group at the N-3 position has significant impacts on their biological properties.

Previous studies have also found that C-4' truncated neplanocin analogue (DHCDA) is effective against a series of viruses, which is likely due to its inhibition of SAH hydrolase. Its selectivity was even greater than that of neplanocin, particularly against vesicular stomatitis virus (VSV) and rotavirus.

Following the lead of these compounds and as part of the study to L-like carbocyclic nucleosides, the L form of DHCDA (1), its 3-Br derivative (3), and the conformation restricted methanocarba (MC) nucleoside analogues (2 and 4) were set as target structures. The x-ray crystal structure shows L form MC analogues (2 and 4) adopt similar (North-like) locked conformation as conventional D-MC nucleosides, while DHCDA analogues (1 and 3) preferred South-like conformer. Compounds 1 and 4 show potent antiviral activity against norovirus, while compound 2 and 3 were inactive. (data shows as below) The antiviral activity combining with L-configuration, and possible syn and anti-conformation, provide interesting structural information for the mechanism as antiviral agents. More antiviral activities are under investigation.

8. Antiviral Activity Of Ursolic Acid In Rotavirus Infections

Maria Julieta Tohme, MS[1], Maria Cecilia Gimenez, MS[1], Andrea Peralta, PhD[2], Maria Isabel Colombo, PhD[3], Laura Ruth Delgui, PhD[4]
[1]*IHEM-UNCuyo-CONICET Juan Agustin Maza University, Mendoza, Argentina;*
[2]*INTA-CONICET, Buenos Aires, Argentina;* [3]*IHEM-UNCuyo-CONICET, Mendoza, Argentina;*
[4]*IHEM-UNCuyo-CONICET Facultad de Ciencias Exactas y Naturales. UNCuyo, Mendoza, Argentina*

Rotavirus (RV) is the main cause of viral diarrhea in children. It is known that ursolic acid (UA), a triterpene present in herbs and fruits, has biological properties such as antiviral and antitumoral. Our group evaluated the potential antiviral activity of UA in RV infections.

For this purpose we employed *in vitro* infections, using the monkey RV strain SA-11 and the MA104 cell line. Firstly, we determined the non-citotoxic concentrations of UA measuring cell viability after treatment with increasing concentrations of the compound at different times. Secondly, to discard virucidal effect, we determined the title of RV preparation after been incubated with UA. Finally, we evaluated UA antiviral activity by Western blot analysis of the major structural viral proteins, VP6 and VP7; and by titration of intra- and extra-cellular viral particles after UA treatment.

We determined the absence of a cytotoxic effect up to 15 μM UA during 24 h. We observed a dosis-dependent decrease in the production of a new viral progeny, demonstrated both by a decrease of the intra- and extra-cellular title, as by a reduction in the level of the viral proteins VP6 and VP7. We also established that the antiviral effect was not due to a virucidal effect since the viral title was not altered by the pre-treatment with UA.

In conclusion, our results suggest that UA interferes with one or more replication steps of RV and we are currently focused in determining the molecular mechanism involved in the antiviral activity of UA in RV infections.

9. Selective Inhibition of Hepatitis C Virus Replication by Alpha-zam, a *Nigella Sativa* Seed Formulation

Olufunmilayo Oyero, PhD[1], Masaaki Toyama, PhD[2], Naoki Mitsuhiro, DVM[2], Abdulfattah Onifade, PhD[1], Akemi Hidaka, BS[2], Mika Okamoto, MD, PhD[2], Masanori Baba, MD, PhD[2]
[1]University of Ibadan, Ibadan, Oyo, Nigeria; [2]Kagoshima University, Kagoshima, Kagoshima, Japan

Hepatitis C virus (HCV) infection became curable because of the development of direct acting antivirals (DAAs). However, the high cost of DAAs has greatly impeded their potential impact on the treatment of HCV infection. As a result, hepatitis C will continue to cause substantial morbidity, and mortality among chronically infected individuals in low and middle income countries. Thus, urgent need exists for developing cheaper drugs available to hepatitis C patients in these countries. Alpha-zam, an herbal formulation from *Nigella sativa* seed, was examined for its anti-HCV activity and cytotoxicity in genotype 1b HCV replicon cells. The antiviral activity was determined by luciferase expression and viral RNA synthesis, while the cytotoxicity was assessed by viable cell number and glyceraldehyde-3-phosphate dehydrogenase RNA synthesis in the replicon cells. Alpha-zam was found to be a selective inhibitor of HCV replication. The 50% effective dilution and 50% cytotoxic dilution of Alpha-zam were 761- and < 100-fold, respectively, in the subgenomic replicon cells LucNeo#2. Its selective inhibition was also confirmed by HCV RNA levels in LucNeo#2 and in the full-genome HCV replicon cells NNC#2 using real-time reverse transcriptase polymerase chain reaction and this was not due to the induction of interferon. Additionally the formulation increased peripheral blood mononuclear cells by 2.2 -2.3 fold following stimulation with 4000 and 400 dilutions of the drug and the effect was sustained after removal of the agent. Alpha-zam selectively inhibited HCV replication and therefore has potential for a novel hepatitis C agent through direct viral inhibition and immune modulation activity.

10. CRISPR/Cas9: An Antiviral Approach to Circumscribe Cotton Leaf Curl Disease

Muhammad Sattar, PhD
King Faisal University, Al-Hafuf, Al-hassa, Saudi Arabia

The begomoviruses (family *Geminiviridae*) associated with cotton leaf curl disease (CLCuD) pose a major threat to cotton productivity in South–East Asia. These viruses have single-stranded, circular DNA genome, of 2800 nt, encapsidated in twinned icosa-hedera, transmitted by ubiquitous whitefly and are associated with satellite molecules alpha- and betasatellite. To circumvent the proliferation of these viruses numerous techniques, ranging from conventional breeding to molecular approaches have been applied. Such devised strategies worked perfectly well for a short time period and then viruses relapse due to various reasons including synergism, recombination, virus proliferation and evolution. Another shortcoming is, until now, that all molecular biology approaches are devised to control only helper begomoviruses but not to control associated satellites. Despite the fact that satellites could add various functions to helper begomoviruses, they remain ignored. Such conditions necessitate a comprehensive approach not only against helper begomoviruses but also their associated DNA-satellites. In the current scenario clustered regulatory interspaced short palindromic repeats (CRISPR)/CRISPR associated nuclease 9 (Cas9) has proved to be a versatile and comprehensive technique that has very recently been deployed successfully to control different geminiviruses. However, like previously used techniques, only a single virus is targeted and hitherto it has not been deployed to control begomovirus complexes associated with DNA-satellites. Here in this article, we proposed an inimitable, unique, and broad spectrum controlling method based on multiplexed CRISPR/Cas9 system where a cassette of sgRNA is designed to target not only the whole CLCuD-associated begomovirus complex but also the associated satellite molecules.

11. CRISPR/Cas9-Mediated Antiviral Immunity Against Geminiviruses

Syed Shan e Ali Zaidi, PhD
Boyce Thompson Institute, Cornell University, Ithaca, NY, Ithaca, New York, United States of America

Geminiviruses cause devastating crop losses that threaten food security. They evolve rapidly by recombination, component capture, and mutation, allowing these viruses to rapidly counter or evade introduced resistance strategies. Clustered regularly interspaced short palindromic repeats (CRISPRs)/ CRISPR associated 9 (Cas9) is a prokaryotic molecular immunity system against invading viruses and has been harnessed as a powerful tool for targeted genomic editing. Recently we have demonstrated that the CRISPR/Cas9 system could be harnessed to confer resistance against geminiviruses in plants by using sgRNAs designed to target viral genomic DNAs. We showed that CRISPR/Cas9 technology could impart molecular immunity against three geminiviruses [i.e., *Tomato yellow leaf curl virus* (TYLCV), *Beet curly top virus* (BCTV), and *Merremia mosaic virus* (MeMV)] in *Nicotiana benthamiana* plants, and revealed that a sgRNA designed to target a conserved sequence (TAATATTAC) in the viral intergenic region could be used to target multiple geminiviruses simultaneously. This sequence is conserved among geminiviruses and is also a hallmark of the betasatellites of begomoviruses. We have also evaluated the differential targeting and possible evasion of geminiviruses in plants having CRISPR/Cas machinery. I would like to present this recently developed technology accompanied with our latest data.

12. Synthesis of Biogenic Silver Nanoparticle from Methanolic Leaf Extract of *Wrightia tinctoria* and Exploration of its Anticancer and Antiviral Activity

Periyasamy Selvam, PhD[1], Ashish Wadhwani, PhD[2], Sameer Kumar Panda, MS[2]
[1]IRC, Kalasalingam Univerity, Krishnankoil 626126, Tamilnadu, India; [2]Dept Pharm biotechnology, JSS College of Pharmacy, JSS University, Ooty, Tamilnadu, India

OBJECTIVE: *Wrightia tinctoria* (Roxb) R.Br (Apocynaceae) is versatile medicinal plant enriched with novel bioactive molecules. *Wrightia tinctoria* Leaf extracts and their isolated compounds demonstrated for inhibition of HIV-1 replication and HIV Integrase enzymatic activity (Selvam *et al.*, ICAR 2010; 2016). Thus, the study planned to biosynthesize anticancer and antiviral potent silver nanoparticles using methanolic leaf extracts of *Wrightia tinctoria* (WT AgNPS). Method: Methanolic leaf extract of *Wrightia tinctoria* used for the synthesis of silver nanoparticles. The synthesized nanoparticles were confirmed by color transformation and Ultra violet-Visible spectrophotometry. The size and morphology of the silver nanoparticle were characterized by SEM and EDEX. The stability of silver nanoparticle were detected by Fourier Transform Infra Red *Spectroscopy* (FT-IR) and PXRD. Anticancer activity of Silver nanoparticles tested against Neuroblastoma cell (SH-SY5Y cells), and cytotoxicity was also investigated against Vero cells by MTT assay. Antiviral activity of WT AgNPs under investigation. Result: The appearance of reddish brown colour and UV absorbtion range 422nm (OD1.123) confirmed the synthesized silver nanoparticles. The silver nanoparticle showed spherical structure and their size were ranging from 18-70 nm under SEM observations. FT-IR spectra of silver nanoparticles showed the peaks N-H, C=O, -C=C, and C-H which indicate the stability of synthesized silver nanoparticles. The obtained nanoparticle showed significant anticancer activity against Neuroblatoma cells (IC_{50}:22 µg/ml, whereus cytotoxicity (CC_{50}) in Vero cells at 170 µg/ml (SI>7). Conclusion: The synthesized WT AgNPS would be helpful for the preparation of potent cytotoxic agents against Neuroblatoma cells to destroy cancer cells.

13. Synthesis of Flexible Purine Analogue Inhibitors of NCp7

Therese Ku, BS, Katherine Seley-Radtke, PhD
University of Maryland, Baltimore County

Anti-HIV-1 drug design has been notably challenging due to the virus' ability to mutate and develop immunity against commercially available drugs. This project aims to discover a new series of nucleobase analogues that not only possess inherent flexibility that could withstand active site mutations, but also target a non-canonical, more conserved target, NCp7. Interestingly, these compounds are not predicted to work

by zinc ejection, which would endow them with significant advantages over currently reported zinc-ejectors, which are toxic. We propose to synthesize several series of these fleximer base analogues using palladium-catalyzed coupling techniques and to test them against NCp7 specifically, and HIV-1 in general. The results to date are reported herein.

14. An Efficient Synthesis of 2′-Fluoro-6′-Methylene-Carbocyclic Adenosine (FMCA) and it's Prodrug FMCAP as an Anti-HBV Agent.

Uma Singh, PhD, Ram Mishra, PhD, Varughese Mulamoottil, PhD, Chung Chu, PhD
University of Georgia, Athens, Georgia, United States of America

2′-Fluoro-6′-methylene-carbocyclic adenosine (FMCA) and it's phosphoramidate prodrug (FMCAP) have shown potent anti-HBV activity *in vitro* against both adefovir- & lamivudine-resistant double mutants (rtL180M/rtM204V) as well as lamivudine/entecavir triple mutants (L180M+S202G+M204V).[1,2] Preliminary *in vivo* screening of FMCAP in female NOD/SCID mouse model showed a high rate of reduction of liver HBV DNA level in comparison to entecavir. In chimeric mice harboring triple mutants, FMCAP also effectively reduces the viral load while entecavir was ineffective.

These results encourage us to conduct additional biological evaluations to determine FMCA and FMCAP as potential anti-HBV candidates. Consequently, the development of an efficient synthetic methodology was critically needed. Initially, FMCA was synthesized in 22 steps by a very inefficient process. Thus, the synthetic process was needed to revise, in which a stereo-selective synthesis of FMCA was developed *via* Vince lactam.[3] However, due to the low yield of certain steps, the utility of the process was also limited for a large-scale synthesis. Herein, we report an efficient and practical synthesis of FMCA in 8 steps using commercially available ketone 1 as shown in Scheme 1.

REF.

1. *Bioorg Med Chem Lett* 2011, 21, 6828.

2. *Bioorg Med Chem Lett* 2013, 23, 503.

3. *J Org. Chem* 2014, 79, 3917.

15. Development of a Rapid In Vitro Assay for Identification of Protein-Protein Interactions in Zika Virus

Shayli Varasteh Moradi, PhD, Dejan Gagoski, PhD, Wayne A. Johnston, PhD, Kirill Alexandrov, PhD
Institute for molecular biosciences, The University of Queensland, Brisbane

The rapid spread of Zika virus (ZIKV) has become a serious public health threat owing to its link to severe neurological disorders such as fetal microcephaly and Guillain-Barré syndrome in adults.

In this study, we attempt to elucidate viral protein interactions in ZIKV which would provide important insight into the understanding of biological function and pathogenesis of virus. We established a robust *in vitro* approach based on AlphaLISA technology in combination with *Leishmania tarentolae* cell-free protein production (LTE) system for this interaction analysis. One of the advantages of using this approach is the ability to identify protein interaction network without any purification and washing steps that results in a rapid screening analysis. Both structural and non-structural of ZIKV proteins were generated in LTE system. Following the co-translation of protein pairs, the physical interaction between viral proteins was determined by AlphaLISA approach as a high-throughput screening assay. All possible pair-wise interactions between viral proteins were tested which led to the identification of 54 intra-viral protein-protein interactions from which some interactions were found to be novel. Using this technology, we could rapidly analyse the protein interaction network in ZIKV enabling us for further investigations to decipher the biological activity of the protein complexes.

16. In Vivo Efficacy of Oral Treatment with Pritelivir Against Acyclovir Resistant Herpes Simplex Virus Type 1 or 2 in BALB/c Mice

Debra Quenelle, DVM, PhD[1], Alexander Birkmann, PhD[2], Thomas Goldner, PhD[2], Tamara Pfaff, DVM[2], Holger Zimmermann, PhD[2], Susanne Bonsmann, PhD[2], Deborah Collins, BS[1], Terri Rice, BS[1], Emma Harden, BS[1], Mark Prichard, PhD[1]

[1]The University of Alabama School of Medicine, Birmingham, Alabama, , United States of America; [2]AiCuris Anti-Infective Cures GmbH, Wuppertal, Germany

Pritelivir (PTV) has excellent activity both in vitro and in vivo against herpes simplex viruses (HSV). Mice were lethally infected intranasally with Acyclovir (ACV) resistant HSV type 1, strain 11360 or type 2, strain 12247. All treatments were initiated 24-72 h post viral infection and given twice daily for 7 consecutive days. PTV was used in mice at 30, 10, 3, or 1 mg/kg/dose (60, 20, 6 or 2 mg/kg/day) at +24 h post viral intranasal inoculation with HSV-1, strain 11360 and reduced mortality at all doses ($p<0.001$). Acyclovir (ACV) was given similarly at 50, 25, 12.5 and 6.25 mg/kg/dose and only the 50 mg/kg reduced mortality ($p<0.05$). When delayed, PTV treatments of 3 and 1 mg/kg doses were effective ($p<0.01$) in reducing mortality at +48 h and also effective at 3 mg/kg for +72 h. Doses of 50 – 6.25 mg/kg of ACV were ineffective at either time against strain 11360. When PTV was used in combination with ACV, there was no antagonism in efficacy against HSV-1. For the HSV-2, strain 12247, PTV was effective at similar doses when treatments were initiated 24 h post viral inoculation while ACV was ineffective at all doses. PTV was effective at 3 mg/kg for +48 or +72 h and 1 mg/kg was also effective at +72 h. ACV was again ineffective at all doses and times. PTV has potent antiviral efficacy against ACV resistant strains and has the potential for the treatment of serious HSV type 1 and 2 infections in humans.

17. HIV-1 Tropism in Patients Living in Grodno Region of Belarus

Natallia Matsiyeuskaya, MD, PhD[1], Irina Tokunova, BS[1], Dmitry Kireev, PhD[2]

[1]State Medical University, Grodno, Belarus; [2]Central Research Institute for Epidemiology, Moscow, Russian Federation

AIM OF STUDY: to establish tropism and HIV-1 subtypes on the nucleotide sequence of V3 loop envelop protein gene (gp120) in HIV infected patients living in Grodno region of Belarus. Detection of HIV tropism was performed by sequencing of V3 loop of env gene by "AmpliSens HIV-Resist-Seq» (Russia) in 98 HIV-infected patients. Algorithm at http://www.geno2pheno.org/ (FPR = 20%) was used. HIV subtype was carried out on the nucleotide sequences of the envelop protein gene fragment using the resource REGA HIV-1 Subtyping Tool – Ver, 3.0. Phylogenetic analysis of the sequences of the HIV V3 loop env was done using software MEGA ver.6.06.

RESULTS. The number of patients infected by R5-tropic HIV was 63 (64,3%): age, Me – 36 (30-43) years, AIDS was in 19 (30,2%). The number of ones infected by non R5-tropic HIV was 35 (35,7%): age, Me – 35 (30-42) years, AIDS was in 13 (33,3%) patients ($p>0,05$). Subtype A HIV-1 was detected in 91 (96,8%) patients, subtype B - in 3 (3.2%). Estimated number of patients in the Grodno region infected by R5-tropic HIV on 01.06.2016 was 449 [95% CI: 424 – 474]; infected by not R5-tropic HIV was 250 [95% CI: 226 – 270].

CONCLUSIONS. Subtype A HIV-1 have been revealed in 96,8% cases in observed group. Isolated samples had the high similarity of the nucleotide sequences with isolates obtained in Russia and Ukraine. Presently the number of patients infected by R5-tropic HIV in Grodno is in 1.8 times higher in comparison with non R5-tropic HIV.

18. Design and Synthesis of Biogenic Silver Nanoparticles from Ethanoilc Leaf Extract of *Andrographis peniculata* and Exploration of its Anticancer Activity

Periyasamy Selvam, PhD[1], Ashish Wadhwani, PhD[2], Sameer Kumar Panda, MS[2]
[1]*International Research Centre, Kalasalingam University, Krishnankoil 626126, Tamilnadu, India;*
[2]*Dept. of Pharmaceutical Biotechnology, JSS College of Pharmacy, JSS University, Ooty, Tamilnadu, India*

Andrographis peniculata (Acanthaceae) is novel medicinal plant, enriched with potential bioactive molecules and used in Indian System of medicine for the treatment of Dengue and cancer. Thus, the study planned to biosynthesize anticancer potent silver nanoparticles using ethanol leaf extract of *Andrographis peniculata* (AP-ET). The synthesized nanoparticles (AP-ET AgNPs) were confirmed by color transformation and UV spectrophotometry. The size and morphology of the silver nanoparticles were characterized by SEM and EDEX. The stability of silver nanoparticles was detected by FT-IR and PXRD. Anticancer activity of ethanol, ethyl acetate extracts and Silver nanoparticles tested against Neuroblastoma cell and cytotoxicity was also investigated against Vero cells by MTT assay. The appearance of reddish brown colour and UV absorption range 437 nm confirmed the synthesized silver nanoparticles. The silver nanoparticle showed spherical structure and their size were ranging from 20-70 nm under SEM observations. FT-IR spectra of silver nanoparticles showed the peaks for the functional groups, N-H, C=O, -C=C, C-H, which indicate the stability of synthesized AP-ET AgNPs. AP-ET AgNPs showed anticancer activity against Neuroblatoma cells with IC_{50} value of 90 µg/ml, whereus ethanol and ethyl acetate extracts were found to be 166 and 152 µg/ml, respectively, Cytotoxicity of AP-ET AgNP in Vero cells was found to be 210 µg/ml, ethanol and ethyl acetate extracts were found to be 324 and 226 µg/ml, respectively. AP-ET AgNPs exhibit significant anticancer activity against neuroblastoma than extracts. *Andrographis peniculata* silver nanoparticles would be helpful for designing of cytotoxic agents against Neuroblatoma cells to destroy cancer cells.

19. Surface Modified Chitosan Nanoparticles for Selective Targeting of Lamivudine to Hepatocyte

Mani Bhargava, MD, MPH[1], Vishal Bhargava, PhD[2], Saurabh Bhargava, MD, MPH[3]
[1]*ICFAI University, Knp, India;* [2]*GTB Hospital, India;* [3]*Himalayan University, India*

Hepatitis B is an infection of the liver caused by the hepatitis B virus(HBV). It is a major cause of infectious liver disease throughout the world. Viral hepatitis resides primarily in the liver; hence drug targeting with ligand anchored moiety can be an effective strategy in management of this disease. Lamivudine a "nucleoside analogue" is commonly used in treatment of Hepatitis B and effectively inhibit viral replication, however it shows extra-hepatic toxicity.

The project envisaged that use of receptor-mediated endocytosis may permit the realization of potential of drug targeting that reduces side effects. This necessitates developing surface modified chitosan nanoparticles for hepatocyte selective targeting via conjugation of a ligand (glycyrrhizin).

The chitosan nanoparticles were prepared by Low Molecular Weight Chitosan(LMWC) by Ionotropic gelation method and ligand was anchored.The nanoparticles were then characterized *in-vitro* for their shape, size, drug entrapment, in-vitro drug release and stability. The *in-vivo* study comprised of biodistribution studies in various organs and fluorescence microscopy was performed, hematological and histological examinations were done.

Finally it could be concluded that encapsulation of lamivudine in glycyrrhizin coupled LMWC nanoparticles enhances the residence time. Further bioavailability of the drug in liver is increased which could be utilized in reducing the dosing frequency as well as the dose. This could help in the reduction of dose related toxicity associated with this antiviral drug. Ligand mediated bio-deposition and cellular interaction of LMWC nanoparticles especially at the site would be a focal paradigm for the upcoming research in the field of antiviral drug delivery.

20. Evidence that ER α-Glucosidase Inhibitors Potentiate Other Broad-Spectrum Antivirals Against Multiple Families of Hemorrhagic Fever Viruses In Vitro and In Vivo

Jinhong Chang, MD, PhD[1], Julia Ma, BS[1], Xuexiang Zhang, MS[1], Travis Warren, PhD[2], Veronica Soloveva, PhD[2], Fang Guo, MD, PhD[1], Qing Su, PhD[1], Shuo Wu, PhD[1], Helen Shen, PhD[3], Eric Solon, PhD[3], Yanming Du, PhD[1], Sina Bavari, PhD[2], Ju-Tao Guo, MD1, Timothy Block, PhD[1]
[1]Baruch S. Blumberg Institute; [2]United States Army Medical Research Institute of Infectious Diseases; [3]QPS, LLC

Targeting host functions essential for viral replication has potential to be a broad spectrum and resistance-refractory therapeutic approach. However, up to now only a few of the host factors have been validated as broad-spectrum antiviral targets *in vivo*. ER α-glucosidases I and II have been demonstrated to be involved in the morphogenesis of many enveloped viruses. *In vivo* antiviral efficacy of various iminosugar based ER α-glucosidase inhibitors has been reported in animal models of Dengue, Japanese encephalitis, Ebola, Marburg and influenza viruses. Here we report establishment of Huh7.5 based cell lines with ER-α-glucosidases I and II knockout using CRISPR-CAS technology, and that the replication of Dengue and Yellow fever virus can only be partially inhibited in compete absence of ER-α-glucosidases I or II. This result supported the notion that, similar to other host targeting antivirals, glucosidase inhibitor therapeutics is likely associated with limited antiviral potency. To further explore the possibility of improving antiviral potency, we examined the effect of our lead iminosugar compound IHVR-19029 in combination with other broad-spectrum antivirals. *In vitro* antiviral studies showed that IHVR-19029, in combination with T-705, synergistically inhibited the replication of dengue and Ebola viruses. Moreover, in a mouse model of Ebola virus infection, we demonstrated that combination of sub-optimal doses of IHVR-19029 and T-705 significantly increased the survival rate. Encouraged by these results, we are currently extending iminosugar study to other emerging flaviviruses such as Yellow fever and Zika virus as well as combination with other antivirals such as BDAA or Sofosbuvir.

21. Evaluation of Cross-Sstrain Neutralizing Potency of Monoclonal Antibodies against Crimean-Congo Hemorrhagic Fever Virus

Marko Zivcec, PhD, Lisa Guerrero, MS, Cesar Albarino, PhD, Eric Bergeron, PhD, Christina Spiropoulou, PhD
VSPB, DHCPP, NCEZID, CDC, Atlanta, Georgia, United States of America

Crimean-Congo hemorrhagic fever (CCHF) is an often life-threatening, viral, human disease that is emerging, or re-emerging, in a wide geographic area. The efficacy of antivirals targeting its causative agent, CCHF virus (CCHFV), remains controversial. MAbs targeting the CCHFV surface glycoproteins, Gn and Gc, have previously shown neutralization efficacy against the prototype CCHFV strain, IbAr10200, and/or protection in a lethal suckling mouse model of CCHF. However, due to extensive sequence diversity of CCHFV Gn and Gc, whether these MAbs neutralize other CCHFV strains is unknown. We initially used our CCHF virus-like particle (VLP) system to generate 12 VLP moieties, each containing a different set of glycoproteins, in order to efficiently screen MAbs in BSL-2 conditions. We obtained a panel of 14 previously reported mouse MAbs targeting Gn or Gc, and screened them for neutralizing activity against our panel of CCHF VLPs. Of the MAbs tested, 3 Gc-targeting MAbs (8A1, 11E7, and 30F7) demonstrated cross-neutralizing activity against CCHF VLPs, with 8A1 neutralizing all VLPs tested. To confirm our findings, we tested 8A1, 11E7, and 30F7 against a panel of CCHFV strains. In line with the VLP data, 8A1 maintained strong neutralizing potency against all tested CCHFV strains, while the neutralizing potency of 30F7 and 11E7 was more variable. This study highlights an important role CCHF VLPs could play in future efforts to screen CCHFV antivirals in the absence of high containment laboratories and provides the first evidence that a single MAb can effectively neutralize a number of diverse CCHFV strains in vitro.

22. Combining Hepatitis B Surface Antigen with Tetanus for a Single Oral Vaccine

Mani Agarwal, BS[1], Vishal Bhargava, MD, PhD[2]
[1]GTB Hospital, Sagar, India; [2]KRV Hospitals, Kanpur, U.P., India

Infections are still leading cause of morbidity and mortality and most of which can be prevented by vaccination. However, there are too many vaccines to be administered, increasing cost of immunization. Combination vaccines can answer these problems by development of single vaccine containing all possible antigens.

The goal of present study was to see the effect of 2 antigens when given in combination. Bilosomes can provide needle free, painless approach for immunization. Recombinant hepatitis-B surface antigen(HBsAg) and recombinant protective antigen(rPA) were candidate antigens.

Bilosomes containing rPA and HBsAg were prepared by lipid cast film method. Antigen loaded bilosomes were characterized *in-vitro* for shape, size, antigen entrapment and stability in various body fluids. Fluorescence microscopy was done to confirm the uptake of bilosomes. The *in-vivo* study comprised of immunization of Balb/c mice and estimation of IgG response in serum and sIgA in various body secretions using specific ELISA.

Bilosomes formed were multilamellar and stable in gastric and intestinal fluids. Fluorescence microscopy suggested that bilosomes were taken up by gut associated lymphoid tissues. *In-vivo* data demonstrates that combination produced both systemic as well as mucosal antibody responses upon oral administration at higher dose levels as compared to intramuscular immunization but fail to produce any synergistic effect.

When rPA and HBsAg given in combination, HBsAg(high dose) potentiates the production of anti-rPA antibody. Also they elicited measurable sIgA in mucosal secretions, while alum adsorbed antigens failed to elicit such responses. The combination produced both systemic as well as mucosal antibody responses upon oral administration.

23. Antibody Coated Liposomes for Transmucosal Vaccination

Sourabh Jain, MD, MPH[1], Saurabh Bhargava, MD, MPH[2]
[1]Bhagyodaya Pharmacy College, Sagar, M.P., India; [2]Himalayan University, Kanpur, u.p., India

The critical role of vaccine delivery system in "rational vaccine design" has been widely recognized. Thus research work was envisaged involving development of antibody coated liposome for transmucosal immunization against hepatitis-B which may offer increased uptake of nanoliposome through transmucosal surface of nasal route and sustaining release of HBsAg to evoke relatively high IgA titre in mucosal surface.

Liposomes were prepared by a lipid cast film method & then IgG antibody was cross linked on the surface. Coated liposomes were characterized *in-vitro* for their shape, size, percent antigen entrapment and stability. Fluorescence microscopy was performed to confirm the deposition pattern in respiratory tract. The *in-vivo* part of the study comprised of estimation of IgG response in serum and sIgA in various body secretions using specific ELISA.

Observation of fluorescence images of nasal mucosa, lungs and spleen, revealed that these antibody coated liposome, were significantly taken up by mice respiratory mucosal surface, which made them promising carriers for mucosal vaccination.

Considerable immune responses were produced by the developed system that may be due to the induction of MALT as well as contribution of the peripheral airways. The higher immunity induced by ACL HBsAg may be attributed to its cationic nature, antibody coating and subsequent mucoadhesive property. Thus mucosal immunization with lipid vesicle through nasal administration may be effective in prophylaxis of diseases transmitted through mucosal routes as well as systemic infections. The strategy can be made more appropriate by determination of paracellular transport, nasal mucociliary clearance, mucosal toxicity assessment etc.

24. Identification of HSP-90 Inhibitors as Potential Anti-HIV Molecules

Jay Trivedi, MS[1], Afsana Parveen, MS[2], Ashoke Sharon, PhD[2], Debashis Mitra, PhD[1]
[1]National Centre for Cell Science, Pune, India; [2]Birla Institute of Technology, India

HIV-1 is the causative agent of acquired immune deficiency syndrome (AIDS) which is responsible for more than 37 million deaths till date. With an arsenal of more than 25 US FDA approved drugs against HIV, we have succeeded to control AIDS to an extent but have failed to eradicate the disease. With a genomic RNA of around 10 Kb coding for 15 proteins, HIV is unable to complete its life cycle on its own. It efficiently hijacks the host cellular machinery to complete its lifecycle and to escape the human immune defence system. Almost all the events of HIV life cycle are predominantly regulated by a large number of cellular factors. Hence, targeting cellular factors in association with viral factors may give us an advantage in the continuing fight with the virus. Our studies to identify such cellular factors, inhibition of which will not affect the host but at the same time will inhibit virus propagation have led to identification of selected heat shock proteins including HSP90. Inhibitors of HSP90 seem to significantly inhibit HIV-1 replication in HIV-1 infected T-cell lines and PBMCs. In the present study, we have screened several HSP90 inhibitors and their novel analogues for anti-HIV activity and have also analysed their therapeutic index. Some of these analogues have shown significant anti-HIV activity in T-cells including one which seem to have several fold better therapeutic index than the parent molecule. Taken together, our results suggest that HSP90 inhibitors can be studied further for potential anti-HIV therapeutic strategy.

25. Disulfiram Can Inhibit MERS and SARS Coronavirus Papain-Like Proteases via Different Modes

Min-Han Lin, BS, Chih-Hua Hsieh, BS, Chi-Yuan Chou, PhD
Department of Life Sciences, National Yang-Ming University, Taipei, Taiwan

Middle East respiratory syndrome coronavirus (MERS-CoV) is a new highly pathogenic human coronaviruses that emerged in Jeddah and Saudi Arabia and has quickly spread to other countries in Middle East, Korea, Europe and North Africa since 2012. Up to 10 February 2017, it has infected at least 1905 people with a fatality rate of about 36% globally. It far exceeds severe acute respiratory syndrome coronavirus (SARS-CoV), which caused global outbreak with a fatality rate around 10% in 2003. This has resulted in an urgent need to identify antiviral drugs that are active against MERS-CoV. Recently, we have found that an alcohol-aversive drug, disulfiram, is an inhibitor against the papain-like protease (PL^{pro}) of MERS-CoV and SARS-CoV. Interestingly, the inhibition studies by enzyme kinetics suggested that disulfiram acts as an allosteric inhibitor on MERS-CoV PL^{pro}, whereas it acts as a competitive inhibitor on SARS-CoV PL^{pro}. Desalting of SARS-CoV PL^{pro} incubated in a solution containing disulfiram didn't eliminate the inhibitory effect of disulfiram, indicating a time-dependent inhibition via affinity labelling. The results imply that the inhibitor recognition specificity may be different between these two coronaviral PL^{pro}s, despite the similarity of overall structures and catalytic sites.

26. Anti-Hepatitis B Virus (HBV) Activity of Novel Pyrimidotriazinone Derivatives Through the Inhibition of Viral Nucleocapsid Assembly

Masaaki Toyama, PhD[1], Norikazu Sakakibara, PhD[2], Takayuki Hamasaki, PhD[1], Mika Okamoto, MD, PhD[1], Koichi Watashi, PhD[3], Takaji Wakita, MD, PhD[3], Masanori Baba, MD, PhD[1]
[1]Kagoshima University, Japan; [2]Tokushima Bunri University, Japan; [3]National Institute of Infectious Diseases, Japan

Chronic human hepatitis B virus (HBV) infection is currently treated with nucleoside analogs, including lamivudine, entecavir, and tenofovir. They are potent inhibitors of HBV DNA polymerase, which also functions as reverse transcriptase. Although these compounds are effective in treatment of infected patients, emergence of drug-resistant mutants and viral reactivation after treatment interruption are still major concerns in antiviral chemotherapy against HBV. Therefore, it is still mandatory to identify and develop novel inhibitors that target a step other than reverse transcription in HBV replication cycle. HBV capsid assembly is a critical step for viral replication and an attractive target for inhibition of HBV. To find novel inhibitors of HBV, we have established an in silico screening system based on X-ray crystallography

of HBV core protein and examined 170,000 compounds for their interaction with the core protein. Fifteen compounds with high docking scores were selected, and they were examined for their inhibitory effect on HBV replication in HepG2.2.15.7 cells. Among them, 4-(4-tert-butylphenyl)-2-[(2,3-dichlorophenyl) amino]-8-methyl-3H,4H,6H-pyrimido[1,2-a][1,3,5]triazin-6-one was found to be a selective inhibitor of HBV replication. Its 50% effective concentration (EC_{50}) and 50% cytotoxic concentration (CC_{50}) of were 5.3±0.4 and >100 μM, respectively. The compound reduced HBV capsid formation and capsid-associated HBV DNA and RNA in a dose-dependent fashion but did not affect the amount of HBc protein. These results indicate that the pyrimidotriazinone derivative inhibits HBV replication through blocking the capsid assembly and is considered to be a promising lead of novel anti-HBV agents. Further studies are in progress to optimize their chemical structures.

27. Solid Lipid Based Nanoparticulate System for Effective Vaccine Delivery

Aakanchha Jain, PhD[1], Piush Khare, PhD[2], Saurabh Bhargava, MD, MPH[3]
[1]DOPS, Sagar, Sagar, M.P., India; [2]United Institute of Pharmacy, Allahabad, U.P., India; [3]Himalayan University, Kanpur, U.P., India

The search for innovative ways of vaccination has intensified recently with declining vaccine coverage and growing public concern about new virulent disease outbreaks. The work envisaged here explores potential of Solid Lipid Nanoparticles(SLN) in efficient protein delivery through surface modifications using subcutaneous route(SC).

The SLN were prepared by Solvent Injection Method & optimized. The characterization included Transmission & Scanning Electron Microscopy, X-Ray Diffraction Analysis, *In-vitro* release, Kinetics of uptake by flow cytometer, Evaluation of cell apoptosis, T-cell proliferative assay, TH1/TH2 cytokine profile and Internalization studies by spectral bioimaging. *In-vivo* study comprised fluorescence studies and estimation of IgG in serum, sIgA in various body secretions using specific ELISA.

The particulate system is better carrier system for immunization because of less diffusivity and restricted movement.SLNs themselves act as signal for the phagocytic cells.Surface modified SLNs can entrap greater amount of antigen, provide its sustained release and rapidly internalized by the antigen presenting cells. *In-vitro* T-cell proliferation and induction of TH1 type of immune response clearly marks, potential of this novel carrier system. Fluorescence studies showed better uptake of surface modified SLNs. Higher and more sustained antibody titer obtained with surface modified SLNs suggests their better immunological potential. Thus, SC immunization could be an efficient alternative approach for vaccination against hepatitis.

The formulations developed in this study can be further explored for the incorporation and delivery of other proteins and peptides should subsequently be subjected to pilot plant scale-up & clinical trial to establish their potential for subcutaneous immunization against hepatitis-B.

28. Development of Engineered Nanocarrier for Controlled Delivery of a Protease Inhibitor

Saurabh Bhargava, MD, MPH[1], Vishal Bhargava, MD, MPH[2], Aakanchha Jain, PhD[3]
[1]Himalayan University, Kanpur, India; [2]GTB Hospitals, Kanpur, India; [3]DOPS, Sagar, India

AIDS is a chronic, progressive syndrome, characterized by intense viral replication and profound immunosuppression, resulting in the development of life threatening opportunistic infections. HIV infection leads to deterioration of immune functions.

The objective of the present study was to develop engineered nanocarriers for controlled delivery of a protease inhibitor(lopinavir). The uncoupled Solid Lipid Nanoparticles(SLN) were prepared by solvent diffusion method and then coupled with mannose. Characterization studies were done by scanning & transmission electron microscopy (SEM & TEM). X-ray diffraction(XRD) and *Differential scanning calorimetry(DSC) studies were performed along with* the *in-vitro* studies followed by *in-vivo* studies on albino rats.

In-vitro and *in-vivo* studies results show mannose coated SLNs (MSLN) deliver their contents to macrophage rich organs and tissues, which are the reservoir of HIV. Low elimination and better distribution profile can be achieved by MSLNs. The dose of the antiviral agent can be reduced due to the site-specific delivery from this carrier.

Conclusively, ligand-mediated bio-disposition and cellular interaction of MSLNs, especially at the target sites, would be a focal paradigm for upcoming research in the field of anti-HIV drug delivery. MSLNs have paved the way for the bio-stable, site-specific and ligand-mediated delivery systems with desired therapeutics.

29. NonInvasive Topical Immunization Using Cholera Toxin as Adjuvant for the Treatment of Hepatitis B

Gomed Agarwal, MD, MPH, Piush Khare, PhD
[1]*United Institute of Pharmacy, Allahabad, India*

The search for innovative ways of vaccination has intensified recently with declining vaccine coverage and growing concern about new virulent disease outbreaks. Immunization is a prophylactic approach through which body is shielded from any incoming pathogenic invasion. Immune response elicited by Topical Immunization depends upon structure and composition of skin of target species. It provides access to local skin immune system which is dominated by langerhans cells that can be manipulated by adjuvants to orchestrate specific, robust immune response. Topical vaccination induces systemic and mucosal antibodies to coadministered antigen and moreover it avoids first pass phenomenon and also protects antigen from enzymes that are present in gut wall.

Niosomes are nonionic surfactant based vesicles that were used as topical carrier for immunogens for transdermal delivery. The goal of the present study was to investigate potential of niosomes as carrier for HepatitisB antigen(HbsAg) with cholera toxin(CTB) as adjuvant. Niosomes prepared by Sonication. Antigen loaded Niosomes were characterized *invitro* for their shape, size, %antigen entrapment and stability. Confocal laser scanning microscopy(CLSM) was carried out to confirm uptake of Niosomes. The *invivo* part of study comprised of immunization of Balb/c mice and estimation of IgG response in serum and sIgA in various body secretions using specific ELISA.

Niosomes formed were multilamellar and stable. Presence of fluorescence at different skin depths reflected accumulation of these niosomes in the region of epidermis, suggesting better uptake of antigen by langerhans cells. Based on the results obtained, niosomes presented its potential for antigen delivery through transcutaneous route.

30. *In Silico* Discovery of a Protein-Protein Interaction Inhibitor for Influenza Viruses

Gregory Mohl, BS, David Busath, MD
Brigham Young University, Provo, Utah, United States of America

Resistance makes the influenza virus a difficult target for antiviral development. Amantadine and rimantadine are now ineffective against most circulating strains due to point mutations in the M2 channel, and neuraminidase inhibitors are also vulnerable to viral resistance. This highlights the need for the discovery of antivirals that inhibit infection by a new mechanism. We set out to design antivirals that would block influenza replication by inhibiting the interaction of Polymerase Basic 1 (PB1), a subunit of the viral polymerase complex, and Ran-Binding Protein 5 (Ran BP5), a host importin, with a small molecule that would bind to PB1. Compounds that might block nuclear import of the polymerase complex were identified using high-throughput virtual screening coupled with *in vitro* validation assays. The Asinex protein-protein interactions library was docked with Glide to the PB1 subunit of 4WSB. The proposed binding site was chosen due to its proximity to the bipartite nuclear localization signal (NLS), and the top hits were rescreened using a higher exhaustiveness to confirm the results. Four compounds were chosen and tested in cell culture for inhibition of viral replication with an immunofluorescence assay and for cytotoxicity with a vital dye uptake assay. A lead compound was identified that has an EC_{50} of 9.1 μM against A/Victoria/3/75, an EC_{50} of 9.5 μM against A/WS/33, and an EC_{50} of 19.8 μM against A/Taiwan/64. This lead compound is amenable to medicinal optimization.

31. Human Genetic Predisposition to Diseases Caused by Viruses from Flaviviridae Family

Andrey Barkhash, MD, Aida Romaschenko, PhD
Institute of Cytology and Genetics SB RAS, Novosibirsk, Russian Federation

Tick-borne encephalitis (TBE) and chronic hepatitis C (HC) are common diseases in the territory of Russia. These diseases differ significantly in pathogenesis, although both of them are caused by single-stranded positive-sense RNA viruses with similar genome organization from the Flaviviridae family. It is still not clear whether molecular protective mechanisms against these two related viruses are similar or different; however, human genes that encode crucial components of antiviral immune response are most likely involved in these mechanisms. We are for the first time studying possible common genes and their polymorphisms involved in predisposition to these diseases in the same human population (Russians from Novosibirsk city). To date, more than 70 polymorphisms located within more than 15 candidate genes were studied. In total, we found ten SNPs within six genes that are associated with predisposition to TBE, and three of these SNPs were also associated with predisposition to HC. The review of our previously obtained already published data (Barkhash et al., 2010, 2012, 2013, 2014, 2016), as well as our new unpublished results on the topic, will be presented in the report. Identification of genetic variants associated with the predisposition to severe viral diseases is important for increased understanding of viral pathogenesis mechanisms and in future may be useful for the development of the specific therapy for treatment of these virus infections.

32. Establishment of an Antiviral Assay System and Identification of Severe Fever with Thrombocytopenia Syndrome Virus (SFTSV) Inhibitors

Masanori Baba, MD, PhD[1], Masaaki Toyama, PhD[1], Nobukazu Sakakibara, PhD[2], Mika Okamoto, MD, PhD[1], Masayuki Saijo, MD, PhD[3]
[1]Kagoshima University, Kagoshima, Japan; [2]Tokushima Bunri University, Sanuki, Japan; [3]National Institute of Infectious Diseases, Tokyo, Japan

Severe fever with thrombocytopenia syndrome (SFTS) is an emerging tick-borne infectious disease. SFTS is epidemic in Asia, and its fatality rate is around 30% in Japan. The causative virus SFTSV is a phlebovirus of the family *Bunyaviridae*. Although effective treatments are required, there are no antiviral agents currently approved for clinical use. Ribavirin and favipiravir have been examined for their anti-SFTSV activity and found that both compounds selectively inhibit SFTSV replication *in vitro*. However, their activity is not high enough. Therefore, it is mandatory to identify novel compounds active against SFTSV. To this end, we have established a safe and rapid assay system for screening selective inhibitors of SFTSV. The virus was isolated from SFTS patients treated in Kagoshima University Hospital. Vero cells were infected with SFTSV (multiplicity of infection = 0.01) and incubated with various concentrations of test compounds. After 3 days, the cells were treated with TaqMan Gene Expression Cells-to-CT™ Kit (Thermo Fisher Scientific) and examined for intracellular viral RNA levels by real-time reverse transcription (RT)-PCR. The cytotoxicity of test compounds was determined by a tetrazolium dye method. Among the test compounds, the antimalarial agent amodiaquine was identified as a selective inhibitor of SFTSV replication. Its EC_{50} and CC_{50} were 19.1 ± 5.1 and > 100 μM, respectively. The EC_{50} value was comparable to or slightly lower than those of ribavirin and favipiravir. Thus, amodiaquine is considered to be a promising lead of novel anti-SFTSV agents, and further experiments are in progress to evaluate its derivatives.

33. A Proof-of-Concept Study for the Prevention of HIV Infection in 'Immune Humanized' Mice Using a Novel Non-nucleoside Reverse Transcriptase Inhibitor

Steve Ludmerer, PhD[1], Gary Adamson, PhD[1], Jaume Balsells-Padros, PhD[1], Michael Brehm, PhD[2], Christopher Bungard, PhD[1], Christopher Burgey, PhD[1], Smita Jaiswal, PhD[2], Christopher John, PhD[1], Bonnie Howell, PhD[1], Lee Klein, PhD[1], Maya Lipert, PhD[1], Jeremy Luban, PhD[2], Karsten Menzel, PhD[1], James Perkins, PhD[1], Elena Trepakova, PhD[1], Deping Wang, PhD[1], Ming-Tain Lai, PhD[1]

[1]Merck & Co, West Point, Pennsylvania; [2]University of Massachusetts Medical School, Worcester, Massachusetts

Every year 2.0 million people become HIV-1 infected. To reduce this burden, it is important to introduce prophylactic administration of potent HIV drugs capable of preventing viral transmission, especially for high risk populations. We describe a non-nucleoside reverse transcriptase inhibitor (NNRTI) which demonstrates potent activity against HIV-1 *in vitro* (IC_{50} 7.8 nM in 100 % NHS). Low solubility (0.4 ug/ml in PBS) favorable for slow dispersion as a suspension coupled with moderate clearance in pre-clinical animal species suggest it has suitable pharmaceutical properties for a candidate long-acting parentally administered drug. A proof-of-concept prophylaxis study was conducted in mice bearing a humanized immune system. Two groups of 6 mice received a single intramuscular injection of drug (9 mg) or placebo. Subsequently they were infected intraperitonally with 100,000 TCID units of HIV-1. Blood was collected every two weeks prior to a further viral challenge through five viral challenges. Due to the fragility of the humanized mice, only three animals in the drug group and four in the control group survived through study completion. While mice receiving placebo became viremic (range 47,200-197,200 genomes/ml), the three animals in the NNRTI group had undetectable virus for a total duration of 10 weeks protection. The mean plasma drug concentration at study end was 200 nM, 25-fold above the IC_{50} of 7.8 nM. The study demonstrates the potential use of a NNRTI as a prophylactic agent for long-term protection against HIV. Additional studies using vaginal/rectal routes of viral transmission are in progress, and data will be presented.

34. Development and Characterization of an Aerodynamic System for Pulmonary Delivery of Influenza Vaccine

Mani Bhargava, MD, MPH[1], Saurabh Bhargava, MD, MPH[2], Piush Khare, PhD[3]

[1]ICFAi Univ, Kanpur, India; [2]Himalayan University; [3]United Institute of Pharmacy

The name *influenza* is Italian and means "influence", Commonly referred to as the flu, is an infectious disease caused by RNA viruses of the family Orthomyxoviridae , that affects birds and mammals. The aim is also to develop and characterize aerodynamic systems with r-H1N1Ags for safely deposition in alveoli to enhance the bioavailability and control release of influenza antigen after pulmonary administration in animal model. This Induces not only systemic humoral (IgG) responses, but also cell-mediated (IL-4, IFN-γ) and mucosal immune responses (IgA, IgG), non-invasive, propellant & needle free delivery of vaccine.

The chitosan microparticles were prepared by ionic gelation method of chitosan with tripolyphosphate(TPP). The formulations were optimized on the basis of particle size, tap density & entrapment efficiency. The external morphology of the optimized formulation was studied by TEM & SEM. The zeta potential was determined along with stability studies at accelerated tempreatures. The in-vivo studies involved determination of antibody titres in serum and mucosal secretions and uptake studies by fluorescence microscopy.

The results show that as the preparation was reduces to lyophilized form which increased the stability as compared to conventional liquid formulations. The microparticles of uniform size distribution were obtained owing to the repulsion between the positively charged particles. The fluorescence images show the uptake of microparticles by various organs and the ELISA results shows comparable IgG responses along with IgA.

Thus, Antibody production was found to be more in pulmonary route as compared to other routes.

35. Pharmacologic Strategy Against Lassa Virus via Targeting Human Preprotein Vonvertase Site 1 Protease Upstream Glycoprotein Cleavage Pathway

Olaposi Omotuyi, PhD[1], Nash Oyekanmi, PhD[2]
[1]Adekunle Ajasin University, Akungba-Akoko, Nigeria; [2]National Biotechnology Development Agency, Abuja, Nigeria

Lassa virus (LASV) is an arenavirus that causes Lassa hemorrhagic fever in human and non-human primates. LASV fever is highly predominant in West African with 300,000 and 500,000 suspected cases anaually, resulting in 5,000 deaths/year. In 2015, 108 Lassa virus fever-related deaths were recorded in Nigeria. Whilst ribavirin has shown some clinical promise in early stage infection, there is no pharmaceutical agent potent in late stage of infection. This study exploited host sterol regulatory element-binding protein (SREBP) site 1 protease (S1P)/LASV pre-glycoprotein processing mechanism in anti-LASV drug development. Computationally derived PF-429242/S1P complex was used to screen chemical libraries following docking validation (r^2=0.6); LAS981 (dG= -8.2 *vs.* -5.6 (PF429242)) was selected for further studies. In animal studies, LAS981 and PF429242 down-regulated SRE target genes (HMG-CoA synthase, Glucose-6-phosphate dehydrogenase, Acetyl-CoA carboxylase) but not control genes (cyclophilin and glyceraldehyde-3 phosphate dehydrogenase) and blocked Activating Transcription Factor 6 processing. Direct S1P inhibition was confirmed in a dose dependent inhibition experiments with IC50 values of 1.375-μM *vs.* 0.37-μM for PF429242 and LAS981 respectively. LAS981 may be representative antiviral agent against prototypic arenavirus targeting pre-glycoprotein processing mechanism.

36. Selective Inhibitor of Nuclear Export (SINE) Compounds Reduce RSV Replication in vitro

Cynthia Mathew, PhD[1], Reena Ghildyal, PhD[1], Patricia Jorquera, PhD[2], Sharon Tamir, PhD[3], Jennifer Pickens, PhD[2]
[1]University of Canberra, Canberra, Australia; [2]Department of Infectious Disease, University of Georgia, Athens, Georgia, United States of America; [3]Karyopharm Therapeutics Inc, Newton, MA, USA, 2459, Newton, Massachusetts, United States of America

Human Respiratory Syncytial Virus (RSV) is the leading cause of hospitalization in premature infants born at 29-34 weeks gestational age. Severe RSV disease early in life can lead to chronic respiratory conditions like asthma, wheezing and bronchitis. Current standard of care is limited to symptomatic relief with no vaccine or antiviral drugs; therefore a safe and efficacious RSV therapy is needed. The RSV Matrix (M) protein is a major structural protein with key roles in virus assembly and budding. We have shown previously that M protein localises to the nucleus early in infection, but later is localised mostly in the cytoplasm. Nuclear export of RSV M protein, mediated by the nuclear export protein XPO1, is crucial to initiate and complete viral assembly and budding. Inhibition of RSV M export correlates with reduced virus titres. The therapeutic targeting of host proteins hijacked by RSV to facilitate replication holds promise as an effective antiviral strategy. Karyopharm Pharmaceuticals, USA have developed a series of synthetically designed compounds that specifically interrupt XPO1-mediated nuclear export. These compounds are effective against a wide range of haematological and solid malignancies which utilize the over expression of XPO1 for tumorigenesis. We aim to re-purpose these anticancer drugs as antivirals targeting RSV. In this study, we show that inhibition of XPO1 by the SINE compounds KPT-185 and -335 in cell culture significantly reduces RSV infectious titres. This was observed across different cell lines at concentrations below the cytotoxic dose. SINEs hold promise as a novel approach for targeting RSV.

37. Identification of the Target Regions Responsible for Resistance to Compound A, a Novel Antiviral Agent Against Dengue Virus

Haruaki Nobori, MS[1], Shinsuke Toba, MS[1], Ryu Yoshida, PhD[1], Yasuko Orba, PhD[2], Hirofumi Sawa, MD, PhD[2], Akihiko Sato, DVM, PhD[1]

[1]Shionogi & Co., Ltd. Drug Discovery & Disease Research Laboratory, Sapporo, Japan; [2]Hokkaido University Research Center for Zoonosis Control, Sapporo, Japan

BACKGROUND: Dengue virus (DENV) is the causative agent of dengue fever and dengue hemorrhagic fever. There is no antiviral drug for DENV infections. We have found anti-DENV agent, Compound A, which has a benzimidazole skeleton, by screening of cell-based assay. To identify the target regions of Compound A, we attempted to obtain Compound A-resistant DENV type 2 (DENV2) and analyzed their phenotype.

METHODS: To isolate Compound A-resistant virus, DENV2 was cultured in BHK-21 cells in the presence of Compound A (1.8-3.6 μg/ml). Whole genome of Compound A-resistant DENV2 was determined by using Next-Generation Sequencer. DENV2 infectious clones with single amino acid mutations based on the results of whole genome sequencing were constructed using mutational PCR. The growth of DENV2 mutants were measured by qRT-PCR.

RESULTS: Compound A-resistant DENV2 were isolated after passage for 20 days in the presence of Compound A. Whole genome sequencing of Compound A-resistant DENV2 revealed that four amino acid mutations in prM, NS4A, and NS4B regions. Among these mutations, NS4A seems to be important for sensitivity against Compound A.

CONCLUSIONS: We identified target region of anti-DENV2 Compound A by isolation of escape mutant for Compound A and infectious clones of DENV2. NS4A of DENV seems to be a good candidate for development of anti-DENV agents.

38. A Novel Kinase Inhibitor is a Pan-Influenza Antiviral with a High Barrier to Resistance

Ryan O'Hanlon, MS, Megan Shaw, PhD

Icahn School of Medicine at Mount Sinai, New York, New York, United States of America

Antiviral drugs are a necessary part of our armory when it comes to treating and preventing viral infections. This is especially true when vaccines do not exist nor afford adequate protection in some populations. The majority of approved antivirals target viral proteins. This makes them highly specific for a particular virus and susceptible to the development of resistance. An alternative approach is to target a host factor that is essential for virus replication, which should ensure a high barrier to virus resistance. This project focuses on discovering and characterizing novel antivirals that target these host factors.

Initially, 744 primary hits were selected from an influenza antiviral screen of a 900,000 compound library. One of these, M85, was identified as a high priority entry inhibitor of influenza A and B viruses with minimal toxicity. Preliminary studies suggest that M85 targets EGFR and PI3 kinases, which are known host factors required for entry of influenza virus and other viruses. Other preliminary experiments demonstrate the inability to select for M85 resistance mutations. Considering these characteristics, compounds like M85 have the potential to be a novel class of pan-influenza virus inhibitors with a high barrier to resistance.

Studying the mechanism of action of M85 may elucidate other potential host factor targets. In addition, lead optimized analogs of M85 will be assessed for potential synergistic activity with oseltamivir, an FDA-approved influenza drug. A synergistic drug combination will likely be less susceptible to the development of resistance observed in influenza strains with oseltamivir.

39. Systematic *In Vitro* Evaluation of Current and Experimental Hepatitis B Therapeutics: Potential Utility for Combinations Exploiting Multiple and Diverse Mechanisms of Action

Andrea Cuconati, PhD[1], Andrzej Ardzinski, BS[1], Lauren Bailey, PhD[1], Nagraj Mani, PhD[1], Kim Stever, BS[1], Xiaohe Wang, MD[1], Amy Lee, MS[2], Chris Moore, PhD[1], Rene Rijnbrand, PhD[1], Michael Sofia, PhD[1]

[1]Arbutus Biopharma, Inc., Doylestown, Pennsylvania, United States of America; [2]Arbutus Biopharma Corp., Burnaby, British Columbia, Canada

Current treatment regimens for chronic hepatitis B are limited to nucleoside/nucleotide analogue inhibitors ("nucs"), and PEGylated interferon alfa-2a. Despite clear clinical benefits, the potential for a durable off-drug response in patients is still extremely low, and they are not considered curative. It is widely accepted that the next advance in the treatment of hepatitis B virus (HBV) infection will involve combination therapies targeting multiple mechanisms of action (MOAs) as well as modulating the innate and adaptive arms of the immune response, resulting in greater inhibition of viral replication and cccDNA regeneration. To evaluate the possibility of mechanistic antagonism and demonstrate the utility of targeting multiple MOAs, multi-dose two-way combinations were studied in HBV cell culture systems that include stably-transfected HBV-replicating cell lines, and HBV-infected primary human hepatocytes. The endpoints examined were secreted antigens, and secreted or intracellular viral DNA. We focused on classes of small molecule encapsidation inhibitors and Arbutus' lipid nanoparticle-formulated siRNA agents, in combination with PEGylated interferon and the nucs entecavir, TDF and TAF. All combinations were found to be additive to synergistic in the reduction of viral DNA and antigen production; interestingly, almost all combinations of Arbutus' novel therapeutics with interferon exhibited synergy for all three endpoints, suggesting that innate immune modulation could be a major factor in the clinical benefits of combination treatments. The lack of obvious mechanistic antagonism observed in these studies underscores the potential utility of Arbutus' novel candidates in combination regimens with currently approved HBV therapies.

40. 2'-Fluoro-2'-Deoxycytidine Inhibits Arenavirus and Bunyavirus Replication

Donald Smee, PhD, Kie-Hoon Jung, PhD, Jonna Westover, PhD, Brian Gowen, PhD

Institute for Antiviral Research, Utah State University, Logan, Utah, United States of America

2'-Fluoro-2'-deoxycytidine (FdC) has been reported to inhibit many unrelated viruses in vitro, including Borna disease, hepatitis C, Lassa fever, influenza and various herpes viruses. Also, FdC prevented mortality in mice infected with influenza A viruses. FdC is not toxic to uninfected cells at >100 µM. Thus, we investigated the antiviral activity of FdC against arenaviruses and bunyaviruses in 50% cytopathic effect inhibition (CPE) and 90% (1 $\log_{10}$) virus yield reduction (VYR) assays in Vero or Vero 76 cells. VYR assays were employed for viruses not exhibiting robust CPE. FdC inhibited Junin, Pichinde, and Tacaribe arenaviruses in CPE assays at 0.6, 5.1, and 4.7 µM, respectively. Lymphocytic choriomeningitis arenavirus was inhibited at 1.9 µM in VYR assays. La Crosse, Maporal, Punta Toro, Rift Valley fever (RVFV), and San Angelo bunyaviruses were inhibited in CPE assays at 2.2-9.7 µM concentrations. In VYR assays, Heartland and severe fever with thrombocytopenia syndrome bunyaviruses were inhibited at 0.9 and 3.7 µM, respectively. In contrast, ribavirin was inhibitory at an average of 47 µM against all of the viruses. FdC was also evaluated in combination with ribavirin against RVFV in vitro, with strong synergy apparent when each compound was used in its active range, as evaluated by 3-dimensional MacSynergy method. Results from an experiment investigating the antiviral activity of FdC in a lethal mouse model of RVFV infection will be discussed.
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41. Synthesis and In-Vitro Biological Activity of Novel Imine Derivatives

Gholamreza Zarini, PhD
Faculty of Natural Science, University of Tabriz, Tabriz, Iran

A diversity of biological activities and pharmaceutical uses have been attributed to imine derivatives such as antibacterial, antifungal (1,2).

A series of imine derivatives were synthesized and their structure confirmed by FT-IR, ^{1}HNMR, ^{13}CNMR, elemental analysis.

The synthesized compounds were evaluated for their antimicrobial activity against bacterial strains *Staphylococcus aureus* ATCC25923,*Bacillus subtilis* ATCC 6633 *Escherichia coli* ATCC 25922, *Klebsiella sp.* ATCC 700834 and the yeast *Candida kefyr*. The MICs (minimum inhibitory concentration) values of the compounds were determined by two-fold microdilution method. Microbilogical results showed that the compound 3 possed abroad spectrum of antimicrobial activity against Gram-positive and Gram-negative bacteria, and also against the yeast *Candida kefyr* with MIC value lower than 62.5µg/ml. The other compounds were indicated poor antimicrobial potency against test strains.

REFERENCE:

1) Josi S.P., Vagdevi H.M, Vaidya V.P, (2008) European Journal of medicinal Chemistry, 43, 1989-1996.

2) Eun Soo park, Woong sig moon, Gin san yoon (2001) Internatonal Biodeterioration and biodegradation 47, 209-214.

42. Anti-Influenza Activity of the N-Benzylaminomethansulphonic Acid

Ruslan Khoma, PhD[1], Alim Ennan, MD, MPH[2], **Tetyana Grydina, PhD**[3], Karina Radkevich, MS[3], Alla Fedchuk, PhD[4], Viktor Lozitsky, PhD[4]
[1]Odessa I.I. Mechnikov National University, Odesa, Ukraine; [2]Physico-Chemical Institute of Environment and Human' Protection, Odesa, Ukraine; [3]Odesa National Medical University, Odesa, Ukraine; [4]Odessa Research Center for Biological Testing Products and Preparations, Odesa, Ukraine

The influenza virus causes the greatest number of acute respiratory viral infections, which can lead to an exacerbation of chronic systemic diseases, to emergence of bacterial complications, to a significant deterioration of public health. But the majority of human isolates of influenza viruses rapidly become resistant to remantadine, oseltamivir. So, the creation of new effective anti-influenza agents is urgent task of medical science.

The purpose of this study is to research antiviral activity derivative of the N-benzylaminomethanesulphonic acid (BnAMSA) compared reference drug (Tamiflu).

Methods of the compound activity studies in vitro on the tissue culture of chorio-allantoic covers of 10-12-days chicken embryos (CAC) were used. We have studied the influence of BnAMSA to extracellular virus A/PR/8/34 (H1N1) and on the fabric's ability to maintain its reproduction after BnAMSA treatment. We also studied the effect of the substance on the reproduction of the viruses A/PR/8/34 (H1N1) and A/Hong Kong/1/68 (H3N2) in the cell culture CAC.

BnAMSA had not neither efficacy against extracellular virus A/PR/8/34/ nor influence on the fabric's ability to maintain its reproduction. BnAMSA inhibited reproduction of A/Hong Kong/1/68 on 4,08 log10 TID50 and A/PR/8/34 on 1,67 log10 TID50 as compared to control. Tamiflu demonstrated 4,07 and 4,07 log10 TID50 respectively.

So, BnAMSA demostrated antiviral activity against influenza virus A/Hong Kong/1/68 on the level of Tamiflu. Level of inhibition reproduction of influenza virus A/PR/8/34 of BnAMSA was lower than level Tamiflu. Results of this study show that N-benzylaminomethansulphonic acid is promising compounds for searching and design of effective antivirals.

43. CD4 Signaling Induced Cytosolic Localization of Topoisomerase II Isoforms

Sunnam Balakrishna, PhD, Bommakanti Akhila, PhD, and **Anand Kondapi, PhD**
University of Hyderabad, Hyderabad, Telangana, India

Topoisomerase II (Topo II) has diverged cellular functions and characters. Along with the DNA topological maintenance, Topo II has involvement in the replication, transcription, recombination etc. Topo II isoforms are potential targets in cancer, viral infection and ageing. It is unknown about their involvement or response to the external stimulus or signaling. Topo II isoforms Co-localization with HIV-1 Reverse transcriptase in the cytosol of HIV-1 virus infected cells was reported in our previous publication. This communication presents results of the signaling mechanisms associated with the translocation of Topo II isoforms from nucleus to cytosol. Results show that immunoglobulin-activated cellular signaling/stimulus of the CD4 receptor can effectively recruit Topo IIβ in the cytosol than CXCR4 . And it is interesting to know that, only CXCR4 driven signal/stimulus could localize Topo IIβ to cytosol transiently. Neither IgG nor CCR5 has impact on the localization of Topo II isoforms in T cells. Based on these results we propose that both CD4 and CXCR4 signaling plays crucial role in recruitment of Topo II isoforms in the cytosol for the successful reverse transcription and preintegration complexes formation of HIV-1 genome.

44. Design, Synthesis and Characterization of Biogenic Chloroquine Silver Nanoparticles as Potential Anti-HBV and Anticancer Agent

M Chandramohan, MD, PhD[1], P Selvam, PhD[2], D. Sivakumar, MD[1], S.C. Vivekananthan, MD[1], Elanchezhiyan Manickan, MD, PhD[3]
[1]*Bharat Ratna Kamarajar Liver Hospital and Research Centre, Madurai., Madurai. Tamilnadu, India;*
[2]*Sir CV Raman-Krishna International Research Centre, Kalasalingam University, Krishnankoil, Tamildau, India;*
[3]*Dept of Microbiology, University of Madras-Taramani Campus, Chennai, Tamilnadu, India*

OBJECTIVE: Nanotechnology has induced a paradigm shift in biomedical sciences, especially in the field of antiviral and cancer chemotherapy. Chloroquine (CQ) is a versatile bioactive agent and already reported board spectrum of activity including antiviral activity. For novel approach therapy, we synthesized of chloroquine silver nanoparticles(CQ AgNPs), CQ and CQ AgNPs tested for *invitro* anticancer activity in human liver cancer cells and antiviral activity against Hepatitis B virus

METHOD: In order to explore the antiviral potential of chloroquine, nanochloroquine was prepared using silver nitrate. Prepared nanoparticles encapsulated with drug carriers namely polyvinyl alcohol (PVA), polyethylene glycol (PEG) and then encapsulated nanoparticles were characterized using UV-VIS, FT-IR, SEM, EDX and PXRD. CQ and Chloroquine nanoparticles also assayed for *invitro* anticancer activity against human liver cancer cells and antiviral activity against HBV virus by HBsAg binding inhibition assay .

RESULT: The UV-VIS spectrum for prepared silver nanoparticles shows absorptions peak at 450 nm confirms the presence of silver nanoparticles. SEM analysis of prepared nanoparticles shows the spherical shape and average size of the particle is 18-25 nm. Powder XRD studies, 2Θ value 111 and 211 confirms that the nanoparticles are crystalline in nature. CQ and Synthesized chloroquine nanoparticles had significant cytotoxicity in human liver cancer cells and chloroquine exhibits anti-HBV activity by effectively interfere viral binding to its receptors

CONCLUSION: Biosynthesized CQ AGNPs exhibits anticancer potential in human liver cancer cells and Chloroquine demonstrated for anti-HBV activity by HBsAg-Receptor binding inhibition assay, suitable further investigation to explore molecular mechanism to be studied

45. Targeted Oral Delivery of HIV-1 Drug Combination (3TC + TNF + ATV/r) Through Lactoferrin Nanoparticles

Prashant Kumar, PhD, Yeruva Lakshmi, MS, **Anand Kondapi, PhD**
University of Hyderabad, Hyderabad, Telangana, India

Lactoferrin nanoparticles loaded Second Line combination of Highly active antiretroviral drugs (SLHAART-NP) were prepared using sol-oil phase separation method 4:1 cocnttations of protein and drug combination of 3TC, TNF, ATV and RTV. SLHAART-NP are found to be 65-72nm with hydrodynamic radii of 113nm and surface charge of -27mV. The EE% was calculated to be 73, 62 and 68% for 3TC, TNF and ATV respectively. Drugs are released in the endosomal pH, while negligible amount of drugs are released at physiological pH. The individual components of drug regimen have been tested for the anti-HIV (HIV-$1_{93IN101}$) activity and IC_{50} for soluble drugs have found to be sol TNF (28.31nM) & Nano-TNF (12.14nM), sol ATV (6.43nM) & Nano-ATV (4.34nM) and sol RTV (19.71nm) and Nano-RTV (7.23nM). Furthermore, the efficacy of SLHAART-NP was two-fold higher than corresponding soluble combination. The *in-vivo* PK studies have been done in rats and found that, the overall bioavailability of all drugs have been increased with significantly increase in AUC (≈3-5fold). AUMC increased 8fold (3TC), 2.6fold (TNF) and 2.0 fold (ATV). The Drug half-life has been increased by >2 fold for each drugs. The safety analysis data shows that LDH, Urea, Creatinine, AST level were found to be reduced when animals were treated with nanoformulation. Finally the Histopathological analysis revealed the absence of tissue damages in nanoformulation treated rats. In summary, the second line Nano formulated drugs are more efficacious, pharmacologically active, less toxic, and more potent against HIV.

46. Synthesis of Small Molecules for Treating Ebola Virus Infection

Elzbieta Niemiec-Plebanek, PhD[1], Vincent Roy, PhD[1], Maximes Bessieres, PhD[1], Dawid Warszycki, PhD[2], Andrzej Bojarski, PhD[2], Gilles Lalmanach, PhD[3], **Luigi Agrofoglio, PhD**[1]
[1]ICOA UMR CNRS 7311 – Universite Orleans, Orleans, France; [2]Institute of Pharmacology – Polish Academy of Sciences, Krakow, Poland; [3]INSERM, UMR 1100 – UNIVERSITE TOURS, Tours, France

Ebola virus is an RNA virus, belonging to the *Filoviridae* family. Infections by the Ebola and Marburg filoviruses cause a rapidly fatal hemorrhagic fever in humans for which no approved antivirals are available. The EBOV glycoprotein (GP) plays critical roles in the early stage of virus infection, including receptor binding and membrane fusion, making it a potential target for the development of anti-EBOV drugs. EBOV GPs require processing by host cell-derived cysteine cathepsins for productive infection. Then proteolytically processed virus GP binds to Niemann-Pick C1 protein (NPC1) within the late endosomal/lysosomal compartments. Thus, cysteine cathepsins and NPC1 are targets for antiviral therapy.

We will describe the synthesis based on the modern tools of organic chemistry and report on the biological evaluation of :

(1) A series of 1,3,5-triazine derivatives targeting the inhibition of lysosomal cysteine cathepsins. Docking was performed on the most active compounds to define and elucidate their binding mode.

(2) A series of small molecules inhibitors of NPC1. In silicoassays were done to characterize their ADMET profile.

47. Cirsimarit in Inhibits Influenza A Virus Replication by Downregulating NF-κB Signal Transduction Pathway

Haiyan Yan, MS, Huiqiang Wang, MS, Jinqiu Yin, BS, Shuo Wu, PhD, Danqing Song, PhD, Yuhuan Li, PhD
1 Tian Tan Xi Li,Beijing 100050,China

On account of high mutation rate, influenza virus exhibits drug-resistance to the currently available drugs which is a major public concern. Therefore, there remains a need to develop novel anti-influenza drugs. Naturally medicinal plants-derived products have shown great potential in preventing viral infection diseases. This study aims at investigating the antiviral efficacy *of cirsimaritin, a* flavonoids compound isolated from *Artemisia scoparia*, and studying the underlying mechanism of anti-IAV*in vitro*. The study showed that cirsimaritin dose-dependently reduced IAV RNA and protein synthesis, which was, at least in part, the result of inhibition of NF-κB/p65 protein expression and NF-κB/p65 phosphorylation in nuclear. Furthermore, we found that cirsimaritin suppressed the activation of JNK MAPK and P38 MAPK but not ERK MAPK *in vitro*. The expressions of pro inflammatory cytokines (TNF-α, IL-1β, IL-6 and IL-10) and inflammation-related protein COX-2 were downregulated by cirsimaritin. Furthermore, haemagglutination inhibition (HI) and neuraminidase (NA) assays suggested that cirsimaritin had no inhibitory effect on the entry and release steps of the viral lifecycle. These results demonstrated that cirsimaritin inhibited IAV replication by downregulating NF-κB signal transduction pathway. In a conclusion, cirsimaritin may be a potential agent or supplement against IAV infection.Further *in vivo* efficacy and pharmacological studies will identify the potential preclinical candidates for development of the flavonoids as therapeutics of influenza infections in future.

48. Antiviral Activity of New Fluorinated Thioacyl Derivatives of Amino Acids

Liubov Biliavska, PhD[1], Yulia Pankivska, MS[1], Olga Povnitsa, PhD[1], Svitlana Zagorodnya, PhD[1], Nadya Pikun, PhD[2], Yuriy Shermolovich, MD, MPH[2]
[1]D.K. Zabolotny Institute of Microbiology and Virology of the NASU, Kiev, Ukraine; [2]Institute of Organic Chemistry NAS of Ukraine, Kyiv, Ukraine

INTRODUCTION: The diseases caused by Herpes Simplex Virus 1 are widely spread. The shortage of the antiviral compounds due to their high toxicity and emergence of resistant viruses is a major problem in the treatment of patients. This work is related to the determination of the antiviral activity of new fluorinated heterocyclic molecules against HSV-1.

METHODS: Nnew fluorinated *N*-alkylthioamides (8 compounds) were synthesized in Institute of Organic Chemistry NAS of Ukraine [Pikun N.V., 2016]. The effect on infectivity of virus was determined by MTT-assay. Were studied: adsorption, penetration, virucidal effect and reduction the titers of virus infectivity in the presence of substances.

RESULTS: Compounds 10S-20, 10S-23 and 10S-24 showed CC_{50} values of ≥1000 μg/ml. Other compounds had higher cytotoxicity. It was shown that 10S-23, 10S-24, 10S-28 and 10S-29 suppressed HSV-1/US reproduction by 50% at concentrations of 71μg/ml, 35μg/ml, 87μg/ml and 16μg/ml, respectively. Compounds 10S-19, 10S-20, 10S-21 and 10S-22 had lower efficiency. The 10S-23 and 10S-24 compounds showed anti-HSV-1 activity, their selectivity indexes (SI) were 14 and 29, respectively. These compounds prevented the adsorption and penetration of HSV-1 into cells up to 30%. It was found that at the concentration of 150 – 33μg/ml compounds reduce the titer of virus obtained *de novo* by 93-98% and 99%, respectively. The absence of virucidal activity was shown for all compounds.

CONCLUSIONS: At least two compounds can be used as starting points for the development of effective anti-HSV drugs in the future.

REFERENCES: Pikun N.V. (2016) Journal of Fluorine Chemistry 185, 86–90.

49. Inhibition of Hepatitis B Virus Replication by N-hydroxyisoquinolinediones and Related Polyoxygenated Heterocycles

Tiffany Edwardstc, MS[1], Elena Lomonosova, PhD[1], Jenny Patel, MS[1], Qilan Li, PhD[1], Fabrice Bailly, PhD[2], Philippe Cotelle, PhD[2], Erofili Giannakopoulou, MS[3], Grigoris Zoidis, PhD[3], Kelly Long, MS[4], John Sagartz, PhD[4], John Tavis, PhD[1]

[1]Saint Louis University, School of Medicine, St Louis, Missouri, United States of America; [2]University of Lille, Lille, France; [3]University of Athens, Athens, Greece; [4]Seventh Wave Laboratories, St. Louis, Missouri

We previously reported inhibition of Hepatitis B virus (HBV) replication by an N-hydroxyisoquinolinedione (HID) compound via suppression of the viral ribonuclease H (RNaseH). Subsequently we found the biochemical RNaseH assay underestimated the number of HBV replication inhibitors. Here, we screened 44 HIDs and structurally related polyoxygenated heterocycles for viral replication inhibition.

Inhibiting the HBV RNaseH blocks synthesis of the (+) polarity DNA strand and causes accumulation of RNA:DNA heteroduplexes. The compounds were screened for preferrential suppression of (+) polarity DNA in hepatoma cells. 12 compounds selectivley inhibited HBV (+) polarity DNA strand. EC_{50}s ranged from 0.69 – 19 µM. CC_{50}s by three assays ranged from >100 -11 µM. Accumulation RNA:DNA heteroduplexes in HBV capsids was assessed by Southern blotting for a representative HID. It caused accumulation of RNA:DNA heteroduplexes, indicating inhibition of the HBV RNaseH in cells. An endogenous polymerase reaction was used to determine the effects of the inhibitors on HBV reverse transcriptase activity in isolated HBV capsids. None of the active compounds affected the polymerase's elongation of DNAs within capsids. *In vivo* efficacy for an N-hydroxypyridinedione (HPD) compound was evaluated in HBV-infected FRG chimeric mice with humanized livers. The compound suppressed viremia by 1.3 log after two weeks of therapy ($p = 0.00003$).

Therefore, the HID scaffold is more promising for anti-HBV drug discovery than we originally reported, and the HPD scaffold appears to hold potential for antiviral development. The negative results with the other compounds lend insight to structure activity relationships.

50. Study the Influence of Process and Formulation Parameters on Solubility and Dissolution Enhancement of Efavirenz Solid Solutions Prepared by Hot Melt Extrusion Using Box Behnken Factorial Design

Dilipkumar Suryawanshi, PhD

Institute of Chemical Technology, Mumbai, Mumbai, Maharashtra, India

BACKGROUND: Efavirenz EFV is a non-nucleoside reverse transcriptase inhibitor and categorized in to BCS class II drug.

METHOD: In this context, we have investigated the dissolution performance of amorphous solid solutions (SSs) of efavirenz (EFV) in polymeric matrix systems with drug loading of 30% 50% and 70 % of EFV with using Box-Behnken design approach processed by hot melt extrusion. The polymers were selected based on the Hansen solubility parameter calculation. The formulated solid dispersions were further termed as ASS and BSS for Soluplus® and Kollidon® VA64 respectively.

RESULTS: In DoE experiments, Box Behnken factorial design was conducted to evaluate effect of three independent variables viz, Soluplus® ratio (A_1), HME screw rpm (A_2), and processing temperature (A_3) similarly for Kollidon®VA64 ratio (B_1), screw rpm (B_2),and processing temperature (B_3) on responses such as solubility (X_1 and Y_1), Dissolution rate (X_2 and Y_2) for both ASS and BSS systems respectively. Developed SSs were characterized by X-ray powder diffraction, differential scanning calorimetry, Fourier transform infrared, Atomic force microscopy, 2D COSY NMR and FTIR chemical imaging to determine the solid state of EFV. DSC and XRD data confirmed that bulk crystalline EFV transformed to the amorphous form during the HME processing. FTIR chemical imaging and AFM analysis results inferred that EFV was dispersed homogenously in respective polymeric systems.

CONCLUSION: EFV oral tablet dosage form has been developed successfully using Soluplus® and Kollidon® VA64 as carrier by industry feasible and scalable hot melt extrusion technology. The formulation was found stable for a period of six months.

51. Antiviral Potential of Fluorinated Analogs of Uracil in EBV-Associated Cell System

Svitlana Zagorodnya, PhD[1], Krystyna Naumenko, MS[1], Anna Golovan, PhD[1], Galina Baranova, MS[1], Ganna Gudz, PhD[2], Yriy Shermolovich, PhD[2]
[1]Zabolotny Institute of Microbiology and Virology of the NAS of Ukraine, Kyiv, Ukraine; [2]Institute of Organic Chemistry NAS of Ukraine, Kyiv, Ukraine

The combination of prediction of biological activity with *in vitro* studies is an optimal way for creation of potential drugs. With this regard, the fluoride analogues of nucleosides are of a special interest. The aim of the study was to evaluate an antiviral activity of fluorinated analogs of uracil using *in vitro* and *in silico* approaches.

Three analogs (G26, G27, G28) on the base of 5-(p-tolilsulfonil)-6(polifluoroalkil)uracil were used in the study. Research was carried out by using MTT-assay, PCR and flow cytometry, PASS and idTarget software.

According to PASS prediction all compounds may possess the antiviral activity; the potential activity values were in range 0,649-0,425. Two compounds (G26, G27) may possess an anticancer activity. The *in vitro* study let to reveal the low level of cytotoxicity of these uracil derivatives. Anti-EBV activity was observed for all compounds and EC_{50} values were 75, 65 and 80μg/ml. Several possible targets for compound G27 were identified with a help of molecule docking. It was established, that majority of the targets are enzymes that participate in the synthesis of nucleic acids and apoptotic proteins. Based on computer modeling, the 95-8 and Raji cells treated with G27 were analyzed with a help of flow cytometry. It was marked an induction of apoptosis in the presence of G27, at 500μg/ml of which, the portion of apoptotic cells reached almost 90%.

The research of fluorine-containing uracil derivatives had shown antiviral ability and apoptosis inducers at B-lymphomas, which opens new directions in the study of this class compounds.

52. 6'-Fluoro-3-Deazaneplanocin: Synthesis and Antiviral Properties

Chong Liu, PhD[1], Qi Chen, PhD[2], Steven Cardinale, PhD[3], Terry Bowlin, PhD[3], Stewart Schneller, PhD[1]
[1]Auburn University, Auburn, Alabama, United States of America; [2]Slippery Rock University of Pennsylvania, Slippery Rock, Pennsylvania; [3]Microbiotix, Inc., Worcester, Massachusetts

Neplanocin A (1) is a representative carbocyclic nucleoside known for its antiviral promise by inhibiting S-adenosylhomocysteine hydrolase (SAH). Its 6'-fluoro derivative (2) has been studied as a more effective, irreversible SAH inhibitor. Our recent extended investigations focusing on 3-deazaneplanocin and 3-deaza-1',6' *iso*neplanocin derivatives suggested that 6'-fluoro-3-deazaneplanocin (3) would be a worthy target. In that direction, a convenient stereoselective synthesis of 3 has been accomplished from D-ribose and will be reported along with its favorable antiviral activity towards Ebola, measles and dengue and its potent inhibitory effect on SAH.

53. Development of Small and Specific Herpes Virus Inhibitors Based on Abalone Hemocyanin Proteins

Negar Talaei Zanjani, PhD[1], Monica Miranda-Saksena, PhD[2], Peter Valtchev, PhD[1], Russell J. Diefenbach J. Diefenbach, PhD[3], Jessica Yichen Zhong, PhD[4], Eve Diefenbach Diefenbach, PhD[2], Vincent G. Gomes, PhD[1], Joel P. Mackay, PhD[4], Anthony L. Cunningham, MD, PhD[2], Fariba Dehghani, PhD[1]
[1]School of Chemical and Biomolecular Engineering, The University of Sydney, Australia; [2]Centre for Virus Research, Westmead Millennium Institute for Medical Research, Sydney, Australia; [3]Department of Biomedical Sciences, Macquarie University, Australia; [4]School of Life and Environmental Sciences, The University of Sydney, Australia

Infections caused by herpes simplex viruses (HSV) are prevalent worldwide and ranges from mild illnesses such as oral or genital lesions to more severe conditions such as encephalitis. Common treatments against HSV, share the same mechanism of action, which involves inhibition of viral replication by interfering with

DNA elongation. In order to increase the efficacy of current treatments, it is necessary to develop antiviral agents affecting other viral replication events, particularly the early steps such as viral attachment and entry. We have recently discovered that a marine protein, known as hemocyanin, inhibits HSV type 1 infections by interacting with viral surface glycoproteins that are involved in viral binding and entry. However, due to the complex structure and large size (8 MDa) of hemocyanin, it is essential to find a smaller active domain on this protein. To this aim, we have broken down the protein into its 16 structural building blocks, known as functional units (FU) with a molecular mass of 50 kDa, using limited enzymatic digestion. The conditions of digestion were optimized to obtain high yields of FU while maintaining the antiviral activity. Various analytical techniques including one- and two-dimensional gel electrophoresis and mass spectrometry demonstrated that 14 out of 16 FUs were obtained. The functional units were then purified by size exclusion chromatography and assayed for cell viability and antiviral activity against HSV-1 on Vero cells. The results demonstrated that functional units are active against HSV-1, although the activity is 2-3 fold less than native hemocyanin.

54. Antiviral Activity of Oroxylin A against Coxsackievirus B3 Alleviates Virus-Induced Acute Pancreatic Damage in Mice

Bo-Eun Kwon, MS[1], Hyuk-Hwan Song, PhD[2], Eun-Hye Hong, PhD[1], Hyun-Jeong Ko, PhD[1]
[1]College of Pharmacy, Kangwon National University; [2]Agency for Korea National Food Cluster

The flavonoids mosloflavone, oroxylin A, and norwogonin, which were purified from *Scutellaria baicalensis* Georgi, significantly protected Vero cells against Coxsackievirus B3 (CVB3)-induced cell death. To investigate the in vivo antiviral activity of oroxylin A, we intraperitoneally inoculated CVB3 into 4-week-old BALB/c mice. Body weights and blood glucose levels of the mice were decreased after CVB3 infection, and these changes were attenuated by the administration of oroxylin A. Importantly, treatment of mice with oroxylin A reduced viral titers in the pancreas and decreased the serum levels of the inflammatory cytokines including interleukin-6 (IL-6) and tumor necrosis factor (TNF)-α. Additionally, the administration of oroxylin A mitigated the histological pancreatic lesions and apoptotic cell death induced by CVB3 infection and increased the levels of phospho-eIF2a in infected pancreata. The results suggest that oroxylin A may represent a potent antiviral agent against CVB3 infection.

55. Nucleoside Inhibitors of Tick-borne Encephalitis Virus: Structure-Activity Relationships and Viral Resistance Study

Luděk Eyer, PhD[1], Radim Nencka, PhD[2], Daniel Růžek, PhD[1]
[1]Veterinary Research Institute, Department of Virology, Brno, Czech Republic; [2]Institute of Organic Chemistry and Biochemistry, The Czech Academy of Sciences, Prague, Czech Republic

Tick-borne encephalitis virus (TBEV) represents one of the most serious arboviral neuro-infection in Europe and northern Asia, against which no specific antiviral therapy is available at present. A structure-activity relationship study of 29 nucleoside analogues revealed that the methylation at the C2′ position or azido modification at the C4′ position exerted a strong TBEV inhibition activity (EC_{50} 0.3 – 11.1 μM) and low cytotoxicity *in vitro*. However, substitutions of the O2′ and O3′ positions and the heterobase moiety resulted in a complete loss of anti-TBEV activity (EC_{50} > 50 μM) or increase of cytotoxicity *in vitro*. A low micromolar TBEV inhibition *in vitro* was demonstrated for the imino-C-nucleoside BCX4430 (EC_{50} of 1.5 μM). Antiviral activity of 7-deaza-2′-C-methyladenosine was evaluated *in vivo* using a lethal rodent model of TBEV infection; intraperitoneal application of 25 mg/kg twice a day resulted in survival rate of 60%. We identified a signature mutation S603T within the active site of the viral NS5 RNA-dependent RNA-polymerase which conferred a high-level resistance of TBEV to the family of 2′-C-methylated nucleosides. This mutation led to a resistance-associated loss of viral fitness in cellular culture and in decreased virulence potency (attenuation) *in vivo*. Two amino acid changes (Y453H and R424G) located outside of TBEV NS5 active site were found to determine the acquired resistance to 4′-C-azidocytidine. High antiviral activity and low cytotoxicity of C2′ methylated or C4′ azido substituted pharmacophores suggest that such compounds represent promising candidates for further development of potential therapeutic agents against TBEV infection.

56. Formulation of Antiretroviral Drugs into Single and Dual Component Solid Drug Nanoparticles for Improved Oral Bioavailability

Alison Savage, PhD[1], Samantha Chadwick, PhD[1], Darren Moss, PhD[2], Helen Box, PhD[2], Joanne Sharp, PhD[2], Andrew Owen, PhD[2], Steve Rannard, PhD[1]

[1]Department of Chemistry, University of Liverpool, Liverpool, United Kingdom; [2]Department of Molecular and Clinical Pharmacology, University of Liverpool, Liverpool, United Kingdom

Antiretroviral drugs are often taken in combinations as part of a HIV drug regimen which act on multiple viral targets. This is known as highly active antiretroviral therapy (HAART) and often involves antiretroviral drugs being administered with protease inhibitors known to boost the efficacy of the antiretroviral. For oral administration, we have prepared solid drug nanoparticles (SDN) with different antiretrovirals to prepare both single component SDNs and dual component SDNs to combine two APIs into one powder. This emulsion-templated freeze drying method that screens excipients to produce solid drug nanoparticles has previously reported for the formulation of the antiretroviral drugs Efavirenz and Lopinavir, the resulting products of which are currently under investigation in human trials.[1,2] The combination SDNs have the potential for a two part benefit; with the SDNs improving the oral bioavailability of the antiretroviral drug thus reducing the dosage of antiretroviral administered for the same therapeutic benefit, whilst the combination therapy has the potential to reduce pill burden often experienced by people with HIV by combining two APIs into one medicine.

1. Giardiello, M.; Liptrott, N. J.; McDonald, T. O.; Moss, D.; Siccardi, M.; Martin, P.; Smith, D.; Gurjar, R.; Rannard, S. P.; Owen, A.,. *Nature Communications* 2016, *7*.

2. McDonald, T. O.; Giardiello, M.; Martin, P.; Siccardi, M.; Liptrott, N. J.; Smith, D.; Roberts, P.; Curley, P.; Schipani, A.; Khoo, S. H.; Long, J.; Foster, A. J.; Rannard, S. P.; Owen, A., *Advanced Healthcare Materials* 2014, *3* (3), 400-411.

57. Activity of SAMHD1 in Cycling Cells Permissive to HIV-1 Infection

Maria Pujantell, MS[1], Roger Badia, PhD[1], Javier Torres-Torronteras, PhD[2], Luis Menéndez-Arias, PhD[3], Ramón Martí, PhD[2], Albert Ruzo, PhD[4], Eduardo Pauls, PhD[1], Bonaventura Clotet, MD, PhD[1], Ester Ballana, PhD[1], José Esté, PhD[1], Eva Riveira-Muñoz, PhD[1]

[1]AIDS Research Institute – IrsiCaixa; [2]Vall d'Hebron Institut de Recerca; [3]Centro de Biología Molecular "Severo Ochoa"; [4]The Rockefeller University, New York

SAMHD1 is a triphosphohydrolase that restricts HIV-1 by limiting the intracellular dNTP pool required for reverse transcription. Although SAMHD1 expression and activation is variable in distinct human cell lines and tissue types, its restriction activity is thought to be relevant only in non-cycling cells, where availability of dNTPs is limited. Here, we show that SAMHD1-induced degradation by HIV-2 Vpx affects the the dNTP pool and HIV-1 replication capacity in the presence of the 3'-azido-3'-deoxythymidine (AZT) in cycling cells, without affecting susceptibility to HIV-1 infection. Then, a knock-out (KO) of SAMHD1 was constructed by CRISPR-Cas9 genome-editing, specifically targeting the exon 5 that encodes the HD domain responsible of nucleotidase and phosphodiesterase activities of the SAMHD1 protein. Similarly, SAMHD1 KO cells showed increased replicative capacity in the presence of AZT that was reverted by re-expression of wild type SAMHD1. In addition, SAMHD1 KO effectively modified the anti-HIV-1 and cytotoxic activity of nucleoside analogues commonly used as antiviral or anti-cancer/leukemic agents, whereas sensitivity to a non-nucleoside inhibitor (nevirapine) or the integrase inhibitor raltegravir was not affected. The activity of SAMHD1 became apparent by addition of a competitive thymidine analogue. Our results demonstrate that the dNTPase activity of SAMHD1 remains active in apparently HIV-1 permissive cells and plays a role in modulating the sensitivity to nucleoside antiviral and anti-cancer agents.

58. Development of Nanoparticulate System for Vaccine Delivery

Marut Agarwal, MD, MPH[1], Saurabh Bhargava, MD, MPH[2]
[1]Manav Bharti University, India; [2]Himalayan University, India

Worldwide more than 300 million peoples are infected with the Hepatitis-B virus (HBV) and more than 1 million people die each year of liver failure hepatocellular carcinoma (HCC). The aim is to prepare controlled release hepatitis-B antigen containing poly ξ-caprolactone (PCL) based nanoparticulate system that provide a long-circulating antigen reservoir from which can be released into the vascular compartment in a controlled manner. Nanoparticulate system have an adjuvant properties, they are used for the antigen targeting to dendritic cell and activate dendritic cells to induce HBV antigen specific T-cell response.

In the present study, PCL nanoparticles were prepared by double emulsion (w/o/w) solvent evaporation method. Hepatitis-B surface antigen (HBsAg) was selected as model antigen to optimize formulation & process variables. The formulations were optimized on the basis of particle size & entrapment efficiency. The external morphology of the optimized formulation was studied by SEM & TEM. Stability studies were performed at accelerated temperatures. The in-vivo study included fluorescence microscopy and estimation of serum for antibody titres.

The results show that mice immunized with intramuscular injection of nanoparticles loaded HBsAg produced higher immune responses as compared to the marketed preparation.

In conclusion, we have demonstrated that hydrophobic nature of PCL provides danger signals for dendritic cells and activated, so PCL based nanoparticles a valuable means of delivering a soluble antigen to dendritic cell and provides enormous potential in developing new vaccines against hepatitis B.

59. Butyrate Prodrugs of IHVR-19029 with Enhanced Oral Exposure and Prevention of Gastrointestinal Glucosidase Interaction

Yanming Du, PhD, Jia Guo, PhD, Fang Guo, MD, PhD, julia Ma, BS, Xuexiang Zhang, MS, Qing Su, PhD, Nicky Hwang, BS, Ju-Tao Guo, MD, Timothy Block, PhD, Jinhong Chang, MD, PhD
Baruch S. Blumberg Institute, Doylestown, Pennsylvania, United States of America

We discovered a novel N-alkyl-ureanyl-deoxynojirimycin, IHVR-19029, that has been demonstrated to significantly protect mice from lethal infection of Marburg and Ebola virus when administrated via injection route through inhibition of the host endoplasmic reticulum (ER) α-glucosidases I and II. However, the major obstacles toward development of oral available IHVR-19029 are their short plasma half life, low oral bioavailability and inhibition of carbohydrate-metabolizing gut glucosidases, which results in osmotic diarrhea side effect. To overcome these problems, several types of prodrugs were designed and synthesized, including ester, carbonate, and amino acid prodrugs each with fully protection or partial protection of the four hydroxyl groups on DNJ core of IHVR-19029. As enzymatic assays showed, all the prodrugs lost the ability to inhibit ER α-glucosidases I and II, suggesting that these ester prodrugs would have reduced activity against GI α-glucosidases, and potentially overcome the off-target effects of the parent compound. In addition, *in vitro* ADME profiling studies demonstrated that while all the prodrugs remained intact in simulated gastric fluid, most of them subjected to rapid conversion to the parent drug either within the circulation and/or inside the cells. Pharmacokinetic profiling of the representative acetate, butyrates, isobutyrate prodrugs in mice demonstrated that oral delivered butyrate prodrugs can be very rapidly and efficiently metabolized leading to the significantly increased overall exposure to parent compound than that of direct administration of parent compound. The selected prodrugs will be advanced into animal efficacy studies in mouse and non-human primate models of Ebola virus infection.

60. Development of a Model for Enterovirus D68 Infection in Mice

Bart Tarbet, PhD, Brett Hurst, MS, Joseph Evans, BS
Institute for Antiviral Research, Utah State University, Logan, Utah, United States of America

In 2014, a mutated strain of enterovirus D68 (EV-D68) emerged in the U.S. This novel strain has only six coding differences from previous strains found in the U.S. However, of the six genetic changes in the EV-D68 polyprotein, five are present in neuropathogenic poliovirus. In 2015, we began development of a model for EV-D68 infection in AG129 mice. EV-D68 virus was serially-passaged in 4-week-old mice. For each passage, mice were infected intranasally (i.n.), and virus recovered from lung tissue three days post-infection (p.i.). Following inoculation, virus titer increased in the lung to 10^7.5 $CCID_{50}$ by 8 hours p.i., and to 10^8.5 $CCID_{50}$ by 24 hours p.i., A time course of infection determined that virus titer peaked in blood at 10^5.5 $CCID_{50}$ on day 2 p.i., and also spread to liver, kidney, and spleen, with clearance of virus by day 9 p.i. In addition, histological changes and an increase in pro-inflammatory cytokines were observed in lung tissues as the virus adapted to mice. Histological lesions, with a peak on days 3-4, included interstitial inflammation and alveolar wall injury. Cytokine involvement included increases in MCP-1 and RANTES on days 1-5, and 1-7, respectively. An evaluation of lung function by plethysmography showed a significant increase in enhanced pause (Penh), on days 5-7 p.i., indicative of morbidity. This is the first report of a respiratory disease model for EV-D68 infection in mice, and has potential use for evaluation of experimental therapeutics and vaccines. *[Supported by Contract HHSN272201000039I from the Virology Branch, DMID, NIAID, NIH]*

61. Discovery and Characterization of Broad-spectrum Inhibitors of Coronaviruses

Kin Kui LAI, MS, Jun DAI, PhD, Kwok Yung YUEN, MD, Richard Yi Tsun KAO, PhD
Department of Microbiology, Li Ka Shing Faculty of Medicine, The University of Hong Kong, Hong Kong

A decade after the SARS-CoV outbreak, zoonotic MERS-CoV has emerged in 2012 and caused approximately 36% of mortality for reported cases. This has prompted the WHO to put Coronaviruses (CoVs) on the list of "top 8 diseases likely to cause severe epidemics" in order to raise public concerns. No vaccine or specific treatment is currently available to treat CoV infections. Our group has previously identified 104 anti-SARS-CoV compounds from high-throughput screening (HTS). These hits were further tested against MERS-CoV and 229E-CoV, and finally two hit compounds, CA-616 and CA-607, were found to be effective. Time of addition and pseudovirion assays have revealed that these two compounds may target CoV entry, while the immunofluorescence assay indicated that the virus was trapped in endosome in compound-treated groups. We thus hypothesized that both compounds may inhibit the viral fusion in endosome by either targeting the Spike protein (S) of CoVs or prohibiting cathepsin L (CTSL) activity that shoulders S activation in endosome. Differential scanning fluorimetry suggested that CA-616 and CA-607 neither physically interact with the purified S of SARS-CoV, nor block cell-cell membrane fusion in polykaryon formation assay. Instead, they carry cell-free and cell-based inhibitory effect on CTSL activity, suggesting that two compounds inhibit CoVs by acting as CTSL inhibitors. This study not only illustrates the biological importance of CTSL in CoV infection, but also provides two lead compounds for the development of broad-spectrum anti-CoV agents.

62. Ribavirin-Imprinted Polymers as Drug Delivery System

Mohamed AYARI, MS, Patrick FAVETTA, PhD, Vincent HERVIN, MS, Vincent Roy, PhD,
Luigi Agrofoglio, PhD
ICOA UMR CNRS 7311 – Universite Orleans, Orleans, France

Respiratory infections, such as influenza, are still one of the major causes of death, which have an impact on other chronic pathologies such as asthma and bronchitis. Ribavirin (l-β-D-ribofuranosyl-l,2,4-triazole-3-carboxamide) has long been employed as an antiviral agent, mainly through an oral and intravenous administration. In some pulmonary viral diseases, ribavirin is known to be administered by aerosol inhalation but, unfortunately, this requires slow dosing of 12-20 hours per day of drug inhalation over at least a three-day period, all of which provides no more than a few hundred milligrams of drug. The molecular imprinting technology (MIP) has an enormous potential for creating satisfactory drug dosage forms through rate-programmed drug delivery, where drug diffusion from the system has to follow a specific rate profile.

Thus, herein, we describe the synthesis of a ribavirin-MIP (bulk, hydrogel, beads) as a new drug delivery system (DDS) for sustained release of ribavirin. Some MIPs were synthetized with ribavirin as template, using various monomers (e.g., acrylamide, 5-vinyluracil) and cross-linkers in different solvent. After optimization of the ratio template/monomer/cross-linker, the adsorption isotherms, the swelling ratio, and the kinetics of release were determined for designed MIPs and NIPs. The releasing kinetics of ribavirin were realized in a buffer mimicking lung biofluid and were function to temperature.

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63. Evaluation of the Innate Immune Modulator Acitretin as a Novel Strategy to Clear HIV Reservoir

Edurne Garcia-Vidal, MS, Maria Pujantell, MS, Roger Badia, PhD, Bonaventura Clotet, MD, PhD, Eva Riveira-Muñoz, PhD, Ester Ballana, PhD, José Esté, PhD
Irsicaixa – AIDS research institute, Badalona, Spain

The persistence of latent HIV despite suppressive antiretroviral therapy is a major roadblock for HIV eradication. Current strategies focused on inducing the expression of latent HIV fail to clear the persistent cellular reservoir, prompting the development of new approaches for killing HIV reactivated cells. Recently, acitetrin, a retinoic acid derivative, has been proposed as a pharmacological innate cellular-defense network enhancer that led to reactivation and preferential death of reactivated cells. Here, we have evaluated the capacity of acitretin to reactivate and/or facilitate immune-mediated clearance of HIV latently infected cells. When assessing the effect of acitretin on ACH-2 latently infected T-cells, we could only observe a modest induction of HIV transcription in comparison to suboptimal concentrations of current latency-reversing agents (LRAs), measured by RNA production in the supernatant. Acitretin was not able to induce HIV reactivation in GFP-expressing latently infected cell lines J-Lat (clones 8.4 and 9.2) or in latent GFP-HIV infected Jurkat cells. Additionally, acitretin induction was insignificant when compared to other LRAs optimal concentrations. Similarly, acitretin failed to induce reactivation in a primary model of latently infected CD4+ T-cells. Combination of acitretin with LRA did not show any significant change in HIV reactivation in any of the models tested. Acitretin treatment was able to induce RIG-I and MAVS expression in infected and uninfected cells, confirming the role of acitretin as an innate immune modulator. Our results suggest that acitretin-mediated stimulation of the RIG-I pathway over HIV reactivation is modest and thus may not meaningfully impact the HIV reservoir.

64. Antiadenoviral Activity of New Nanoparticles

Yulia Pankivska, MS[1], Liubov Biliavska, PhD[1], Olga Povnitsa, PhD[1], Svitlana Zagorodnya, PhD[1], Anatoly Dorovskykh, PhD[2], Mykhailo Lokshyn, PhD[3], Valery Lozovski, PhD[4], Volodymyr Lysenko, PhD[3], Valentyn Tertykh, PhD[4]

[1]Zabolotny Institute of Microbiology and Virology of the NASU, Kyiv, Ukraine; [2]LLC Scientific-Production Enterprise «International Medical Center», Kyiv, Ukraine; [3]V. Laskariov Institute of Semiconductor Physics of the NASU, Ukraine; [4]Institute of High Techologies T. Shevchenko National University, Kyiv, Ukraine; [5]Chuiko Institute of Surface Chemistry of the NASU, Kyiv, Ukraine

Nanoparticles occupied an important place in antiviral therapy. Nanoparticles that were functionalized with different types of biomolecules and medicines were used as drug delivery agents. However, non-functionalized nanoparticles were also demonstrated to have antiviral action. Previously, the authors have shown that the field action due to local-field enhancement effect is one of the main reasons of antiviral activity of nanoparticles*. In the present work, the antiviral activity of preparation of composite SiO_2-Au, Au and Au with SiO_2 shell nanoparticles against adenovirus was studied.

The reference strain of human adenovirus serotype 5 and monolayer cell line MDBK were used in the experiment. The preparations of nanoparticles with initial concentration ~ 10^{12}-10^{13} cm^{-3} were studied. As a result, the preparation of composite Au-SiO_2 in the dilution range of 10^{-2}–10^{-6} was shown to inhibit the virus reproduction by 90-100%. Similar results were obtained for preparations of shelling nanoparticles that inhibited virus reproduction by 60-90% at the same range of dilutions. In addition, analysis of the virucidal action of the nanoparticles preparations during 1-4 hours showed that the test component of composite Au-SiO_2 in dilution of 10^{-5} inhibited the virus activity by 92% after 3 hours of incubation, whereas viral infectivity was inhibited by 86% after 2 hours of incubation with nanoparticles in the dilution of 10^{-6}.

The nonmonotonic correlation between nanoparticles concentration or incubation time and the level of virus inhibition demonstrates complicated picture of the interaction between nanoparticles, viruses and living cells.

*V.Lysenko, V.Lozovski, M.Spivak, *Ukr.J.Phys*.58, 77 (2013)

65. Kinetic, Structural and Thermodynamic Analysis of the H275Y, I223V and S247N Neuraminidase Resistant Mutants of H1N1 2009 Pandemic Influenza Virus

Milan Kožíšek, PhD, Jana Pokorná, PhD, Petr Pachl, PhD, Pavlína Řezáčová, PhD, Aleš Machara, PhD, Jakub Hejdánek, BS, Elena Karlukova, BS, Jan Konvalinka, PhD

Institute of Organic Chemistry and Biochemistry of the Czech Academy of Sciences, Prague, Czech Republic

Influenza is an acute viral infection that can cause serious complications and death. The influenza glycoprotein, neuraminidase (NA), is an essential enzyme for the last step of viral cycle, the release of the viral particles from the host cell. To date, two neuraminidase inhibitors (NAIs) have been licensed worldwide for therapeutic and prophylactic uses (oseltamivir marketed as Tamiflu and zanamivir as Relenza) and two others have been authorized in various countries for the emergency treatment during pandemics.

However, the clinical isolates of recently emerged pandemic type H1N1 („swine flu", 2009) were reported to show increasing frequency of resistence. New neuraminidase inhibitor resistance substitutions I223V and S247N alone or in combination with a major oseltamivir resistance mutation H275Y have been observed recently in the 2009 pandemic H1N1 viruses.

We overexpressed the ectodomain of the wild type neuraminidase from the influenza virus A/California/07/2009 (H1N1) as well as enzymes containing H275Y, I223V, and S247N single mutation and the H275Y, I223V and H275Y, S247N double mutants in *Drosophila* Schneider S2 cells and purified them by one-step purification using a streptavidin derivative. In order to quantify the level of resistance we enzymologically characterized these enzymes with oseltamivir carboxylate. Thermodynamic analyses of oseltamivir carboxylate binding to neuraminidase monomutants were performed by isothermal titration calorimetry. Finally, we co-crystallized neuraminidase variants in complexes with oseltamivir carboxylate to structurally explain the resistance mechanism.

66. Cardiovascular Mortality of HIV-infected Patients

Valentina Golyshko, PhD, Victor Snezhitskiy, MD, PhD, **Natallia Matsiyeuskaya, MD, PhD**
Grodno State Medical University, Grodno, Belarus

AIM OF STUDY: to evaluate the frequency and structure of cardiovascular pathology leading to death in HIV-infected patients in dependence of HAART receiving.

MATERIAL & METHODS: There was made a retrospective analysis of medical records of 210 HIV-infected patients who died in Grodno region of Belarus since 2000 to 2015. Among them there were 143 (68.9%) men and 67 (31.9) women; the age (Me) was 36,5 [33,0;42,0] years. The 1st clinical stage (WHO, 2012) was in 55 (26.1%) cases, the 2nd – in 10 (4.8%), and the third – in 63 (30%;) patients, the 4th stage of HIV infection – in 82 (39.1%) patients. HAART was received by 62 (29,5%) HIV-positive persons.

The results were processed with «STATISTICA 7.0» program.

RESULTS: Blood circulatory system diseases were the main cause of death in 3 (4.8%) patients receiving HAART – and in 13 (8.9%) patients – without HAART (p>0,05). Chronic forms of coronary artery disease as the cause of death have been established in 5 (2.4%) patients without HAART and in 2 (0.9%) on treatment (p>0,05). Dissecting aortic aneurysm in a patient on HAART was found at one (0.5%) case. AIDS-related pathology of cardiovascular system was installed only in 8 (3.8%) patients without HAART: in 6 (2, 8%) of them were diagnosed with cardiomyopathy, in 2 (0.9%) cases – infective endocarditis.

CONCLUSION: The absence of HAART in patients in the study group was associated with a higher mortality rate from AIDS-related cardiovascular disease compared with patients receiving therapy (p = 0,05).

67. High-Throughput Screening to Identify Inhibitors of BSL-4 Viruses

Mike Flint, PhD, Payel Chatterjee, MS, Stephen Welch, PhD, Michael Lo, PhD, Laura McMullan, PhD, Eric Bergeron, PhD, Cesar Albarino, PhD, Stuart Nichol, PhD, Christina Spiropoulou, PhD
Viral Special Pathogens Branch, Centers for Disease Control and Prevention, Atlanta, Georgia, United States of America

The tragic 2013–2015 Ebola virus epidemic in West Africa highlighted the need for medical countermeasures, both vaccines and antivirals, for viral hemorrhagic fevers. To facilitate campaigns to identify small molecule inhibitors of these viruses under biosafety level 4 containment, we generated recombinant viruses expressing fluorescent reporter proteins: Ebola virus, Lassa fever virus, Crimean-Congo hemorrhagic fever virus, Nipah virus, and Rift Valley fever virus. Each of these recombinant viruses undergoes a full replication cycle and is infectious. Compounds with antiviral effects may be detected by reduction of fluorescence in cells treated with the compound and infected with the recombinant viruses. A sixth virus, Alkhurma hemorrhagic fever virus (AHFV), causes a robust cytopathic effect in A549 cells, and an assay based on cell viability was used to test for compounds with anti-AHFV activity.

Each of these assays was optimized for use in 384-well plates, and each demonstrated Z -factors > 0.6, indicating their suitability for high-throughput screening. Details of assay setup and performance will be described. A test library of FDA-approved compounds was used to establish the reproducibility of these assays, and data on hits from this test library against each of the viruses will be presented. Screening of a diverse library of 50,000 compounds is underway.

68. Development of Quinazolinone-Based Inhibitors Against Venezuelan Equine Encephalitis Virus

Nikhil Madadi, PhD[1], Omar Moukha-Chafiq, PhD[1], Saibal Chakraborthy, PhD[1], Daniel Streblow, PhD[2], Nicole Haese, PhD[2], Thomas Morrison, PhD[3], Nicholas May, PhD[3], Mark Heise, PhD[4], Victor DeFilippis, PhD[2], Corinne Augelli-Szafran, PhD[1], Mark Suto, PhD[1], Ashish Pathak, PhD[1]
[1]Southern Research; [2]Oregon Health & Science University; [3]University of Colorado School of Medicine; [4]University of North Carolina at Chapel Hill

Venezuelan equine encephalitis virus (VEEV) is a positive-sense RNA virus capabale of causing neurological disease in humans and lethal encephalitis in equines. During natural outbreaks, humans become infected by the bite from an infected arthropod. However, VEEV is highly infectious when transmitted by aerosol and thus, has been developed as a bio-warfare agent. Currently, there are no FDA approved treatments to treat VEEV infection and the available vaccines show insufficient efficacy or cause adverse side effects that limit their use. Given the increased incidence of this virus and the absence of effective treatments, a research program aimed at identifying small molecule-derived VEEV inhibitors was undertaken. A high throughput screen yielded a quinazolinone hit that inhibited a VEEV-induced cytopathic effect (CPE) in the low micromolar range. Medicinal chemistry efforts on this quinazolinone hit led to the identification of SRI-36959 which showed inhibition of a VEEV virus plaque formation in the NHDF fibroblast cells (EC_{90} = 0.78 µM) with no cytotoxicity (CC_{50}> 50 µM). Furthermore, SRI-36959 dramatically reduced viral titers by 3 log units at 0.78 µM. Further investigation of SRI-36959 and its analogs may provide novel therapeutic avenues for the treatment of VEEV infection.

69. Using a Recombinant Crimean-Congo Hemorrhagic Fever Virus Expressing a Fluorescent Protein to Rapidly Evaluate Synergistic Properties of Antiviral Compounds.

Stephen Welch, PhD, Florine Scholte, PhD, Mike Flint, PhD, Éric Bergeron, PhD, Christina Spriropoulou, PhD
Centers for Disease Control and Prevention, Atlanta, Georgia, United States of America;

Crimean-Congo hemorrhagic fever virus (CCHFV), a tick-borne nairovirus (family *Bunyaviridae*), is a cause of severe hemorrhagic disease in humans. Currently, there are no licensed vaccines and treatment is limited to supportive care and the use of ribavirin, although its therapeutic benefit remains unclear. CCHF is part of WHO's priority list of infectious diseases warranting further research and development. To aid in the identification of new antiviral compounds we generated a recombinant CCHFV, based on the IbAr10200 strain, that expresses the ZsGreen (ZsG) fluorescent reporter protein. This was achieved by creating a modified CCHFV S-segment in which the ZsG coding sequence, followed by the self-cleaving P2A sequence derived from porcine teschovirus-1, was inserted immediately upstream of the nucleoprotein coding sequence. Inhibition of replication of the recombinant virus is assessed by measuring the reduction in ZsG fluorescence in infected cells treated with candidate compounds. The assay was readily adaptable to high-throughput screening (HTS) of compounds, with a signal-to-noise ratio of 40:1, and Z'-factors >0.6 in both a 96- and 384-well format. A screen of compounds with known antiviral activity identified both 2'-deoxy-2'-fluorocytidine (EC_{50} = 52±12 nM), and mycophenolic acid (EC_{50} = 57±43 nM) as having >300× the potency of ribavirin (EC_{50} = 15.5±2.6 µM). Use of a fluorescent reporter allowed us to rapidly assess the synergistic potential of these three compounds in conjunction with another compound with reported anti-CCHFV activity, favipiravirir (EC_{50} = 810±256 nM). This approach will allow high-throughput screening to identify more effective therapeutic options to treat CCHFV infection.

70. Benzoannulenes as Inhibitors Against Chikungunya Virus

Syed Ahmed, PhD[1], Vibha Pathak, MS[1], Jaden Cowan, BS[1], Daniel Streblow, PhD[2], Nicole Haese, PhD[2], Nicholas May, PhD[3], Thomas Morrison, PhD[3], Mark Heise, PhD[4], Victor DeFilippis, PhD[2], Corinne Augelli-Szafran, PhD[1], Mark Suto, PhD[1], Ashish Pathak, PhD[1]

[1]Southern Research, Birmingham, Alabama, United States of America; [2]Oregon Health & Science University, Beaverton, Oregon, United States of America; [3]University of Colorado School of Medicine, Aurora, Colorado, United States of America; [4]University of North Carolina at Chapel Hill, Chapel Hill, North Carolina, United States of America

Arthropod-borne viruses have developed a complex life cycle that alternates between insect and vertebrate hosts. These arthropod-borne viruses mainly belong to the virus families called Togaviridae, Flaviviridae, and Bunyaviridae. *Alphavirus* is a genus within the Togaviridae family. This genus includes viruses such as chikungunya virus (CHIKV), Venezuelan Equine Encephalitis virus, and Eastern Equine Encephalitis virus. Despite the widespread distribution and severity of viral infection, a virus specific treatment for CHIKV is still lacking. In search for inhibitors of CHIKV, benzoannulene, SRI-33366 was identified that showed inhibition in CHIKV virus plaque formation in the NDHF fibroblast cells (EC_{90} = 2.5 μM) and reduced viral titers by 2.5 log units at 10 μM with no cytotoxicity (CC_{50} > 50 μM). Medicinal chemistry was initiated to improve the potency and efficacy of SRI-33366. This structure-activity relationship study and results will be discussed.

71. Anti-Dengue Activity of Traditional Chinese Medicinal Plants

Maqsood Maryam, MS[1], Kian Keong Te, PhD[2], Fai Chu Wong, PhD[3], Tsun Thai Chai, PhD[3], Seng Chiew Gan, PhD[2], Gary Low, PhD[2], Hui Yee Chee, PhD[1]

[1]Faculty of Medicine and Health Sciences, University Putra Malaysia, Serdang, selangor, Malaysia; [2]Faculty of Medicine and Health Sciences, University Tunku Abdul Rahman, Sungai Long, Selangor, Malaysia; [3]Department of Chemical Science, Faculty of Science, University Tunku Abdul Rahman, Kampar, Perak, Malaysia

INTRODUCTION: Dengue virus (DENV) is emerging as a major virus spread by mosquito. Recently it has spread to more than a hundred countries around the globe and still lacks a specific treatable medication. Many Traditional Chinese Medicine (TCM) plants are in practice for dengue fever in dengue endemic regions of the world. Research can help TCM practitioners identify the anti-dengue potential of the plants in clinical practice.

METHODS: Twelve TCM plant (extracts) described as cool herbs used for the diseases with high fever were screened for their anti-dengue potential. Lead plants were established through detailed *in vitro* foci forming unit reduction analysis against all four serotypes and were validated through quantitative real time RT-PCR.

RESULTS: Four plants were potentially inhibiting the virus in primary phenotypic *in vitro* screening. Two lead plants *Dryopteris crassirhizoma* (DC) and *Morus alba* (MA) were identified with IC50 values 130ug/ml and 221ug/ml respectively and the Selectivity index (SI) were 4.21 and 4.62 respectively. These two were tested against all four serotypes of DENV and were found equally inhibiting all. Whereas, qRT- PCR RNA copy number reduction analysis suggested DC to have slightly more anti-dengue activity than MA.

CONCLUSION: Rhizome of DC is identified as potential anti-dengue and can be made available for the future studies to help TCM practitioners. Leaves of MA are also inhibiting but inhibiting dose is very high so it can be considered as potential anti-dengue but further research is required to improve its activity.

72. Inhibition of Dengue Virus by Novel Inhibitors of RNA-Dependent RNA Polymerase and Protease Activity

Giuseppe La Regina, PhD[1], Valeria Famiglini, PhD[1], Valentina Naccarato, MS[1], Antonio Coluccia, PhD[1], John Hiscott, PhD[2], Jin-Ching Lee, PhD[3]

[1]Sapienza University, Institut Pasteur Italy – P.le A. Moro 5, I-00185 Roma, Italy, Roma, Italy; [2]Institut Pasteur Italy, Viale Regina Elena 291, 00161 Roma, Italy; [3]National Cheng Kung University, Taiwan, Taiwan

Dengue virus (DENV) is the leading mosquito-transmitted viral infection in the world. With more than 390 million new infections annually, and up to 1 million clinical cases with severe disease manifestations, there is an urgent need to develop new antiviral agents that inhibit DENV infectivity.[1] Currently, no licensed antiviral drugs are available to block DENV infection and vector control efforts remain the only means to stop the spread of the infection.

In the present study, we focused our attention on the identification of potential anti-DENV inhibitors by targeting the enzymatic activities of the NS5 RdRp polymerase and NS3 protease, *in vitro and in vivo*. As part of a continuation of our studies,[2,3] we developed new pyrazole derivatives 1-3 as inhibitors of NS5 RdRp polymerase (Chart 1). Furthermore, virtual screening studies on the NS2B/NS3 protease led us to identify indole derivatives 4-5 as inhibitors of NS3 protease (Chart 1). New compounds exhibited anti-DENV replication activity without cytotoxicity; two compounds exhibited anti-DENV activity in ICR suckling mouse model of DENV infection. Interestingly, combination treatment with several compounds demonstrated a synergistic inhibitory effect on DENV replication.

[1] Wilder-Smith, A.; Ooi, E. E.; Vasudevan, S. G. et al. *Curr. Infect. Dis. Rep.* 2010, *12*, 157-164.

[2] Silvestri, R.; Cascio, M. G.; La Regina, G. et al. *J. Med. Chem.* 2008, *51*, 1560-1576.

[3] La Pietra, V.; La Regina, G.; Coluccia, A. et al. *J. Med. Chem.* 2013, *56*, 10066-10078.

73. Lupeol and Other Compounds from Natural Sources Exhibited Antiviral Activity Against Enteroviruses 7, 13 and 19

Omonike Ogbole, PhD[1], Abidemi Sunmola, BS[2], Adekunle Adeniji, PhD[3]

[1]Department of Pharmacognosy, Faculty of Pharmacy, University of Ibadan, Ibadan, Nigeria; [2]Department of Pharmaceutical Microbiology, Faculty of Pharmacy, University of Ibadan, Ibadan, Nigeria; [3]Department of Virology, College of Medicine, University of Ibadan, Ibadan, Nigeria

Enteroviruses are a large family of viruses responsible for many infections especially in children. These viruses live in the intestinal tract, but can cause a wide variety of illnesses. Enteroviruses 7, 13 and 19 are part of the diseases causing enteric viruses identified from Nigeria. Presently, no vaccines or antiviral drugs have been clinically available to employ against these viruses. Previous work has established the antiviral activity of lupeol, chrysophanol and physcion and β-sitosterol isolated from natural product (*Cassia siamea* plant) against poliovirus (enterovirus). In continuation of our work we determined the antiviral activities of this compounds against EV 7, EV13 and EV19.

In this study, the ability of these compounds to reduce the viral cytopathic effect on rhabdomyosarcoma cells was evaluated. Using a serial dilution of the maximum non-toxic dose (MNTD) of each compound, the ability to inhibit viral-induced cell death in tissue culture was assessed three days' post-infection by colorimetric method using MTT dye (3-[4,5-dimethylthiazol–2-yl]-2,5-diphenyltetrazolium bromide). Data obtained were analyzed statistically using Microsoft excel.

Lupeol was the most active with an IC_{50} of 0.417 and 0.329μg/mL on EV 7 and EV19 respectively. EV13 was only slightly inhibited by β-sitosterol at the highest concentration tested.

EV 13 was not sensitive to tested compound, a compound is considered sensitive only if its active in two subsequent dilution of MNTD, thus it cannot be considered sensitive to β-sitosterol. The active compounds may represent a potential therapeutic agent to control infections due enterovirus 7 and 19

74. Antiviral Potentials of *Omidun* and Selected Lactic Acid Bacteria Against Selected Human Enteroviruses (HEV)

Abidemi Sunmola, BS[1], Funmilola Ayeni, PhD[1], Omonike Ogbole, PhD[2], Temitope Faleye, MS[3], Adekunle Adeniji, PhD[3]

[1]Department of Pharmaceutical Microbiology, Faculty of Pharmacy, University of Ibadan, Ibadan, Nigeria; [2]Department of Pharmacognosy, Faculty of Pharmacy, University of Ibadan, Ibadan, Nigeria; [3]W.H. O Polio Laboratory, Department of Virology, University of Ibadan, Ibadan, Nigeria

Various microorganisms, in particular, lactic acid bacteria (LAB) are involved in common food fermentation processes. *Omidun* is the supernatant of fermented cereal from grains, the bacteria responsible for its fermentation belong to the LAB group. *Omidun* has been traditionally used to reduce stooling frequency in young diarrhoeal patients. Diarrhoeal caused by enterovirus are a major health problem, particularly among children, accumulating evidence suggests that specific probiotic bacteria are able to decrease the risk and symptoms of these infections. This study aimed to determine whether *Omidun* may confer protection in the GIT against HEV.

In this study, the antiviral activity of *Omidun, Lactobacillus plantarum, Lactobacillus amyllovorus* and *Enterococcus hairea*, the cell free supernatant (CFS) and bacterial pellet of the LABs were determined against Enterovirus 7 (EV7), Enterovirus 13 (EV13) and Enterovirus 19 (EV19) using MTT colorimetry assay.

Omidun has the highest activity against EV7 compared to the three LABs used; *Omidun* and the LABs used did not display any significant are not active against EV13 while *Lactobacillus amyllovorus* shows highest activity against EV19 followed by *Omidun*. But the CFS of *Enterococcus hairea* shows a high degree of activity. There is a statistical difference ($P > 0.05$) between the activity of the CFS and pellet of the LABs.

The data generate indicate that O*midun* has the potential to inhibit the infectivity of certain Human Enteroviruses thus further research should be done to explore it mode of action. In vivo experiment should be done to support the claim.

75. Synthesis of γ-Modified Nucleoside Triphosphates

Simon Weising, MS[1], Dominique Schols, PhD[2], Jan Balzarini, PhD[2], Chris Meier, PhD[1]

[1]Organic Chemistry, Department of Chemistry, University of Hamburg, Hamburg, Germany; [2]Rega Institute for Medical Research, Katholieke Universiteit Leuven, Leuven, Belgium

In the last few decades a variety of antiviral active nucleosides were developed to combat antiviral infections. Inside cells these nucleosides need to be phosphorylated stepwise to their corresponding active triphosphate form, but often one of these phosphorylation steps is rate limited. To bypass this problem it's possible to use the appropriate mono-, di- or triphosphate prodrugs, which have to be stable and lipophilic enough to penetrate the cell membrane. It is known that short alkyl- or phenol groups can lead to increasing stability while still being substrates for different polymerases. However, such compounds can not be used as potential antivirals due to their high polarity that prevent cellular uptake. In contrast, higher lipophilic residues may combine high stability and high lipophilicity.

Herein we present the synthesis of γ-lipophilic-modified nucleoside triphosphates. The synthesis was achieved by nucleophilic opening of an activated *cyclo*Sal-phosphotriester (see figure) by nucleoside diphosphates. The stability of these γ-modified nucleoside triphosphates in CEM/0 cell extracts will be shown.

76. Pyrrole and Pyrazole-3-Carbothioamide Derivatives Act as Dual Allosteric Inhibitors of HIV-1 Reverse Transcriptase

Angela Corona, PhD[1], Valentina Onnis, PhD[1], Alessandro Deplano, PhD[1], Monica Demurtas, MS[1], Simona Distinto, PhD[1], Giulia Bianco, PhD[1], Stefano Alcaro, PhD[2], Francesca Esposito, PhD[1], Enzo Tramontano, PhD[1]

[1]University of Cagliari, Cagliari, Italy; [2]University of Magna Graecia

In the continuous effort to identify new HIV-1 inhibitors endowed with innovative mechanisms, the dual inhibition of different viral functions is a particularly interesting goal since it would provide a significant advantage against the selection of drug resistant variants. The HIV-1 Reverse Transcriptase (RT) associated Ribonuclease H (RNase H) is the only viral encoded enzymatic activity that still lacks an efficient inhibitor, and its co-presence in the RT with the RNA dependent DNA polymerase (RDDP) activity strongly encourages the possibility to reach the simultaneous inhibition of both activities with one molecule. In the present work we synthesized a small library of 3,5-diamino-N-aryl-1*H*-pyrazole-4-carbothioamide and 4-amino-5-benzoyl-N-phenyl-2-(substituted-amino)-1*H*-pyrrole-3-carbothioamide derivatives and tested them against both HIV-1 RT associated activities. We identified a pyrazole-4-carbothioamide, A15, able to inhibit both RNase H/RDDP RT-associated activities in the low micromolar range, being also active against viral replication. Its binding mode, investigated by molecular dynamics, provided interesting insights for the scaffold optimization. The V108A substitution strongly affected the A15 IC_{50} values (12.6 fold in RNase H and 4.7 fold in RDDP, respectively), and the A502F substitution caused a 9.0 fold of increase in RNase H IC_{50} value only, reinforcing the hypothesis of a dual inhibition achieved by the binding to two different sites on the same enzyme. Moreover, A15 retained a good inhibition potency against three NNRTI resistant enzymes, confirming a mode of action unrelated to NNRTIs, and suggesting a great potential for A15 as a starting point for development of new RT dual inhibitors active against drug resistant viruses.

77. Novel Cell Targeting L-ddBCNA Antiviral Inhibits Autophagy in Virus Infected Cells

Rohan Narayan, MS[1], Laura Farleigh, PhD[2], Alina Tscherne, MS[3], Daniela Friese, BS[3], Ed Sayers, PhD[4], Arwyn Jones, PhD[4], **Joachim Bugert, MD, PhD**[3]

[1]Cardiff School of Pharmacy & Pharmaceutical Sciences, Cardiff, Wales, United Kingdom; [2]University of Cambridge, Cambridge, United Kingdom; [3]Institut für Mikrobiologie der Bundeswehr, München, Bavaria, Germany; [4]Cardiff School of Pharmacy & Pharmaceutical Sciences, Cardiff, Wales, United Kingdom

We have previously reported that dideoxy bicyclic pyrimidine nucleoside analogues with L-chirality (L-ddBCNAs) exhibit antiviral activity against measles and vaccinia viruses and suggested a cellular target (1). Here we report, that the lead compound cf2642 completely inhibits the formation of measles wildtype virus syncytia in both B95a and VerohSLAM cells with an IC90 of 10µM, without reduction of cellular ATP production or cell-to-cell adhesion at 12 hours' post-infection. Measles viruses are known to induce successive stages of autophagy in virus infected cells (2). Autophagy flux was monitored at 12 hours' post-infection by fluorescent cell sorting and quantitative Western blotting of LC3-II (microtubule-associated protein 1 light chain 3) and p62 protein, also known as sequestosome 1 (SQSTM1) in the presence and absence of cf2642. cf2642 inhibits measles virus induced autophagy in infected cells with an IC50 of 1µM. cf2642 was subsequently found to inhibit Zika virus plaque formation on A549 cells with an IC50 of 2.5µM. Flaviviruses were previously reported to be sensitive to autophagy inhibitors. We propose that cf2642 is a novel autophagy inhibitor affecting viruses inducing autophagy in their lifecycle. Autophagy is a cellular pathway modulated in many virus infections and in cancer cells.

78. Identification of Candidate Immunomodulatory Viral-Host Cell Interaction Between Dengue NS5 and Cellular PML Protein

Federico Giovannoni, MS[1], Peter Hemmerich, PhD[2], **Cybele Garcia, PhD**[3]
[1]Lab de Estrategias Antivirales, QB, FCEyN, UBA- Instituto de QB FCEyN-CONICET, Buenos Aires, Argentina; [2]Leibniz Institute on Aging – Fritz-Lipman-Institut, Jena, Germany; [3]Lab de Estrategias Antivirales, QB, FCEyN, UBA- Inst. QB FCEyN (IQUIBICEN)-CONICET., Buenos Aires, Argentina

Intrinsic immunity is a form of innate immunity that is mediated by constitutively-expressed cellular proteins which can block viral replication immediately.

Promyelocytic leukemia (PML) protein contributes to intrinsic immunity against many viruses. PML forms structures called nuclear bodies (PML-NBs) which are frequently targeted by viruses to overcome PML-mediated immunity.

Previously, we described the antiviral role of PML against dengue virus serotype 2 (DENV-2). Here, we performed further studies to characterize the role of PML in the *in-vitro* replication of other DENV serotypes (DENV1-4) as well as the molecular mechanism underlying this effect.

Confocal microscopy revealed that the number of PML-NBs was significantly lower in DENV1-4-infected A549 cells. Even though flaviviruses, such as DENV, replicate in the cytoplasm DENV proteins C and NS5 can localize to the nucleus. This nuclear localization is still considered enigmatic in DENV biology.

To determine if the disruption of PML-NBs could be consequence of an interaction between PML and C or NS5, cells were transfected with vectors encoding for C or NS5. Confocal images showed that quantitative expression of NS5, but not C, was sufficient to reduce the number of PML-NBs. Immunoprecipitation studies confirmed the interaction between NS5 and PML. Finally, confocal images also showed that only PML isoforms III and IV co-localize with NS5.

Overall, we show for the first time the interplay between PML and DENV1-4. Our data shows that NS5 interacts with PML and that it is responsible for disrupting PML-NBs. Therefore, NS5 nuclear localization could be important for inhibiting PML-mediated immune response.

79. Synthesis of Heterocycles Targeting the Inhibition of DNA Glycosylases Involved in Base Excision Repair Pathway

Zahira Tber, PhD[1], Charlotte Rieux, MS[2], Franck Coste, PhD[2], Norbert Garnier, PhD[2], Bertrand Castaing, PhD[2], Vincent Roy, PhD[1], **Luigi Agrofoglio, PhD**[1]
[1]ICOA UMR CNRS 7311 – Universite Orleans, Orleans, France; [2]CBM UPR CNRS 4301, Orleans, France

DNA, the carrier of genetic information, is constantly submitted to negative effects of exogenous and endogenous chemical and physical agents which modify its native structure. The decay of DNA is likely to be a major factor in mutagenesis, carcinogenesis, ageing but also in viral pathogenic diseases. To counteract these effects, living organisms have developed specific DNA repair strategies; like retrotransposons, recent data indicate that retroviral DNA integration is completed with the aid of host cellular proteins involved in DNA break repair. although a strong link between host cellular DNA damage response proteins and retroviral integration exists, the exact mechanism(s) still remain to be discovered.

In previous work, we established that 2-thioxanthine (2TX) is a suicide inhibitor of eukaryote and prokaryote zinc finger Fpg/Nei DNA glycosylases, one of the two families of glycosylases that recognize oxidized DNA bases, the other being the HhH/GPD (or Nth) superfamily. By combining chemical synthesis of 2TX derivatives, enzyme inhibition assays, molecular dynamics simulations, molecular docking, and crystal structure analysis, herein we will present a new enzyme binding site for nucleobase derivatives, common to all Fpg/Nei enzymes as well as the synthesis of 2nd generation inhibitors. Those data which provide foundation for the rational design of more effective inhibitors for this class of enzymes.

80. Drug Design and Synthesis of New Indolylarylsulfones as HIV-1 Non-nucleoside Reverse Transcriptase Inhibitors

Valeria Famiglini, PhD[1], Giuseppe La Regina, PhD[1], Antonio Coluccia, PhD[1], Domiziana Masci, PhD[1], Roger Badia, PhD[2], José A. Esté, PhD[2], Emmanuele Crespan, PhD[3], Giovanni Maga, PhD[3]
[1]Sapienza University, Institut Pasteur Italy – P.le A. Moro 5, I-00185 Roma, Italy, Roma, Italy; [2]AIDS Research Institute – IrsiCaixa, Universitat Autonóma de Barcelona, Badalona, Spain; [3]IGM-National Research Council, via Abbiategrasso 207, I-27100 Pavia, Italy, Pavia, Italy

HIV-1 non-nucleoside reverse transcriptase inhibitors (NNRTIs) are key drugs of highly active antiretroviral therapy (HAART) in the clinical management of AIDS/HIV-1 infection. Our recent studies showed that indolylarylsulfones (IASs) bearing a cyclic moiety at the 2-carboxamide nitrogen linked through a short spacer group were endowed with potent antiretroviral activity.[1,2]

Based on the results previously obtained, we aimed to expand the SAR studies by the introduction of new aryl or heteroaryl portions to the indole nucleus.

Interestingly, for the first time IASs endowed with asymmetric centre have shown significant differences in term of antiretroviral potency. In particular, the *R*-enantiomer proved to be exceptionally potent and uniformly superior to the *S*-enantiomer against the whole viral panel. Docking studies showed that the methyl group of the *R*-enantiomer (Figure 1) pointed toward the cleft created by the K103N mutation, differently from the corresponding group of (S) counterpart. By calculating the solvent accessible surface, we observed that the exposed area of the RT in complex with *S*-enantiomer was larger than the area of the (R) complex.[3]

REFERENCES. 1) La Regina, G., Coluccia A. *et al. J. Med. Chem.* 2012, *55*, 6634−6638. 2) Famiglini, V., La Regina, G. *et al. Eur. J. Med. Chem.* 2014, *80*, 101-111. 3) Famiglini, V., La Regina, G. *et al. J. Med. Chem.* 2014, 57, 9945-57.

81. 25-Hydroxycholesterol Inhibition of Lassa Virus Infection Through Aberrant GP1 Glycosylation

Punya Shrivastava-Ranjan, PhD, Eric Bergeron, PhD, Ayan Chakrabarti, MS, César Albariño, PhD, Mike Flint, PhD, Stuart Nichol, PhD, Christina Spiropoulou, PhD
Centre for Disease Control and Prevention, Atlanta, Georgia, United States of America

Lassa fever is an acute viral hemorrhagic fever in humans caused by Lassa virus (LASV). No vaccine for LASV is currently available. Treatment is limited to administration of ribavirin, which is only effective when given early in the course of illness. Cholesterol 25-hydroxylase (*CH25H*) is a recently identified interferon-stimulated gene (ISG); it encodes an enzyme that catalyzes the production of 25-hydroxycholesterol (25HC), which inhibits several viruses. Here, we identify a novel antiviral mechanism of 25HC that is dependent on inhibiting glycosylation of Lassa virus (LASV) glycoprotein and reducing the infectivity of LASV as a means of suppressing viral replication. Since N-linked glycosylation is a critical feature of other enveloped virus glycoproteins, 25HC may be a broad inhibitor of virus infectivity.

82. Brazilian Natural Compounds and Derivative Synthetic Analogues Efficiently Inhibit CHIKV and ZIKV Infection

Jacqueline Shimizu, MS[1], Suely Silva, BS[1], Daniel Martins, BS[1], Debora Oliveira, BS[1], Zsofia Igloi, PhD[2], Cintia Bittar, PhD[3], Paula Rahal, PhD[3], Luis Regasini, PhD[3], Andres Merits, PhD[4], Mark Harris, PhD[2], **Ana Carolina Jardim, PhD**[5]

[1]Laboratory of Virology, Federal University of Uberlândia, Uberlândia, MG, Brazil; [2]Institute of Molecular and cell Biology, University of Leeds, Leeds, UK; [3]Sao Paulo State University – UNESP, São Jose do Rio Preto, SP, Brazil; [4]University of Tartu; [5]Federal University of Uberlandia, Uberlandia, Minas Gerais, Brazil

Diseases caused by arboviruses involve complex cycles between vertebrate and hematophagous arthropod vectors and represent important causes of outbreaks and epidemics. Among these diseases, Chikungunya and Zika fevers have been receiving attention by the Brazilian government and public health authorities worldwide. There is no specific antiviral against chikungunya virus (CHIKV) and Zika virus (ZIKV) and current treatment is palliative. The efforts to develop innovative and specific drugs against these viruses are challenged by the high viral mutation rate and the need to develop drugs which can impair the virus with low damage to the host cell. In this context, natural and synthetic compounds are attractive candidates in the search for new therapeutic approaches, as numerous modern drugs have been developed from natural prototypes. Therefore, this study aims to investigate the antiviral effects of a panel of natural compounds isolated from Brazilian natural sources and synthetic analogues based on natural scaffolds. Cells were infected with CHIKV or ZIKV in vitro and immediately treated with compounds at maximum nontoxic concentration for 16 and 72 hours, respectively. Initial screening of a hundred compounds was performed and 12 compounds demonstrated anti-CHIKV or -ZIKV activity. More interestingly, a synthetic alkaloid reduced a minimum of 70% of both CHIKV and ZIKV infections, and a natural compound blocked up to 97% of both viruses infectivity. These data are the first description of Brazilian natural compounds possessing anti-CHIKV and -ZIKV activities and further analyses are being performed in order to investigate the mode of action of those compounds.

83. Identification Of PIK-III as a Novel Antiviral Against Pathogens Entering Cells by Macropinocytosis

Olena Shtanko, PhD, Robert Davey, PhD

Texas Biomedical Research Institute, San Antonio, Texas, United States of America

Macropinocytosis is a way for eukaryotic cells to engulf extracellular liquid and dissolved molecules. Many viruses, including those belonging to families *Filoviridae* and *Poxviridae*, subvert this endocytic process to enter host cells. Here, we report that PIK-III, a selective inhibitor of PI3K type III enzymatic activity, is a potent inhibitor of macropinocytosis. The compound efficiently inhibited uptake of the fluid-phase marker dextran and infection by viruses pseudotyped with either Ebola or Marburg virus glycoproteins, each known to require macropinocytosis for trafficking into the cell. Treatment of cells with PIK-III blocked macropinosome formation at the earliest detectable step when Ankfy1 protein and PtdIns(3,4,5)P3 lipid mark sites of macropinocytic cups. We further show that the compound blocked infection of cells by replication-competent Ebola and Marburg viruses at nanomolar concentrations. We are currently testing whether PIK-III and its derivatives have an inhibitory effect on additional filoviruses as well as vaccinia virus, a member of the poxvirus family. This work was funded by the Ewing Halsell foundation.

84. Cell Line Stably Expressing a zsGreen Minigenome Enables Drug Screening Using all Known Ebolaviruses

Markus Kainulainen, PhD, Mike Flint, PhD, Cesar Albarino, PhD, Christina Spiropoulou, PhD
Viral Special Pathogens Branch, Centers for Disease Control and Prevention, Atlanta, Georgia, United States of America

The findings and conclusions in this report are those of the authors and do not necessarily represent the official position of the Centers for Disease Control and Prevention.

Genus *Ebolavirus* of the family *Filoviridae* contains four viruses known to cause disease in humans (Ebola virus, Sudan virus, Bundibugyo virus, Taï Forest virus) and one so far not associated with human disease (Reston virus). While outbreaks caused by Ebola and Sudan viruses have been recorded since 1976, Bundibugyo emerged as recently as 2007.
Drug discovery efforts against ebolaviruses have used Ebola as the representative virus for the genus. Ebola minigenome systems, virus-like particles and infectious reporter viruses have been used for primary screens, and hits of such screens have then typically been tested against some of the related viruses as well. We are interested in comparative screens using all known ebolaviruses as well as strains emerging during future outbreaks.
To this end, we have generated an Huh7-based cell line that stably expresses an Ebola minigenome with a fluorescent reporter gene, zsGreen. Importantly, we show that this minigenome can be activated by infection with any of the five ebolaviruses. Coupling wild-type virus infections at low multiplicity with activation of a quantifiable reporter signal enables comparative screens that are based on complete viral replication cycle. We are interested in using our system especially for screening compounds that target viral proteins, such as nucleoside analogs. Libraries of such compounds may contain hits against over-looked but medically important relatives of Ebola.

85. Mechanistic Analysis of Benzoannulene Alphavirus Inhibitors

Nicole Haese, PhD[1], *Kaleem Ahmed, PhD*[2], *Clayton Morrison, PhD*[3], *Wes Sanders, PhD*[3], *Nathaniel Moorman, PhD*[3], *Nicholas May, BS*[4], *Vibha Pathak, PhD*[2], *Corinne Augelli-Szafran, Ph.D*[2], *Mark Suto, PhD*[2], *Victor DeFilippis, PhD*[1], *Thomas Morrison, PhD*[4], *Mark Heise, PhD*[3], *Ashish Pathak, PhD*[2], *Daniel Streblow, PhD*[1]

[1]Vaccine & Gene Therapy Institute, Oregon Health & Science Univeristy; [2]Southern Research; [3]University of North Carolina at Chapel Hill; [4]University of Colorado School of Medicine

Chikungunya virus (CHIKV), is an Alphavirus that causes debilitating polyarthritis. In recent years the largest recorded outbreak of CHIKV occurred spreading from the Islands of the Indian Ocean and India to the Western hemisphere, causing millions of cases across more than 40 countries. Despite the global re-emergence, increased global spread of the virus, and its high morbidity rate there are no virus-specific treatments or vaccines available. To close this gap, we used a high-throughput screen as a primary assay to screen drug libraries and identify compounds that blocked alphavirus replication. We identified a family of Benzoannulenes capable of inhibiting CHIKV replication at micromolar concentrations with little to no cytotoxicity. We performed studies to understand the antiviral breadth and mechanism of action for this compound family. Time of addition studies revealed that the most potent analog, SRI-34963 (IC_{90}= 1.5 mM), requires administration prior to 4 hours post-infection in order to inhibit virus replication. Consistent with these results, RNA profiling experiments showed a decrease in viral RNA synthesis with SRI-34963 suggesting an early block in viral replication. Sequencing of resistant mutants generated against a Benzoannulene identified mutations in the nonstructural protein 3 (nsP3). Reverse genetic reintroduction of the mutations into wt CHIKV conferred the resistance phenotype against the Benzoannulene; also confirming nsP3 as the target for Benzoannulenes. This novel group of anti-CHIKV compounds offers a new potential treatment option for CHIKV infections.

86. High Throughput Screen Measuring Marburg Virus VP24 Activation of the Nrf2 Antioxidant Response

Megan Edwards, PhD, Christopher Basler, PhD
Institute for Biomedical Sciences, Georgia State University, Atlanta, Georgia, United States of America

Marburg virus (MARV), a member of the family *Filoviridae*, causes highly lethal hemorrhagic fever in humans. Despite the need, there are currently no approved antivirals or therapeutics. One potential avenue for therapeutic intervention is the interaction between MARV VP24 (mVP24) and the host protein Kelch-like ECH-associated protein 1 (Keap1). Keap1 is an E3 ubiquitin ligase specificity factor that targets nuclear factor (erythroid-derived 2)-like 2 (Nrf2) for degradation. This serves to negatively regulate the Nrf2 antioxidant response pathway under homeostatic conditions. However, oxidative stress and other stresses disrupt Keap1 degradation of Nrf2. This leads to nuclear translocation of Nrf2, where it binds to antioxidant response elements (AREs), upregulating transcription of cytoprotective genes. Previous studies demonstrate that mVP24 interaction with Keap1 disrupts the Keap1:Nrf2 interaction and upregulates ARE genes. Suggesting that this is important for MARV pathogenesis, Nrf2 knockout mice better control MARV infection as compared to wild-type animals. We have developed and optimized a high-throughput screening assay for mVP24 dependent activation of the Nrf2 antioxidant response that can used to identify inhibitors of this response. An ARE firefly luciferase reporter and mVP24 are stably expressed in 293T cells. Cells are plated into 384 well plates in the presence of compounds and 24 hours post-treatment the luciferase signal is read. The assay is robust, with a Z-factor >0.5. We have performed screening of more than 9500 compounds, identifying lead hits that inhibit the Nrf2 antioxidant response pathway, providing both potential anti-MARV therapeutics and furthering our understanding of the role of mVP24.

87. Probing Membrane Lysis of Individual Virus Particles Induced by an Amphipathic Peptide and Correlations with Antiviral Activity

Nam-Joon Cho, PhD, Joshua Jackman, PhD
School of Materials Science and Engineering, Nanyang Technological University, Singapore

Antiviral drugs that destabilize viral membranes hold significant promise for developing broad-spectrum antiviral strategies, and fulfilling this potential would be aided by a deeper mechanistic understanding of how membrane-lytic agents within this class operate. However, real-time observation of membrane-lytic activity against single, nanoscale virus particles remains elusive. Herein, we investigate the process of individual virus particle lysis by using a total internal reflection fluorescence microscopy approach that is optimized for tracking viral membrane degradation and capable of highly parallel measurements. An amphipathic, -helical peptide with reported antiviral activity was tested, and it was discovered that the peptide rapidly lyses synthetic lipid vesicles in two stages, whereas a gradual, single-stage lytic process occurs against Dengue (DENV) and Zika (ZIKV) virus particles. The detailed steps of membrane lysis strongly depended on nanoscale curvature, sterol composition, and temperature-dependent conformational changes in virus structure. Collectively, our findings demonstrate a broadly applicable experimental approach to monitor lysis of individual virus particles, and highlight the potential clinical significance of viral membrane targeting.

88. A Novel Class of Replication Inhibitors of RSV N/P Interaction

Roberto Manganaro, MS[1], Dirk Jochmans, PhD[2], Johan Neyts, PhD[2], Pleter Leyssen, PhD[2], Andrea Brancale, PhD[1]
[1]*Cardiff School of Pharmacy and Pharmaceutical Sciences, Cardiff, United Kingdom;* [2]*Rega Institute – University of Leuven, Belgium*

Respiratory Syncytial Virus (RSV), part of to the *Paramyxoviridae* family, is an enveloped, negative-sense, single-stranded RNA virus and it is the principal etiological cause of LRTIs (Low Respiratory Tract Infections) worldwide, both in infants and elderly patient. Severe RSV infection in childhood seems to be involved also in long-term respiratory diseases in adulthood, such as asthma. Currently, there are no small-molecule antiviral on the market for the treatment or prevention of this viral infection.

The N protein, which is part of the ribonucleocapsid complex (RNP) and plays a key role in the transcription and replication of the viral RNA, was chosen as a target for the identification of novel potential inhibitors of the virus replication. The X-ray structure of the N protein co-crystallised with a known inhibitor of the N-NTD (N protein N-terminal domain) and P-CTD (P protein C-terminal domain) interaction was used for a structure-based virtual screening of commercially available drug-like compounds.

24 compounds were selected and bought on the basis of their ability to bind deeply inside the hydrophobic pocket and on the number of interactions. Among these compounds four hits were identified in virus-cell-based assay, showing a complete inhibition of the viral infection. The best of the four hits was chosen as a starting point for Structure-Activity relationship (SAR) analyses. A series of analogues were synthetized and they were evaluated for antiviral activity in a virus-cell-based assay. The results obtained will be discussed in this presentation.

89. A Novel Mutation in N1 Neuraminidase Confers Resistance to Multiple Neuraminidase Inhibitors Without Impacting Viral Fitness

Jin Jung Kwon, BS[1], Won-Suk Choi, MS[1], Ju Hwan Jeong, BS[1], Ji Won Han, BS[1], Su Jeong Ahn, BS[1], Hyeok-il Kwon, PhD[1], Eun-Ha Kim, PhD[1], Sun-Woo Yoon, PhD[2], Young Ki Choi, PhD[1], Yun Hee Baek, PhD[1], Min suk Song, PhD[1]

[1]College of Medicine and Medical Research Institute, Chungbuk National University, Cheongju, Korea, Republic of; [2]Viral Infectious Disease Research Center

The increased use of the neuraminidase inhibitor (NAI) Oseltamivir (OS) as a therapeutic intervention for influenza virus infection has selected for H275Y in N1 neuraminidase (NA), which confers resistance to OS. This has led to the increased use of the alternative NAI Zanamivir (ZA), which in turn has given rise to multi-NAI resistant viruses. To study molecular markers conferring multi-NAI resistance, the NA gene of OS-resistant pandemic H1N1 2009 influenza virus (pH1N1) was enriched with random mutations in addition to Y275 and the randomly mutated virus library was propagated under ZA. A substitution together with Y275 conferred highly increased resistance to the NAIs OS and Peramivir (PER) and increased resistance to the NAIs ZA and Laninamivir (LAN). The single mutation without Y275 alone also increased resistance to all NAIs, suggesting this was a multi-NAI resistance marker. The single mutation did not impact viral fitness or pathogenicity *in vitro* or in the murine model, whilst together with Y275 impacted viral fitness and pathogenicity. The single mutation alone also did not appreciably impact viral replication in the ferret upper respiratory tract or transmissibility. However, the double mutations containing Y275 abrogated both direct contact and aerosol transmission in the ferret model. Overall, these results highlight the potential for pH1N1 viruses to attain multi-NAI resistance and the importance of the novel mutation in NA as a marker for multi-NAI resistance.

90. Profiling of Neuraminidase Inhibitor Resistance among subtype N4, N5, N6 and N8 Avian Influenza Viruses

Won-Suk Choi, MS[1], jin Jung Kwon, BS[1], Ju Hwan Jeong, BS[1], Ji Won Han, BS[1], Su Jeong Ahn, BS[1], Su-Jin Park, BS[1], Young-il Kim, MS[1], Chul-Joong Kim, PhD[2], Young Ki Choi, PhD[1], Yun Hee Baek, PhD[1], Min-Suk Song, PhD[1]

[1]Chungbuk National University, Cheongju, Republic of Korea; [2]Chungnam National University, Dae Jeon, Republic of Korea

Increased rates of human infections with various subtypes of avian influenza viruses (AIVs) have recently been reported. This is of concern as current vaccines against seasonal influenza virus strains show limited efficacy against AIV infections. Further, widespread resistance to M2 ion channel blockers means that neuraminidase (NA) inhibitors (NAIs) are a critical therapeutic intervention against AIV infections. However, NAI resistance is also observed, as we found in N3, N7 and N9 AIV subtypes. Here, we profiled NA mutations that conferred NAI resistance in N4, N5, N6, and N8 AIV subtypes using Gene-segmented Random Mutagenesis. We generated libraries of mutant viruses using reverse genetics (RG) and resistant

phenotypes were selected using the NAIs Oseltamivir, Zanamivir or Peramivir in MDCK cells. NA activity, IC50, stability and infectivity of RG NAI resistant variants were determined. We identified 30 mutations in NA, 17 of which (N4 = 4, N5 = 5, N6 = 4, and N8 = 4) conferred the resistance to NAIs as measured by fluorescence-based NA inhibitory assay. Mutations conferring NAI resistance were mainly categorized as either novel, subtype-specific or previously reported in other subtypes. Knowledge of these NA mutations in AIVs is important in facilitating surveillance and monitoring of NAI resistance in AIVs.

91. Anti-Influenza Activity of Plant Flavonoids – Di- and Tetramethoxy-Quercetin Derivatives

D. Starosyla, PhD[1], M. Platonov, PhD[2], O. Vacylchenko, PhD[2], L. Palchykovska, PhD[2], S. Zagorodnya, PhD[3], S. Dsadiun, PhD[1], S. Rybalko, MD[1], L. Varbanets, PhD[3], V. Atamanyuk, PhD[1]
[1]Gromashevsky L.V. Institute of Epidemiology and Infection Diseases, NAMS of Ukraine, Kyiv, Ukraine; [2]Institute of Molecular Biology and Genetics, NAS of Ukraine, Kyiv, Ukraine; [3]D.K.Zabolotny Institute of Microbiology and Virology NAS of Ukraine, Kyiv, Ukraine

The chemical analysis of the preparations from the mixture extracts of *Deschampsiya caespitosa L* and *Calamagrostis epigeios L.*, was carried out anti-influenza activity of major and minor components of extracts mixture – 7,3′ –dimethoxy quercetin (DMQ) and 5,7,3,4′-tetramethoxy quercetin (TMQ), respectively, was defined. The monosaccharides ramnoza and glucose and small amounts of protein in DMQ and TMQ preparations were revealed, their impact on general activity of both drugs was evaluated as well. Antiviral activity of DMQ and TMQ in vitro and in vivo has been determined, an effective doses for prophylaxis and therapeutic regimens against influenza virus of this preparations amounted to 4.8 mcg/ml and 0.96 mcg/ml, accordingly. The anti-influenza virus mechanism of DMQ is explained dy neuraminidase activity inhibition that realized via the DMQ binding to active site of enzyme. The stability of enzyme-inhibitor complex is provided to by the carbohydrate moiety.

92. Delineate the Dimeric Flip-Flop Mechanism of Coronaviral Main Protease During Catalysis

Lin Shi, BS, Chi-Yuan Chou, PhD
Department of Life Sciences, National Yang-Ming University, Taipei, Taiwan

SARS and MERS are the deadly respiratory infectious diseases by two highly pathogenic human coronaviruses. Their main protease (M^{pro}) is essential to viral maturation involving the auto-cleavage of polyproteins and as a great target for development of antiviral-drugs. Previous studies suggest that dimeric M^{pro} may perform a flip-flop mechanism that only one of the subunit is catalytically active at any time during the catalysis. To clarify this, we try to express and purify heterodimeric M^{pro} (one subunit is wide type and another is inactive mutant, C145A). Interestingly, the K_{cat} of heterodimeric M^{pro} have 70 fold decrease than that of the wild-type enzyme. It indicates that the wild-type subunit is also inactive due to the tie with the inactive mutated subunit. To delineate how the inactive subunit transforms the active one into inactive, we try to solve the heterodimeric M^{pro} by X-ray crystallography. However, the heterodimeric M^{pro} originated from SARS-CoV cannot be clearly identified in the asymmetric unit in that there is only one residue's difference between the two subunits. To solve this, the "chimeric" heterodimeric M^{pro} whose one subunit (wide-type) is from SARS-CoV and another (inactive mutant) is from MERS-CoV have been expressed and purified. Surprisingly, the chimeric heterodimer also show dramatically k_{cat} decrease, indicating the existence of flip-flop mechanism. Now we are going to solve the chimeric M^{pro} to validate the possible conformational change during the flip-flop.

93. Multiple Effects of Toxins Isolated from *Crotalus durissus terrificus* on the Hepatitis C Virus Life Cycle

Jacqueline Shimizu, MS[1], Cintia Bittar, PhD[2], Mariana Batista, PhD[2], Guilherme Campos, MS[2], Suely Silva, BS[3], Adélia Cristina Silva, PhD[4], Suely Vilela, PhD[4], Victor Hugo Quintana, PhD[4], Paula Rahal, PhD[2], **Ana Carolina Jardim, PhD**[3]

[1]Institute of Biomedical Science, Federal University of Uberlandia; [2]Sao Paulo State University; [3]Federal University of Uberlandia; [4]University of Sao Paulo

Hepatitis C virus (HCV) is one of the main causes of liver disease and transplantation worldwide. Current therapy is expensive, presents additional side effects and viral resistance has been described. Therefore, studies for developing more efficient antivirals against HCV are needed. Compounds isolated from animal venoms have shown antiviral activity against some viruses such as Dengue virus, Yellow fever virus and Measles virus. In this study, we evaluated the effect of the complex crotoxin (CX) and its subunits crotapotin (CP) and phospholipase A_2 (PLA_2-CB) isolated from the venom of Crotalus durissus terrificus on HCV life cycle. Huh 7.5 cells were infected with HCVcc JFH-1 strain in the presence or absence of these toxins and virus was titrated by focus formation units assay or by qPCR. Toxins were added to the cells at different time points depending on the stage of virus life cycle to be evaluated. The results showed that treatment with PLA_2-CB inhibited HCV entry and replication but no effect on HCV release was observed. CX reduced virus entry and release but not replication. By treating cells with CP, an antiviral effect was observed on HCV release, the only stage inhibited by this compound. Our data demonstrated the multiple antiviral effects of toxins from animal venoms on HCV life cycle.

94. A High Content Screen Identifies Cellular microRNAs with Anti-Flavivirus Activity

Jessica Smith, PhD, Ashleigh Murphy, BS, **Alec Hirsch, PhD**

Vaccine and Gene Therapy Institute, Oregon Health & Science University, Beaverton, Oregon, United States of America

Many critical aspects of the biology of mosquito-borne flaviviruses, including virus-host interactions, host cell requirements for replication, and how virus-host interactions impact pathology, remain to be fully understood. Our research is focused on identifying cellular microRNAs (miRNAs) that modulate flavivirus replication as a means of characterizing cellular pathways that support or limit viral replication. We have screened a library of known human miRNA mimics for their effect on replication of three flaviviruses—DENV, WNV, and Japanese encephalitis virus (JEV)—using a high content immunofluorescence screen. Members of the miR-34 family demonstrated strong anti-flaviviral effects, and this inhibitory activity extended to other viruses, including ZIKV, alphaviruses, and herpesviruses. These miRNAs potentiate IRF3 phosphorylation and translocation to the nucleus, induction of IFN-responsive genes, and release of type I IFN from transfected cells. We further demonstrate that the intersection that miR-34 acts through repression of the Wnt signaling pathway, resulting in increased GSK3b-TBK1 binding, inducing TBK1 to phosphorylate IRF3 and initiate downstream IFN signaling. In a second study, miR-424, another inhibitor of flavivirus replication, was found to target the E3 ubiquitin ligase SIAH1, which is induced during DENV infection as part of the unfolded protein response. During infection, SIAH1 positively contributes to DENV replication via targeting the innate immune adaptor protein MyD88 for degradation, and miRNA or siRNA-mediated inhibition of SIAH1 expression in infected cells results in a significant reduction in viral replication. These miRNAs, or the targets identified by this approach, may inform development of novel anti-flaviviral therapeutic approaches.

95. Broad-Spectrum Antiviral Molecules with Biophysical Mechanisms of Action

Sietske Speerstra, BS[1], Alexey Chistov, BS[2], Gleb Proskurin, BS[2], Vladimir Korshun, PhD[2], Luis Schang, DVM, PhD[3]

[1]Department of Biochemistry, University of Alberta, Edmonton, Alberta, Canada; [2]Shemyakin-Ovchinnikov Institute of Bioorganic Chemistry, Moscow, Russian Federation; [3]Baker Institute, Cornell University & Dept. of Biochemistry, University of Alberta, Ithaca, New York, United States of America

Most antivirals target virus-specific proteins. Antivirals with conserved targets can be broadly active. Lipid envelopes, which fuse with cellular membranes, are common to most pathogenic viruses. Envelope fusion occurs through an hemifusion stalk in which only the outer leaflets are fused, bent with a smaller radius for their polar heads than for their hydrophobic tails (negative curvature). Outer leaflets enriched in "inverted cone" phospholipids (head groups of larger cross sections than those of their lipid tails) disfavor negative curvature. The RAFIs (rigid amphipathic fusion inhibitors), synthetic compounds of inverted cone molecular geometry, inhibit infectivity of enveloped viruses. The original RAFIs have ethynyl-perylene hydrophobic and arabino-uracil polar moieties (EC_{50} 16–43 nM). Their activities require inverted cone shape, amphipaticity, and rigidity of the hydrophobic moiety. We now tested 26 chemically distinct new RAFIs.

The ethynyl linker could be replaced by butadiynyl (EC_{50} 32 nM), or perylene-ethynyl by butadiynyl-pyrene (EC_{50}, 73 nM), without affecting antiviral activity. The sugar could be modified (EC_{50}, ~50 nM) or replaced by carboxymethyl or carboxamidomethyl residues (EC_{50} 29–60 nM). Uracil to cytidine replacement increased EC_{50} (1,260 nM) as did triazoles substitutions for ethynyl, shortening the hydrophobic moiety to 9.2 Å (≤2,300 nM). Phenyl replacement for perylene, shortening it to 5.3 Å, disrupted activity (EC_{50} >2,000 nM). Most RAFIs were not cytotoxic (CC_{50} >2,000 nM; SI >250–1200). Carbonyl or butylamide substitutions for arabino, or cytidine replacement for uracil, increased cytotoxicity (CC_{50} <600–1,700 nM). The definition of the RAFIs molecular requirements allows for their further development.

96. Identification of a Substituted Thienopyrimidine Scaffold with Antiviral Activity against Zika Virus.

Marcella Bassetto, PhD[1], Juliane Nolte, MS[2], Benno Schreiner, MS[2], Joachim Bugert, MD, PhD[2], Andrea Brancale, PhD[1]

[1]Cardiff University; [2]Institut für Mikrobiologie der Bundeswehr, München

Zika virus (ZIKV), a *Flavivirus* transmitted to humans by mosquitoes of the *Aedes* species, represents an ongoing public health emergency of international concern. Responsible for an acute febrile illness with rash, arthralgia and conjunctivitis, infection with this virus is linked to Guillan-Barré syndrome in adults, and microcephaly and neurological disorders in newborns to ZIKV-infected mothers. Despite the serious consequences associated with the infection, no therapeutic options are currently available to treat or prevent the disease.

In an attempt to identify potential anti-ZIKV agents, we have performed an *in silico* screening of our internal library of small-molecule compounds. A series of docking simulations were carried out against the available crystal structures for ZIKV non-structural proteins, and the top hits were evaluated for their antiviral activity in cell-based assays. A substituted thienopyrimidine scaffold, which we had previously reported as potent inhibitor of HCV replication in cells, was found to inhibit ZIKV replication in a $TCID_{50}$ assay. Different derivatives showed IC_{50} values in the low micromolar range, in both a neuronal (DBTRG) and a lung (A549) cell line. Starting from these early findings, we have designed a series of novel analogues, modifying different portions of the original scaffold. The synthetic strategies optimised for the preparation of these new small molecules will be discussed, along with their antiviral activities against ZIKV replication in cellular systems.

97. Membrane-Permeable Nucleoside Triphosphate-Prodrugs against Hepatitis C Virus

Matthias Winkler, MS[1], Elena Vedove, MS[2], Chris Meier, PhD[1]
[1]*University of Hamburg, Hamburg, Germany;* [2]*University of Camerino, Camerino, Italy*

The antiviral activity of nucleosides is often limited due to ineffective phosphorylation. To overcome this limitation, a wide variety of different prodrug concepts has been developed in the past years. Herein, we report the first synthesis of membrane-permeable triphosphate-prodrugs of well-known anti-HCV nucleosides and 2´-*C*-methyl-T-1106 using the Tri*PPP*ro-approach.

The Tri*PPP*ro-approach, first applied to deliver anti-HIV-NTPs into cells, was used to synthesize a first generation of potential membrane-permeable-Tri*PPP*ro-anti-HCV-NTPs. Utilizing this approach, the γ-phosphate was masked with two highly lipophilic and cleavable units, which are enzymatically hydrolyzed inside the cell to release the NTP.

The synthesis of these novel Tri*PPP*ro-compounds was carried out using either the phosphoramidite or the H-phosphonate route. However, it was found that the H-phosphonate route is more beneficial, due to the easier access of the required NMPs. First, the masked pyrophosphate 6 was synthesized from the readily available H-phosphonate 5. The nucleoside monophosphates 7a-d were synthesized in a one-step procedure based on a known protocol. The synthesis of the Tri*PPP*ro-compounds 8a-d was achieved by a stepwise activation of 6 with trifluoroacetic acid anhydride and *N*-methylimidazole, followed by the addition of the NMPs 7a-d (Scheme 1).

We successful applied the Tri*PPP*ro-approach to three anti-HCV nucleosides and the 2´-*C*-methyl-T-1106-analogue using the H-phosphonate route. Hydrolysis data proved the formation of the corresponding triphosphates in cell extracts and under chemical conditions. Evaluation of the antiviral data will be shown as well.

98. Evaluation of Lamivudine Sustained Release Tablet for Hepatitis B Infection

Lila Nath, PhD[1]
[1]*Department of Pharmaceutical Sciences, Dibrugarh University, Dibrugarh, Dibrugarh, India*

Hepatitis B infection remains a relevant clinical problem with high morbidity and mortality in long-term follow up in renal,liver or lung transplant recipients. Lamivudine is approved for clinical use in the treatment of Hepatitis B and AIDS either alone or in combination with another antiviral drugs. Owing to its water solubility and shorter half-life, it requires frequent dosing by oral route. Various recent techniques for sustaining drug release have been explored, and matrix system offer various advantages of ease of formulation better control on release profile of drug and better patient compliance. This study concerns with the development of sustained release tablets of lamivudine using cross-linked moth bean starch. The cross-linked moth bean starch was synthesized with phosphorous Oxy chloride in the basic pH medium. The cross-linked moth bean starch was tested for acute toxicity and drug-excipient compatibility study. The formulation was evaluated for physical characteristics like hardness, friability, % drug content and weight variations. The *in vitro* release study showed that the optimized formulation exhibited highest correlation (R) value with zero order kinetic model and the release mechanism study proved a combination of diffusion and erosion process. There was a significant difference in the pharmacokinetic parameters (T_{max}, C_{max}, AUC, $T_{1/2}$ and MDT) of the optimized formulation as compared to the marketed conventional tablet Lamivir® in rabbit model. The *in vitro* and *in vivo* results of the optimized formulation revealed sustained release nature of this novel formulation that will augment patient compliance and cost effectiveness in patients with hepatitis B.

99. The STING Agonist SB 11285 is a Broad-Spectrum Antiviral Agent

Cybele Garcia, PhD[1], José Peña Carcamo, PhD[1], Maria Morell, MS[1], Sandra Cordo, PhD[1], Sreerupa Challa, PhD[2], Shenghua Zhou, PhD[2], Anjaneyulu Sheri, PhD[2], Seetharamaiyer Padmanabhan, PhD[2], Geeta Meher, PhD[2], Diane Schmidt, PhD[2], Niraj Shil, PhD[3], Meleri Jones, PhD[3], Graham Foster, PhD[3], Santanu Bose, PhD[3], Nezam Afdhal, PhD[2], Brent Korba, PhD[5], Radhakrishnan Iyer, PhD[2]

[1]Lab Estrategias Antivirales, Bioquímica y Biología del virus Junín, Univ Buenos Aires, Buenos Aires, Argentina; [2]Spring Bank Pharmaceuticals, Inc., Milford, MA 01757, Massachusetts, United States of America; [3]Washington State University, Pullman, Washington, United States of America; [4]The Liver Unit, Blizard Institute, Barts Health, Queen Mary University of London, London, United Kingdom; [5]Georgetown University Medical Center, Division of Molecular Virology and Immunology, Washington DC, District of Columbia, United States of America

BACKGROUND: Cyclic guanylate adenylate (cGAS), an important pattern recognition receptor (PRR) for viral genomes, uses Stimulator of Interferon Genes (STING) as the key adapter protein in the IRF3-IFN signaling axis to trigger innate and adaptive immune response for antiviral defense. We report here the antiviral evaluation of a novel nucleotide compound SB 11285, a potent STING agonist that is being developed for immuno-oncology.

METHODS: In vitro antiviral evaluations of SB 11285 were conducted as follows: **RSV**: We used RSVA2-infected (0.5 MOI) A549 cells and viral titer was estimated by plaque assays; **Norovirus**: A Replicon of Norovirus strain GI NoV in HG23 (hepatoma) cell line was used and activity assessed by RNA hybridization and quantitative PCR; **HCV**: Activity against HCV genotypes 1a and1b was tested using the capture fusion assay. Briefly, THP-1 cells were exposed to donor serum, fused with Huh7 derivative cells and qPCR was used to assess HCV replication. **Hemorrhagic fever viruses**: Activity against JUNV and Dengue-2 was conducted using strain JV 4454 and DENV-2 (strain NGC) respectively in Vero cells and extracellular DENV/JUNV yields were determined by plaque assays. Cytotoxicity assays were done in parallel by MTT or MTS methods.

RESULTS: SB 11285 elicited potent antiviral activity against all tested RNA viruses with EC_{50} ranging from 0.34 to 5.5 µM, and with high selectivity index.

CONCLUSION: Consistent with its mechanism of action, the STING agonist SB 11285 showed potent antiviral activity against several RNA viruses including hemorrhagic fever viruses. Additional preclinical studies are under way.

100. Advances in Mouse Models of Crimean-Congo Hemorrhagic Fever: Viral Tropism and Neurological Disease in Hu-NSG™-SGM3 Humanized Mice

Jessica Spengler, DVM, PhD[1], M. Kelly Keating, DVM[1], Anita McElroy, MD, PhD[1], Marko Zivcec, PhD[1], JoAnn Coleman-McCray, BS[1], Jessica Harmon, MS[1], Brigid Bollweg, MS[1], Cynthia Goldsmith, MS[1], Eric Bergeron, PhD[1], James Keck, PhD[2], Sherif Zaki, MD[1], Stuart Nichol, PhD[1], Christina Spiropoulou, PhD[1]

[1]Centers for Disease Control and Prevention, Atlanta, Georgia, United States of America; [2]The Jackson Laboratory, Sacramento, California

Crimean-Congo hemorrhagic fever (CCHF) is a tick-borne viral disease seen exclusively in humans. Existing mouse models of disease are limited to severely immunocompromised strains that develop a rapid and uniformly lethal disease. We infected SGM3 humanized mice (Hu-NSG™-SGM3) with 2 strains of CCHFV isolated from human patients in Oman (CCHFV-OM) or Turkey (CCHFV-TR). All mice infected with CCHFV-TR developed progressive disease and had high levels of virus in the brain. Mice infected with CCHFV-OM had mild clinical signs and recovered, and no viral antigen was detected in the brain. Although overt hemorrhagic signs are often described in severe disease, central nervous system (CNS) infection and neurological involvement have been also reported. To date, mechanisms of virus entry in the brain, targets of infection, and neuropathology have not been investigated. In mice that succumbed to disease, viral antigen was detected in rare neurons, but was primarily found in glial cells, including microglia and astrocytes, such as Bergmann glia, and in meninges, resulting in gliosis, meningitis, and meningoencephalitis. Here we present a novel animal model of CCHF, identify cell populations and regions in the brain for future investigation, and highlight considerations for therapeutic treatment of CCHF.

101. Comparison Between Various Strains of Chikungunya in Disease Phenotype and Response to Antiviral Treatment

Makda Gebre, BS, Justin Julander, PhD
[1]Institute for Antiviral Research, Utah State University, Logan, Utah, United States of America

Antiviral countermeasures are needed to prevent or treat Chikungunya. First discovered in 1953 in Tanzania, Chikungunya virus (CHIKV) recently reemerged in 2004 extensively spreading in Africa, Asia, Europe and the Americas and causing outbreaks resulting in over 3.3 million cases. Different strains currently distributed around the world have been grouped into three phylogenetic groups, namely, West African, Asian and East/Central/South African (ECSA). In order to identify broadly active agents against all clades of CHIKV, we investigated two strains each from the three phylogenetic groups. To understand differences in phenotypic consequences and antiviral response, the viruses were characterized in two mouse models, an arthralgic DBA/1J mouse model and a lethal AG129 mouse model. Subsequently, the differential efficacy of T-705, a broad-spectrum pyrazine derivative antiviral, was evaluated in both mouse models.

102. Antiviral Treatment Efficiently Inhibits Chikungunya Virus Replication in the Joints of Mice During the Acute but not During the Chronic Phase of Infection

Rana Abdelnabi, MS[1], Dirk Jochmans, PhD[1], Erik Verbeken, PhD[2], **Leen Delang, PhD**[1], Johan Neyts, PhD[1]
[1]Rega Institute for Medical Research, KU Leuven, Leuven, Belgium; [2]Department of Pathology, University of Leuven and Leuven University Hospitals, Leuven, Belgium

We reported earlier that favipiravir (T-705), a broad-spectrum antiviral, inhibits the replication of chikungunya virus (CHIKV) *in vitro* and protects against disease progression in CHIKV-infected immune-deficient mice. We here explored whether favipiravir is also effective in a CHIKV-induced arthritis mouse model during the acute and the chronic phase of the infection. C57BL/6J mice were infected with CHIKV in the left footpad and were treated with favipiravir (300 mg/kg/day, orally) either during the acute phase (from day zero to day 3 post-infection (p.i.)) or during the chronic phase of infection (from day 49 to 55 p.i.). Favipiravir treatment during the acute phase resulted in complete inhibition of systemic viral spread. In contrast, when favipiravir treatment was initiated late, no significant differences in viral RNA levels were noted between the treated and untreated mice. Moreover, viral RNA was still present in the ankles of infected mice up to 14 weeks p.i.; however, no infectious virus could be detected. Interestingly, when attempting to amplify the viral genome from these samples by PCR, some parts of the genome, such as the viral polymerase gene, could not be amplified and the amplicons for the non-structural protein 1 (nsP1) and the envelope glycoprotein (E2) were shorter than expected. Collectively, these results suggest that the viral RNA detected during the chronic phase is likely defective, which also explains the lack of effect of a viral replication inhibitor. It will have to be explored whether a same situation would hold for CHIKV infections in man.

103. Influenza A Inhibitors Based on Copper Binding to a Highly Conserved Histidine

Nathan Gordon, MS[1], Kelly McGuire, BS[2], Spencer Wallentine, BS[2], Gregory Mohl, BS[2], **Mackenzie Hart, BS**[2], Jonathan Lynch, BS[2], Roger Harrison, PhD[2], David Busath, MD[2]
[1]Dept. of PDBio, Provo, Utah, United States of America; [2]Brigham Young University, Provo, Utah, United States of America

Influenza M2 proton channel mutations in the primary target site for amantadine have caused desensitization to anti-M2 drugs. New M2 blockers effective against the ubiquitous S31N M2 mutation are needed. Divalent copper has previously been shown to block wild type M2 channels in transfected Xenopus laevis oocytes and binds near the His37 imidazoles according to solid state NMR. We have synthesized, purified and characterized by NMR, ICP-MS, and UV-vis a set of four copper complexes based on known M2 blockers amantadine and cyclooctylamine, two iminodiacetates and two iminodiacetamides, as well as substituent molecules, for controls and structure-activity relationships. In two-electrode voltage clamped Xenopus laevis oocytes transfected with Influenza A M2 S31 or N31, the compounds show thorough, poorly-

reversible block of acid-induced inward currents. When compounds lack Cu^{2+} or M2 channels lack imidazole side chains in the histidine tetrad (i.e. H37A mutations), block was incomplete and rapidly reversible. For example, with Cu(amantadine-iminodiacetate), which was stable at pH>4 and in the buffers tested, the EC_{50} for *in vitro* mini-plaque assays with amantadine-resistant A/Calif/07/2009 H1N1 was 0.7 ± 0.1 μM and the CC_{50} was 147 μM, in contrast to $CuCl_2$, which had EC_{50} of 3.8 ± 0.9 μM and CC_{50} of 19 μM. Given the stability and affinity of the compounds and the high level of conservation of M2 H37 in nature, these compounds show promise for further testing as inhalant or injectable drugs against all strains of influenza A.

104. High-Throughput Screening to Identify Dengue Virus Entry Inhibitors

Wenlong Lian, PhD, Priscilla Yang, PhD
MBIB, Harvard Medical School, Boston, Massachusetts, United States of America

Dengue virus (DV) is a major human pathogen infecting 300 million people annually. Current approved countermeasures are limited to a single vaccine, Dengvaxia, which can reduce illness and hospitalizations in dengue endemic regions but potentially exacerbate disease in areas of low dengue prevalence. Alternatives to combat DV infection and disease are thus urgently needed.

The DV E protein on the virion surface presents a novel target for direct-acting antivirals that act at the earliest stage of the viral life cycle and thus mimic the humoral immune system. We previously identified compound GNF-2 and related disubstituted pyrimidines as inhibitors of DV that bind directly to E on the virion surface and inhibit viral entry. In our broader efforts to investigate inhibitors of E as potential antivirals, we sought to identify additional compound classes that can inhibit DV by this mechanism.

We established a high-throughput competitive AlphaScreen (amplified luminescent proximity homogeneous assay) utilizing a biotinylated variant of GNF-2. Via high-throughput screen with this assay, we identified 8 new chemical leads that inhibit DV in cell culture, with excellent correlation of activity in the AlphaScreen with antiviral potency. Since prior efforts to target DV E have relied on in silico and phenotypic screens, the assay and proof-of-concept assay we report provide important tools to discover inhibitors of E, to define the SAR for antiviral activity mediated by this target, and ultimately to develop small molecule inhibitors of DV entry as potential anti-DV therapeutics.

105. Role of RSV Polymerase in the Antiviral Effect of Ribavirin

Jerome Deval, PhD, Amy Fung, MS, Jia Meng, PhD, Andreas Jekle, PhD, Guangyi Wang, PhD, Natalia Dyatkina, PhD, Marija Prhavc, PhD, Julian Symons, PhD, Leo Beigelman, PhD
Alios BioPharma

Ribavirin is the only small molecule approved for the treatment of severe respiratory syncytial virus (RSV) infection in infants, and its mechanism of action is not well understood. It has recently been shown that ribavirin treatment of RSV-infected cells increases the number of virus mutations. It is conceivable that the mutagenic effect of ribavirin is caused by incorporation of ribavirin-MP into viral RNA by RSV polymerase. However, the interaction between ribavirin triphosphate (RBV-TP) and RSV polymerase has not been reported. Here, we conducted a comparative study between ribavirin and ALS-8112, a cytidine analog known to inhibit RSV. In cell-based assays, the EC_{50} value of ribavirin and ALS-8112 was 4.3 and 0.15 μM, respectively. In a biochemical assay, RBV-TP was about 10,000-fold less potent than ALS-8112-TP at inhibiting the polymerase activity of crude RSV ribonucleoprotein. This difference in antiviral potency at the triphosphate level came from poor RBV-TP substrate recognition by RSV polymerase, with a discrimination of >1,000-fold compared to GTP (opposite C template) and ATP (opposite U template). This level of discrimination is comparable to the G:U mismatch measured for RSV polymerase. These results indicate that RBV-TP is overall a poor substrate for RSV polymerase. The apparent discrepancy between the 30-fold and 10,000-fold lower potency in cell-based and biochemical assays most likely involves additional host targets. In conclusion, the mutagenicity of ribavirin against RSV is likely to be mediated by a decrease of intracellular nucleotide pools that indirectly increases base-pair mismatch by the error-prone RSV polymerase.

106. MERS-CoV Infection of Human Monocyte-Derived Cells and Antiviral Efficacy of Select FDA-Approved Drugs

Yu Cong, MD[1], Brit Hart, MS[2], Robin Gross, BS[1], Huanying Zhou, BS[3], Lisa Hensley, PhD[4], Jahrling Peter, PhD[5], Julie Dyall, PhD[6], Michael Holbrook, PhD[1]
[1]NIAID DCR IRF Battelle, Frederick, Maryland, United States of America; [2]APHL – Association of Public Health Laboratories, Gaithersburg, Maryland, United States of America; [3]NIAID DCR IRF LBERI, Frederick, Maryland, United States of America; [4]NIH NIAID DCR IRF, Frederick, Maryland, United States of America; [5]NIAID DCR IRF, Frederick, Maryland, United States of America; [6]NIAID DCR IRF Tunell, Frederick, Maryland, United States of America

Middle East respiratory syndrome coronavirus (MERS-COV) presents an emerging threat to public health worldwide by causing severe respiratory diseases in humans with high virulence and case fatality rate (about 30%). Little is known about the innate antiviral response in primary human monocytes-derived macrophages (MDM) and DCs (MDDCs) upon MERS-CoV infection. In this study we assessed MERS-CoV replication and the induction of inflammatory cytokines/ chemokines in MDMs and MDDCs and the efficacy of two drug compounds that have shown effective against MERS-CoV infection in immortalized cells. Both MDM and MDDC are permissive to MERS-CoV infection which triggers the production of several proinflammatory mediators. We have previously shown that chloroquine (CQDP) and chlorpromazine (CHLOR) have anti-MERS-CoV activity in Vero E6 cells. In order to determine the efficacy of these compounds in primary human antigen presenting cells, cells were treated with the test compounds and subsequently infected with MERS-CoV. In these studies, very little (CQ) or limited efficacy (CHLOR) was detected with higher cytotoxicity (CHLOR) than in Vero E6 cells. Therefore, more research effort evaluating viral pathogenesis and the mechanism of innate immune regulation following MERS-CoV infection is necessary in order to improve the control and treatment of MERS-CoV infection. These results also provide valuable evidence that antiviral drug screen studies should be evaluated in primary cells before moving to preclinical and clinical studies for MERS treatment.

107. Identification of Small Molecule Inhibitors of Ebola Virus Replication

Priya Luthra, PhD[1], Jue Liang, PhD[2], Colette Pietzch, MS[3], Sudip Khadka, PhD[1], Sampriti De, MS[1], Alexander Bukreyev, PhD[3], Joseph Ready, PhD[2], Christopher Basler, PhD[1]
[1]Georgia State University; [2]UT Southwestern; [3]University of Texas Medical Branch

Ebola viruses are enveloped, negative-sense, single-stranded RNA viruses which are causative agent of highly lethal hemorrhagic fevers in humans. The 2014-2016 Ebola outbreak in West Africa was of unprecedented scope, causing over 10,000 deaths. This emphasizes the dire need for effective drugs and vaccines to combat this virus. Here, we utilized our previously established high throughput screening Ebola virus minigenome assay that assesses the function of virus polymerase complex and can be used to identify small molecule inhibitors of Ebola virus RNA synthesis. We screened 200,000 compounds, identifying top 56 hits with more than 70% inhibition of Ebola minigenome activity and less than 20% cell toxicity. We further identified five top chemical scaffold that had selective index values > 10, suggesting the specificity against EBOV RNA synthesis. Here, we discuss the optimization of one of the lead compound classes – benzoquinoline as inhibitors of Ebola virus infection. To identify chemical features of the benzoquinoline that accounts for its antiviral activity, a broad set of about 50 structural analogs were synthesized and were tested in the minigenome assay to compare Structure-Activity Relationship trends. This led to identification of the optimized compound with submicromolar 50% inhibitory concentration in the minigenome assay. Furthermore, this compound inhibited the replication of infectious Ebola virus in vitro. Interestingly, this compound was also effective against other RNA viruses including vesicular stomatitis virus and Zika virus, suggesting broad spectrum antiviral activity of this compound. Our data demonstrate potential of benzoquinolines to be developed as broad spectrum antiviral therapeutics.

108. Development of an In Ovo System for Evaluation of Antivirals Against Zika Virus

Jasper Chan, MD, Kwok-Hung Chan, PhD, Shuofeng Yuan, PhD, Kenn Chik, BS, Zheng Zhu, MS, Kah-Meng Tee, BS, Jessica Tsang, BS, Cyril Yip, PhD, Vincent Poon, MS, Chris Chan, MS, Winger Mak, BS, Anna Zhang, PhD, Kwok-Yung Yuen, MD
Department of Microbiology, The University of Hong Kong, Hong Kong

Zika virus (ZIKV) infection may be associated with congenital malformations in infected fetuses and severe complications in infected adults. Rapid and reproducible viral culture systems are needed for evaluation of antivirals against ZIKV. Viral culture using chick embryos has been shown to be suitable for antiviral evaluation for other viruses because of the eggs' large size, low cost, and potentially higher throughput than other ranimal models. We investigated whether chick embryos could support virus replication and facilitate antiviral evaluation for ZIKV. Inoculation of chick embryos with 0.2 ml (3.6 log plaque forming units/ml) of either epidemic (ZIKV-Puerto Rico: ZIKV-PR, human isolate) or pre-epidemic ZIKV (ZIKV-Uganda: ZIKV-U, primate isolate) strains led to significantly higher mean viral loads in the brain (1.6-2.4 log10 copies/ml), muscle (1.2-2.4 log10 copies/ml), and viscera (0.3-2.1 log10 copies/ml) of the chick embryos ($P \leq 0.018$). No significant increase in mean viral load was observed in the egg yolk. Abundant ZIKV NS1 protein expression was seen in the brain, muscle, and viscera of ZIKV-inoculated chick embryos. Embryonic lethality was observed in all the ZIKV-inoculated chick embryos. Compared to untreated controls, addition of azithromycin led to a ≤0.88 log10 copies/ml reduction of mean viral load in ZIKV-inoculated chick embryos. These results corroborated with previous findings that showed the moderate *in vitro* anti-ZIKV activity of azithromycin in cell lines. Further optimization of this *in ovo* culture system would facilitate the development of therapeutic measures against the emerging ZIKV.

109. Successful Design of Ribonucleoside Di- and Triphosphate Prodrugs to Improve the Anti-Influenza Virus Activity of T-705 and its Analogue T-1105

Evelien Vanderlinden, PhD[1], Johanna Huchting, PhD[2], Chris Meier, PhD[2], Lieve Naesens, PhD[1]
[1]Rega Institute for Medical Research, Leuven, Belgium; [2]Institute of Organic Chemistry, Hamburg University, Hamburg, Germany

The nucleobase analogue T-705 (6-fluoro-3-hydroxy-2-pyrazinecarboxamide; favipiravir) is a unique antiviral drug possessing broad anti-RNA virus activity and a high barrier for resistance. When we compared the influenza virus inhibition by T-705 and its non-fluorinated analogue T-1105, the latter proved to be five-fold more potent in Madin-Darby canine kidney (MDCK) cells. In an enzymatic RNA elongation assay with influenza virus-derived viral ribonucleoproteins, T-1105 ribosyl-5'-triphosphate (RTP) was even seven-fold superior to T-705 RTP (IC_{50} values: 0.41 μM vs. 2.7 μM) in inhibiting GTP incorporation into viral RNA.

We previously reported[1] that human hypoxanthine guanine phosphoribosyltransferase (HGPRT) is crucial to convert T-705 and T-1105 into their ribosyl-5'-monophosphates which are then further phosphorylated to the active RTP metabolites. Since both pyrazine derivatives are poor HGPRT substrates, we applied our Di*PP*ro and Tri*PPP*ro prodrug approaches to increase the intracellular RTP levels.[2,3] In influenza A and B infected-MDCK cells, JH580 and JH642 (both Di*PP*ro-T-1105-RDPs), JH625 (Tri*PPP*ro-T-1105-RTP) and T-1105 displayed average antiviral EC_{50} values of 0.83, 0.98, 2.8 and 3.8 μM respectively, meaning that the best prodrug (JH580) afforded five-fold gain in antiviral potency. In contrast to T-705 and T-1105, the three prodrugs retained full antiviral activity in HGPRT-deficient MDCK cells, indicating that they release a phosphoribosylated metabolite inside the cells.

1) Naesens et al., *Mol Pharmacol* 2013, *84*, 615-629; 2) Meier et al., *Curr. Med. Chem.* 2015, *22*, 3933-3950; 3) Gollnest et al., *Angew. Chem, Int. Ed.* 2016, *55*, 5255-5258.

110. Treatment of Old and New World Arenavirus Infections with Favipiravir

Brian Gowen, PhD[1], Jonna Westover, PhD[1], Eric Sefing, MS[1], Brady Hickerson, BS[1], Kevin Bailey, BS[1], Luci Wandersee, BS[1], Brittney Downs, BS[1], Skot Nielson, MS[1], Yousuke Furuta, PhD[2]
[1]Utah State University; [2]Toyama Chemical Co., Ltd.

A collection of Old and New World arenaviruses are etiologic agents of viral hemorrhagic fever, a syndrome that features hematologic abnormalities, vascular leak, hypovolemia, and multi-organ failure. Treatment is limited to ribavirin for Lassa fever and immune plasma for Argentine hemorrhagic fever. Improved therapeutic options that are safer and are more effective and accessible are needed. In the present work, we describe continued efforts towards the development of favipiravir as a pan-arenavirus antiviral drug. We show that modification of treatment to include a high-dose loading period achieves complete protection in the guinea pig model of Junin virus infection when therapy is initiated two days following challenge. This modified loading dose strategy also protected 50% of animals from lethal disease when treatment was delayed until 5 days post-infection and extended the survival time in those that succumbed. Consistent with the survival data, dramatic reductions in serum and tissue viral loads were observed in the animals treated with favipiravir. Our efforts also extend to lymphocytic choriomeningitis virus (LCMV), an opportunistic pathogen of the immune compromised and a virus closely related to Lassa fever virus. A study to evaluate favipiravir in a model of hemorrhagic disease in NZB mice challenged with LCMV is underway and the results will be presented. *This work was supported by the National Institutes of Health (HHSN272201000039I).*

111. Vaccinating Effect of Attenuated Zika Virus Candidates in a Lethal Mouse Model

Justin Julander, PhD[1], Steffen Mueller, PhD[2], Skot Neilson, MS[1], J. Robert Coleman, PhD[2]
[1]Institute for Antiviral Research, Utah State University, Logan, Utah, United States of America; [2]Codagenix Inc., Farmingdale, New York, United States of America

Effective vaccines are needed to stem the tide of emergent Zika virus (ZIKV), which has caused significant amounts of morbidity and mortality in recent years. Various synthetic, codon-pair deoptimized ZIKV viral variants were generated and tested as live-attenuated vaccines for efficacy in a mouse model of lethal ZIKV disease. Synthetic wild-type (WT) viruses were also made for use as infection controls, and challenge of AG129 mice with these strains resulted in morbidity and mortality similar to what one would expect after infection with WT ZIKV, including detectable viremia, weight loss and mortality. The objective of the study was to determine the suitability of codon-pair deoptimized, attenuated ZIKVs as vaccines. Groups of AG129 mice were injected subcutaneously with the attenuated viruses followed by a boost 28 days after the initial challenge. Serum samples were collected prior to boost and just before lethal challenge with ZIKV. Various levels of protection were observed with different constructs; complete protection, however, was observed after a prime-boost protocol when vaccinationg with $10^{4.0}$ PFU of candidates CDX-ZKV-1 and CDX-ZKV-2. Protection correlated with neutralizing Ab levels present in the serum prior to virus challenge. The effective constructs had only a portion of their genome codon-pair deoptimized. This study demonstrates the potential utility of reverse engineered synthetic attenuated Zika viruses in the protection from severe disease. [Supported in part by HHSN272201000039I from the Virology Branch, NIAID, NIH]

112. Investigation of Stem Cell-Derived Alveolar like Macrophages as a Novel RSV Therapeutic

Yuchen Cen, BS[1], Michael Litvack, PhD[2], Wenming Duan, PhD[2], Martin Post, PhD[1], Theo Moraes, MD, PhD[1]

[1]University of Toronto, The Hospital for Sick Children, Toronto, Ontario, Canada; [2]The Hospital for Sick Children, Toronto, Ontario, Canada

BACKGROUND: Respiratory syncytial virus (RSV) is the leading cause of lower respiratory tract infection in infants and young children worldwide. There are no specific drugs available to treat RSV; supportive care is provided until the virus is cleared by the immune system. Macrophages play an important role *in vivo* protecting hosts from RSV infections. Our group has recently described a method to generate stem cell-derived alveolar-like macrophages (PSC-AM). Here we investigate the interaction between PSC-AMs and RSV.

METHODS/RESULTS: To study RSV infection, we utilized a modified RSV strain expressing green fluorescent protein (GFP). Initially, we co-incubated RSV-GFP with PSC-AM to determine if PSC-AM become infected. No appreciable GFP signal was detected over 72 hours. In contrast, human epithelial cells were robustly infected. Upon further investigation, gene expression of a cellular receptor for RSV, nucleolin, was found to be insignificant in the PSC-AMs. PSC-AMs co-incubated with RSV-GFP led to a dose and time dependent reduction in RSV infectivity of human epithelial cells. Importantly this reduction was not seen when RSV-GFP was incubated with Sf9 cells (an insect cell line that is not infected by RSV). Upon further characterization, immunofluorescence and electron microscopy revealed viral like particles associated with PSC-AMs.

CONCLUSION/SIGNIFICANCE: PSC-AMs were found to be resistant to productive RSV infection and inhibitory towards RSV infectivity. These findings warrant further investigation into this interaction, with the potential to elucidate novel therapeutic strategies for RSV.

113. Suramin Inhibits Zika and Chikungunya Virus Replication by Interfering with Attachment and Later Steps of the Replication Cycle

Irina Albulescu, MS[1], Kristina Kovacikova, MS[1], Ali Tas, BS[1], Tabitha Hoornweg, PhD[2], Salvatore Ferla, PhD[3], Andrea Brancale, PhD[3], Jolanda Smit, PhD[2], Eric Snijder, PhD[1], **Martijn van Hemert, PhD**[1]

[1]Leiden University Medical Center, Leiden, Netherlands; [2]University Medical Center Groningen, Groningen, Netherlands; [3]Cardiff University, Cardiff, United Kingdom

Chikungunya virus (CHIKV) and Zika virus (ZIKV) are re-emerging arboviruses that have affected millions of people worldwide over the past decade. We and others have shown that the anti-parasitic drug suramin inhibits CHIKV replication by interfering with an early step in the replicative cycle, with an additional minor effect on viral RNA synthesis. In CPE reduction assays suramin also inhibited ZIKV replication with an EC_{50} of ~40 µM (CC_{50} ~1.9 mM; SI ~47.5). In single replication cycle experiments, suramin caused a dose-dependent reduction in intracellular ZIKV RNA levels and a >3-log reduction in infectious progeny titers. Time-of-addition studies revealed that suramin inhibits an early step of the ZIKV replication cycle, as well as the release of infectious progeny. When suramin treatment was initiated post-entry, ZIKV RNA synthesis was unaffected while the release of infectious progeny was reduced. Studies with fluorescently labeled CHIKV and radioactive CHIKV and ZIKV demonstrated that suramin inhibits virus attachment. We have selected suramin-resistant CHIKV variants that exhibited a ~3.5-fold resistance and had acquired two mutations (N5R and H18Q) in the E2 envelope protein, besides several mutations in the non-structural proteins. Reverse genetics studies revealed that the E2 mutations are responsible for suramin resistance. Suramin may prevent binding or fusion of CHIKV with the host cell membrane, possibly by interfering with conformational rearrangements that need to occur in E1E2 glycoprotein heterodimers. Besides interfering with attachment, suramin also inhibited ZIKV biogenesis possibly by interfering with glycosylation and maturation of the virus during its traffic through the secretory pathway.

114. Discovery of Toll-Like Receptor Potentiators

Joe Baldick, PhD, Betsy Eggers, MS, Robert Bertekap Jr., MS, Kevin Pokornowski, MS, Neil Burford, PhD, Andrew Alt, PhD, **Stephen Mason, PhD**
Bristol-Myers Squibb, Wallingford, Connecticut, United States of America

As one of the first lines of defense in triggering a response against invading pathogens, Toll-Like Receptors (TLRs) are expressed by cells within tissues that are most vulnerable to pathogen entry into the host. TLRs are expressed by cells of both the innate and adaptive immune systems within these tissues.

Synthetic agonists of TLR activity induce TLR signaling and have therapeutic effects in human diseases such as certain cancers and chronic viral infections. Although there are a few notable successes, the clinical efficacy of TLR agonists has been somewhat disappointing. The most likely reason for this is the association of TLR agonists with dose limiting toxicities.

Here we report the discovery of TLR Potentiators (TLRP), small molecules that synergize with natural or synthetic agonists of certain TLRs. The TLRPs are not active as agonists on their own, but amplify TLR signaling only when and where TLR agonists are present. The TLRPs described here are specific for TLR7, 8, and 9, and do not potentiate TLR3 or TLR4. Using TLR7 as a model, the profile of cytokines induced by TLRP alone is drastically different than that of a TLR7 agonist alone or with agonist in the presence of TLRP. Thus, while synthetic TLR agonists indiscriminately activate receptors, a TLRP would only exert effects where TLR signaling is naturally occurring, providing a "functional selectivity" to their effects that cannot be achieved with direct-acting agonists. Therefore, TLRPs may be able to circumvent the toxicity issues posed by potent synthetic TLR agonists.

115. Screening of FDA Approved Compounds Library Targeting the mRNA Capping of *Venezuelan equine encephalitis virus* (VEEV)

Ana S. Ramos, MS[1], Changqing Li, PhD[1], Eydoux Cécilia, PhD[1], Aouadi Wahiba, MS[1], Martin Baptiste, MS[1], Contreras Jean Marie, PhD[2], Morice Christophe, PhD[2], Jung Marie-Louise, PhD[2], Bruno Canard, PhD[1], Guillemot Jean-Claude, PhD[1], Decroly Etienne, PhD[1], Coutard Bruno, PhD[1]
[1]Aix Marseille Université, CNRS, AFMB UMR 7257, Marseille, France; [2]Prestwick Chemical, 67400 ILLKIRCH – Strasbourg – France

Venezuelan equine encephalitis virus (VEEV), an important re-emerging alphavirus from the *Alphavirus* genus, *is* a significant human and equine pathogen. Alphaviruses have a unique viral mRNA capping mechanism carried out by the non structural protein 1 (nsP1). NsP1 is thus an attractive target for drug design. First, the S-Adenosylmethionine (AdoMet)-dependent methyltransferase (MTase) activity of nsP1 methylates the N7 position of a GTP molecule to form ^{m7}GTP. Then, the enzyme catalyses the formation of a covalent bond between a conserved histidine and ^{m7}GTP leading to the formation of the ^{m7}GMP-nsP1 complex (guanylylation, GT). The last step is the transfer of the ^{m7}GMP from nsP1 to the 5' end of the diphosphate viral mRNA.

We previously demonstrated that the guanylylation of nsP1 can be monitored by Western blot using an anti-cap antibody. Using this detection strategy, we developed an ELISA assay to quantify the ^{m7}GMP-nsP1 adduct and identify new nsP1 inhibitors. This assay was used to screen a ~1 200 compounds library of FDA approved drugs. We selected 21 hits showing more than 80% inhibition effect at 50µM concentration. The hits were then confirmed by IC50 determination, an orthogonal assay on the nsP1 MTase activity, and specificity against cellular MTases. Compared to the capping inhibitor Sinefungin, the best hits showed higher inhibition potency and specificity on nsP1. Altogether, the results show that this enzyme-based screening is a convenient way to select potent compounds targeting the mRNA capping of Alphaviruses, and provided initial result to drug design for the development of possible antivirals.

116. Inactivation of Respiratory Viruses Using Far-Infrared Radiant Heater

Chong-Kyo Lee, PhD[1], Chonsaeng Kim, PhD[1], Keunbon Ku, DVM[1], Jin Soo Shin, DVM[1], Hae Soo Kim, BS[1], Gi Ppeum Lee, MS[1], Chun Sik Jeon, MS[2], Hee Jung Lee, BS[2], Jaekyung Hyun, PhD[3]

[1]Korea Research Institute of Chemical Technology, Daejeon, Korea, Republic of; [2]Ecopartners Ltd, Seoul, Korea, Republic of; [3]Korea Basic Science Institute, Cheongju, Korea, Republic of

Virus inactivation is important to control the viral diseases and spread. Not like vaccine or pharmacological drug, it can prevent various viruses from spreading between the environment, people and animals. As high temperature exposure could be a good option to inactivate viruses, we have tested the heat sensitivities of several viruses mostly respiratory, such as influenza viruses, human rhinoviruses, coronaviruses, and adenoviruses. Mineral heat ware (produced by Ecopartners Ltd, Seoul, Korea) was used as a heat source. Far-infrared radiant heater is a regenerative FIR heating system applied physical and bioware technologies. The wavelength of far infrared rays was 4 ~ 10 um. Test temperature was 170°C and 210°C. Most viruses were inactivated within 60 seconds at both temperatures but certain viruses showed incomplete inactivation. Understanding the relationship between heat sensitivities and physical characteristics of viruses may help the control of emerging viruses.

117. VIRIP – an Anti-HIV Host Peptide Output Hypothesis

Aitsana A. Maslakova[1], Vera S. Efimova[1], Alexei S. Maslakov[1], Victor E. Spangenberg[2], Mikhail A. Rubtsov[1], Igor V. Orlovsky[3]

[1] Biology Department, Lomonosov MSU, Moscow, Russia; [2] Vavilov Institute of General Genetics, Moscow, Russia; [3] Lomonosov MSU A.N. Belozersky Research Institute of Physical and Chemical Biology, Moscow, Russia

Alpha1-antitrypsin is known to be a precursor of bioactive peptides. Full-length protein proteolytic cleavage has been regarded as the only mechanism of these peptides production. VIRIP (virus-inhibitory peptide) is such a derivative, a potent anti-HIV host peptide (Münch J., 2007). It is also believed to be produced proteolytically, although its C-terminus proteolytic formation hasn't been demonstrated yet. Our previous data on *SERPINA1* gene coding region relative expression analysis implied the existence of a more fine mechanism – regulation at a transcriptional level. Northern hybridization with cRNA probe derived from exon 5 on different total RNA samples proved the *SERPINA1* gene short transcripts existance. Our hypothesis has been supported by an independent research (Matamala N., 2017). Here we hypothesize that VIRIP can be produced by immune cells from unique short transcript(s) containing coding region of exon4/exon5 and/or exon5, which are polyadenylated at a cleavage site (T) located 13 nt downstream of putative AACAAA polyadenylation signal, thus producing a stop-codon and a transcript lacking 3'-UTR and encoding the VIRIP sequence. It has been established that such 3'UTR-depleted transcripts are highly stable, that must be important in case of VIRIP. AAT full-length blood concentration in HIV-infected patients (around 30uM) tends not to exceed its normal levels (Bryan C.L., 2010), but VIRIP IC50 (4-20uM, depending on HIV subtype) is relatively high (Münch J., 2007). The existence of stable unique transcript(s) may increase local VIRIP level. Our hypothesis may help to solve such a discrepancy.

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118. Role of Receptor Tyrosine Kinases and Associated Gangliosides in Influenza Virus Replication

Pieter Vrijens, MS[1], Els Vanstreels, PhD[1], Roberto Ronca, PhD[2], Marco Presta, PhD[2], Sandra Liekens, PhD[1], Lieve Naesens, PhD[1]

[1]Rega Institute for Medical Research, KU Leuven – University of Leuven, Belgium, Leuven, Vlaams-Brabant, Belgium; [2]Experimental Oncology and Immunology, University of Brescia, Italy, Brescia, Italy

Like all viruses, the influenza virus crucially depends on numerous host cell factors for its replication. These cellular factors are potential antiviral targets, since they are less likely to mutate and, consequently, viral resistance is less likely to emerge. In this project, we focus on inhibitors of cellular protein kinases as potential influenza blockers. Through screening of a protein kinase inhibitor library, we identified Ki8751 as a compound with robust and broad activity against influenza A and B viruses and low cytotoxicity. Ki8751, a known inhibitor of the receptor tyrosine kinases VEGFR2, FGFR2 and PDGFR , was shown to act during the viral entry phase but had no effect on virus binding to the cells. In parallel, we observed that influenza virus entry is four-fold more efficient in CHO-K1 cells when compared to CHO-wild type cells. CHO-K1 cells lack complex gangliosides such as GM1 but do express the GM3 ganglioside, which has either a positive or negative effect on RTK signaling pathways. Hence, we are verifying whether the antiviral activity of Ki8751 is based on inhibition of a specific RTK type that is positively regulated by GM3.

Using a panel of CHO transfectant cell lines, we further found that influenza replicates more efficiently in CHO cells that express VEGFR2. Strikingly, VEGFR2 seems to affect viral RNA synthesis and/or nuclear export, not viral entry. We hypothesize that this could be linked to intranuclear VEGFR2 and are currently investigating the viral factor(s) involved.

119. Evaluate the Broad-Spectrum Protective Efficacy of *Lactobacillus* Species Against Influenza Viruses in Mice

Jung-hoon Kwon, DVM, Seong-Su Yuk, DVM, Sol Jeong, DVM, Jei-hyun Jeong, DVM, Ji-ho Lee, DVM, Jun-beom Kim, DVM, Yu-jin Kim, DVM, Chang-Seon Song, DVM, PhD

College of Veterinary Medicine, Konkuk University, Seoul, Korea

Influenza virus infections continue to be a significant public health problem. Recently new subtypes of influenza virus such as H5N6 and H7N9 represent a global pandemic threat. For improved therapies and preventive measures against influenza, the need for a broad-spectrum antiviral therapeutics such as probiotics has been increased. In almost previous studies including ours, antiviral efficacy of *lactobacillus* was evaluated using just one strain of *Lactobacillus* species. In this study, we evaluated the protective efficacy of 2 strains of *lactobacillus* (L. Rhamnosus and L. Sakei) against 3 subtype of influenza viruses (H1N1, H3N2 amd H5N2). Administration of 2 strain of *lactobacillus* reduced mortality in mice challenged with 3 influenza viruses. In H5N2 influenza virus challenged group, the body weights of L. Rhamnosus administered mice were significantly higher than that of L. Sakei administered mice and positive control. There was no significant difference in body weight change in H1N1 and H3N2 challenged group. These results indicated that antiviral efficacy of *Lactobacillus* species were different depend on challenge virus. Although 2 strain of *lactobacillus* showed same antiviral efficacy against H1N1 and H3N2 influenza viruses, however L. Rhamnosus showed higher protective efficacy than L. Sakei in H5N2 challenge group. Therefore, for select efficient *lactobacillus*, optimize challenge model could be critical and our H5N2 challenge model in mice considered appropriate to select *lactobacillus*. Further, we need to use various challenge model to select *lactobacillus* showed broad-spectrum antiviral efficacy.

120. Synergistic Antiviral Effect of Polymerase and Autophagy Inhibitors on Dengue and Zika Virus Infected Cells *In Vitro*

Cecilia Cima, PhD[1], Marcella Bassetto, PhD[1], Daniela Friese, BS[2], Juliane Nolte, BS[2], Gerhard Dobler, MD, PhD[2], Silke Wölfel, MD[2], Andrea Brancale, PhD[1], **Joachim Bugert, MD, PhD**[2]
[1]Cardiff School of Pharmacy & Pharmaceutical Sciences, Cardiff, Wales, United Kingdom; [2]Institut für Mikrobiologie der Bundeswehr, München, Bavaria, Germany

Zika virus is a recently reemerging mosquito borne flavivirus, causing flu-like illness and CNS infection, which can lead to embryopathy/microcephaly *in utero*. Increasing numbers of infections have been reported in 2015 and 2016 in a global context, causing WHO to declare a public health emergency.

Dengue virus is endemic in over 100 countries. In the last 50 years incidence has increased 30-fold and dengue haemorrhagic fever is the cause of 500000 hospitalizations and over 20000 deaths worldwide every year. There is no vaccine or approved treatment for infected or exposed individuals to either virus infection.

We report here the antiviral effect of novel broad spectrum flavivirus polymerase inhibitors of the Cima series versus Dengue and Zika viruses in human cell lines.

Lead compound Cima 4 inhibits dengue and Zika viruses with IC50 of 4.58μM+/-0.1 and 4μM+/-0.2, respectively. In combination with L- ddBCNA cf2642, an autophagy inhibitor originally reported to be active versus vaccinia and measles viruses (McGuigan at al., 2013, and unpublished data), synergy was observed, with a combined IC50 of 1μM+/-0.2. This is a 50-fold improvement *in vitro* over ribavirin alone versus Zika virus. CC50 was determined to be >20μM for all antivirals used, giving a selective index of the drug combination of 20 or better. We believe our results warrant further development of the agents and make a case for combined virus specific and cell targeting therapies.

121. Baicalin as an *In Vitro* Inhibitor for Chikungunya Virus

Adrian Oo, MS[1], Stephen Higgs, PhD[2], Sazaly AbuBakar, PhD[3], **Keivan Zandi, PhD**[4]
[1]Tropical Infectious Disease Research and Education Center, Faculty of Medicine, UM, Kuala Lumpur, KL, Malaysia; [2]Kansas State University; [3]Tropical Infectious Disease Research and Education Center, UM, Kuala Lumpur, WP, Malaysia; [4]1- TIDREC, UM, Malaysia 2- Emory University, Atlanta, USA, Atlanta, Georgia, United States of America

Over the past decade, Chikungunya virus (CHIKV) has re-emerged as one of global health threat, affecting the lives of millions annually. As the coverage of affected geographical regions expands over the years, effective treatment for CHIKV remains elusive. This has prompted us into designing this study for evaluating the bioflavonoid, baicalin's inhibitory activity against CHIKV. Identification of the compound's cytotoxicity in various cell lines was followed by different antiviral experiments to determine the nature of baicalin's inhibitory actions against CHIKV replication. Our MTS assays showed that baicalin was not toxic to all cell lines with MNTD values > 600 μM, 354.39 μM and 110.1 μM recorded for Vero, BHK-21 and HEK 293T cells, respectively. Preliminary antiviral assays conducted, demonstrated the most potent inhibitory activity against free extracellular virus particles with EC_{50} = 7.11 μM, followed by inhibition of virus entry, attachment and intracellular viral replication, respectively. Different CHIKV subgenomic RNAs were effectively inhibited by baicalin, however our luciferase assay demonstrated weaker inhibitory effect of our compound on the viral genome replication. Immunofluorescence and immunoblotting experiments demonstrated dose-dependent inhibitions of different viral proteins expression and production, whereas cellular autophagy and apoptosis markers such as LC3 and Bax were also reduced in the treatment groups. In summary, baicalin possesses virus replication inhibitory properties with great potential for further development into a novel antiviral compound against CHIKV.

122. Mechanism Study of Baicalin and Baicalein Against Dengue Virus

Pouya Hassandarvish, PhD[1], Justin Jang Hann Chu, PhD[2], Sazaly AbuBakar, PhD[3], **Keivan Zandi, PhD**[4]

[1]Department of Medical Microbiology, Faculty of Medicine, UM, Malaysia; [2]Dept of Microbiology,Yong Loo Lin School of Medicine, National University of Singapore; [3]Tropical Infectious Diseases Research and Education Center, UM, Malaysia; [4]1- Emory University 2-TIDREC, University of Malaya, Atlanta, Georgia, United States of America

Dengue virus (DENV) is an important human arbovirus belongs to *Flaviviridae* without any efficient therapeutics available. Therefore, finding an effective antiviral against this virus is crucial. We previously reported the anti-DENV activities of two flavonoids baicalein and baicalin against different stages of virus replication cycle in Vero cells. In order to find out more on the antiviral effects of baicalein and baicalein against all DENV serotypes, the compounds were tested against DENV1, DENV2, DENV3 and DENV4 using virus yield reduction assay in a human cell line. Transmission electron microscopy (TEM) was used the visualized the effects of baicalin and baicalein against DENV-2 entry. Immunoblot assay against capsid protein and DENV polymerase protein (NS5) was used to examine the effects on DENV life cycle. Results obtained suggestted that both compounds inhibited all DENV serotypes. However baicalin inhibited DENV with very low IC50; DENV1 at 9.6 µg/ml, DENV2 at 7.1 µg/ml, DENV3 at 25 µg/ml & DENV4 at 9.8 µg/ml, in comparison to baicalein; DENV1 at 30.5 µg/ml, DENV2 at 17.3 µg/ml, DENV3 at 23.7 µg/ml & DENV4 at 19.7 µg/ml. The electron microscopic (TEM) investigation showed that both baicalein and baicalin can inhibit dengue virus replication from the early stages of infection until after virus entry. Immunoblot assay results suggested that baicalein and baicalin affected synthesis of the capsid protein and NS5 in a dose response manner. In summary, baicalein and baicalin demonstrated effective antiviral properties *in vitro*, hence, these compounds could be explored further as potential antiviral agent against DENV.

123. Exploring Polypharmacology for the Design of Broad-Spectrum Influenza Antivirals

Jun Wang, PhD

Assistant Professor Department of Pharmacology and Toxicology University of Arizona, Tucson, Arizona, United States of America

Influenza viruses are respiratory pathogens that account for both seasonal influenza as well as influenza pandemics. As multidrug-resistant influenza viruses continue to emerge, there is a clear need for the next-generation of antivirals. We have previously shown that by targeting the M2-S31N mutant proton channel in influenza A viruses (AM2-S31N), we were able to design channel blockers that are active against multidrug-resistant influenza A viruses. However, one limitation of the AM2-S31N inhibitors is that they do not inhibit the BM2 channel, thus they are not active against the influenza B viruses. The goal of the current effort is to extend the therapeutic value of AM2-S31N inhibitors to cover influenza B viruses as well.To achieve this, we have recently identified several lead compounds that have broad-spectrum antiviral activity against both influenza A and B viruses. However, the antiviral mechanism in inhibiting influenza A and B viruses is different: inhibition of influenza A viruses is AM2-dependent, while inhibition of influenza B viruses is BM2-independent. We are currently conducting mechanistic studies to delineate the antiviral mechanism towards influenza B viruses. It is expected that antivirals with polypharmacology will have a higher genetic barrier to drug resistance than traditional single-target drugs. Continuous lead optimization of this series of compounds is likely to yield the first-in-class broad-spectrum antivirals that are effective against drug-resistant influenza A and B viruses.

124. Pre- and Post-Exposure Treatment of Quercetin-3-βO-D-Glucoside Against Ebola Virus Infection

Xiangguo Qiu, MD[1], Andrea Kroeker, PhD[1], Shihua He, PhD[1], Robert Kozak, PhD[1], Jonathan Audet, PhD[1], Majambu Mbikay, PhD[2], Michel Chretirn, MD[2]
[1]*Special Pathogens Program, NML/ Public Health Agency of Canada, Winnipeg, Manitoba, Canada;* [2]*Clinical Research Institute of Montreal*

BACKGROUND: Ebola outbreaks occur on a regular basis, with the current outbreak in West Africa being the largest ever recorded. It has resulted in over 11 000 deaths in four African countries and has received international attention and intervention. Although there currently are no approved therapies or vaccines, many promising candidates are undergoing clinical trials and several have had success in promoting recovery from Ebola. However, these prophylactics and therapeutics have only been designed and tested against the species of Ebola that causing the current outbreak. Future outbreaks involving other species would require reformulation and possibly redevelopment. Therefore a broad spectrum alternative is highly desirable.

METHOD AND RESULTS: Using a variant of *Ebola virus* (EBOV) that expresses green fluorescent protein, a flavonoid derivative called Quercetin-3-β-O-D-glucoside (Q3G) has been identified to have antiviral activity in vitro. Similar results were also found with *Sudan virus* (SUDV). Furthermore, we have evaluated the protective efficacy of Q3G as prophylactic and/or post-exposure treatment agent in a mouse-adapted EBOV model. The results showed that Q3G not only can fully protect mice from EBOV infection when given 30 minutes prior to infection, but also can protect mice up to 87.5% when the treatment was initiated at 24 hour post challenge.

CONCLUSION: This study serves as a proof of principle that Quercetin-3-β-O-D-glucoside has potential as a prophylactic/treatment agent against Ebola virus infection.

125. Sensitivity to a Potent Lassa Antiviral is Modulated by a Virulence Determinant

Sean Amberg, PhD[1], Ikenna Madu, PhD[1], Megan Files, BS[1], Tiffany Huelar, BS[1], Kie-Hoon Jung, PhD[2], Brian Gowen, PhD[2], Shawn Iadonato, PhD[1], Kristin Bedard, PhD[1]
[1]*Kineta, Seattle, Washington, United States of America;* [2]*Utah State University, Logan, Utah, United States of America*

LHF-535 is a small molecule inhibitor of virus entry currently in development as a therapeutic candidate for Lassa fever and other hemorrhagic fevers of arenavirus origin. Antiviral activity was recently evaluated against a panel of lentiviral pseudotypes incorporating the arenavirus envelope glycoprotein (GP). This panel included nine Lassa strains, with at least one representative from each of the four lineages of Lassa. While all strains exhibited nM-range sensitivity to LHF-535, the LP strain exhibited less sensitivity. The LP strain is the prototype of lineage I and was isolated from the first documented case of Lassa fever in 1969; to date, it is the only lineage I member described, as lineages II and IV comprise most clinical cases. The modulated sensitivity of the LP strain was found to be mediated by a single amino acid substitution in GP not found in the other lineages (V434I). Interestingly, this site has been implicated as the major attenuation determinant of the Junín virus vaccine strain, Candid1. When the attenuation determinant (F427I) was introduced into the Junín GP, pseudotype sensitivity to LHF-535 was lost. Live virus yield assays were used to confirm the sensitivity difference, using Candid1 and Romero strains. Using Tacaribe virus as a surrogate, viral isolates selected for reduced LHF-535 sensitivity were generally attenuated in the AG129 mouse model of arenavirus pathogenesis, including one isolate with a substitution equivalent to the Candid1 attenuation determinant (F425L). This correlation between inhibitor sensitivity and virulence could have significance for the clinical use of this inhibitor class.

126. Substituted Pyrimidines as Potent Non-Nucleoside Reverse Transcriptase Inhibitors

Petr Simon, PhD[1], Lucie Cechova, MS[1], Ondrej Baszczynski, PhD[1], David Saman, PhD[1], George Stepan, PhD[2], Eric Hu, PhD[2], Eric Lansdon, PhD[2], Petr Jansa, PhD[2], Zlatko Janeba, PhD[1]
[1]*Institute of Organic Chemistry and Biochemistry, Czech Academy of Sciences, Prague, Czech Republic;*
[2]*Gilead Sciences Inc., 333 Lakeside, Foster City, CA 94404, USA*

A series of novel substituted pyrimidines (Fig. 1), as potential non-nucleoside reverse transcriptase inhibitors (NNRTIs) derived from diarylpyrimidine (DAPy),[1] was prepared by new synthetic approach. The pyrimidine ring was substituted with 4-cyanophenylamino moiety and with another aromatic system attached through various linkers. Further modifications are present at the C-5 position of the pyrimidine moiety. Structure and anti-HIV activity relationship (SAR) study was performed on a series of some 20 compounds. The most potent derivative from the series, so far, exhibited low nanomolar anti-HIV-1 activity (EC_{50} = 4 nM) with no significant toxicity (CC_{50} > 57.1 μM).

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LITERATURE:

1) Zhan, P.; Chen, X.; Li, D.; Fang, Z.; De Clercq, E.; Liu, X. *Med. Res. Rev.* 2013, *33* (S1), E1-E71.

2) Simon, P.; Baszczynski, O.; Saman, D.; Stepan, G.; Hu, E.; Lansdon, E. B.; Jansa, P.; Janeba, Z. *Eur. J. Med. Chem*. 2016, *122*, 185-195.

127. Evaluation of Ribavirin Against Recombinant Oncolytic Newcastle Disease Virus Replication In Vitro

Weijia Wang, MS, Xing Cheng, MS, Udaya Rangaswamy, PhD, Hong Jin, PhD
Medimmune

We have generated a recombinant Newcastle disease virus (rNDV) by reverse genetics for virotherapy. The rNDV has three modifications: change of the fusion protein cleavage site, insertion of 198nt sequence between the HN and L genes, and insertion of a human granulocyte-macrophage colony-stimulating factor (GM-CSF) transgene between the P and M gene junction (Cheng,X 2016). rNDV retains oncolytic activity, but is attenuated in chicken species and is no longer classified as a select agent. As NDV is not a human pathogen, a targeted anti-NDV drug has not been developed. However, circumstances could be envisioned under which an antiviral agent would be desired for rNDV used in humans as an oncolytic agent. We have therefore evaluated some RNA virus inhibitors for their antiviral activity against NDV. Ribavirin, approved by the FDA for treatment of severe RSV infection, was shown to inhibit NDV replication in vitro. The drug concentration that inhibits 50% of viral replication (IC_{50}) is approximately 1.5 μg/ml in rNDV-infected HT1080 cells. The half maximal effective concentration (EC_{50}) of the drug that prevented cell death from viral infection ranges from 2.0-4.4 μg/ml at different multiplicity of infection in HT1080 cells. This drug concentration is within the range of the EC_{50} value of ribavirin on RSV replication in vitro, 2-8 μg/ml (De Clercq,E 1996). rNDV appears to be more sensitive to ribavirin's inhibition than wild type NDV-73T strain, which is likely attributable to its decreased cleavage efficiency. Our data indicates that ribavirin could be potentially effective against rNDV infection in vivo.

128. Verdinexor, a Selective Inhibitor of Nuclear Export (SINE) Compound, Exhibits Significant Antiviral Activity Against HIV and SIV

Sharon Tamir, PhD[1], Shelton Cochran, BS[1], Marie Mankowski, BS[2], Priscilla Hogan, BS[2], Trinayan Kashyap, MS[1], Yossi Landesman, PhD[61], Margaret Lee, PhD[1], Sharon Shacham, PhD[1], Roger Ptak, PhD[2]

[1]Karyopharm Therapeutics, Newton, Massachusetts, United States of America; [2]Southern Research Institute, Frederick, Maryland, United States of America

Simian immunodeficiency virus (SIV) is a retrovirus closely related to human immunodeficiency virus (HIV) that can cause infection in at least 45 species of non-human primates. Previous transmission events from non-human primates to humans and adaptation of the virus in humans have produced multiple strains of AIDS-inducing HIV and suggest that future transmissions are highly possible. A promising HIV therapy under development is verdinexor (KPT-335), a well-tolerated, orally-bioavailable inhibitor of the nuclear export protein, exportin 1 (XPO1). In previous studies verdinexor was shown to cause significant nuclear retention of the HIV Rev protein and exhibited significant *in vitro* antiviral efficacy against 7 strains of HIV and 5 significant HIV co-infections. As part of verdinexor's continued development as an anti-HIV therapeutic, its efficacy against SIV was assessed *in vitro* to ensure its activity against potential new strains of HIV in addition to existing strains. Verdinexor was tested in a standard human peripheral blood mononuclear cell (PBMC)-based HIV/SIV replication assay against two SIV isolates and one reference HIV-1 isolate. It demonstrated significant antiviral activity (IC_{50}) and therapeutic index (TI) values against both SIV strains ($SIV_{mac}239$: IC_{50} = 7.54 nM, TI = 30.0; $SIV_{mac}251$: IC_{50} = 4.30 nM, TI = 52.6), and similar, expected levels of activity against the HIV-1 strain (Ba-L: IC_{50} = 6.56 nM, TI = 34.5). The nanomolar level of efficacy and double-digit therapeutic indices demonstrate that verdinexor has the potential to be a powerful antiviral agent against existing and potential future strains of HIV.

129. Human Ex Vivo Lung Tissue as Model System to Investigate Novel Host-Directed Antivirals Against Influenza A Virus

Jessica von Recum-Knepper, PhD[1], Thorsten Wolff, PhD[2], Silke Stertz, PhD[3], Torsten Steinmetzer, PhD[4], Eva Böttcher-Friebertshäuser, PhD[5], Sumit Chanda, PhD[6]

[1]University of California, San Diego, School of Medicine, La Jolla, California, USA, California, United States of America; [2]Influenza Viruses & Other Respiratory Viruses, Robert Koch Institute, Berlin, Germany; [3]Institute of Medical Virology, University of Zurich, Switzerland; [4]Institute of Pharmaceutical Chemistry, Philipps University Marburg, Germany; [5]Institute of Virology, Philipps University Marburg, Germany; [6]Sanford Burnham Prebys Medical Discovery Institute, La Jolla, USA

Influenza A virus (IAV) is an RNA virus encoding up to 13 viral proteins. Due to this limited coding capacity IAV needs the host cellular machinery to complete its lifecycle. To date, the roles of host proteins in IAV infection and their potential as antiviral drug targets have predominantly been characterized using immortalized cell lines like A549. However, the use of cell lines has several drawbacks including differences in host regulatory networks compared to nontumorigenic cells. Therefore, better models are urgently needed to investigate host-directed antivirals against IAV. Human ex vivo lung tissue is a particularly useful model system that applies authentic tissue in which different cell types are still connected in 3D structure and able to interact with each other.

We successfully established an ex vivo human lung tissue model and confirmed that it supports replication of several IAV strains, exhibiting up to 3-log titer increase upon 48 hours of infection. Furthermore, we demonstrated that IAV titers as well as the levels of proinflammatory cytokines like IL-1b and TNF-a were significantly decreased by the addition of antiviral drugs like oseltamivir. Next, we investigated compounds targeting host proteins such as the protease TMPRSS2 or members of the polo-like kinase family. Several compounds like BAPA and BI2536 displayed strong antiviral activity at non-toxic levels.

In summary, ex vivo human lung tissue represents an authentic model system for testing and developing novel antiviral drugs targeting human host proteins.

130. Inhibition of Respiratory Virus Infection by Cholesterol Reducing Agents

Shringkhala Bajimaya, MS
University of Rochester School of Medicine and Dentistry, Rochester, New York, United States of America

Many enveloped viruses utilize cholesterol-rich lipid rafts at the plasma membrane for virus assembly and production. However, the functional role of cholesterol in virus formation and infectivity is unclear. In this study, we investigated the effects of FDA-approved cholesterol-reducing agents on raft formation and the production of infectious parainfluenza virus (PIV), influenza A virus (IAV) and respiratory syncytial virus (RSV) in human airway cells. Depletion of cholesterol with the agents, especially when combined, significantly decreased production of all infectious viruses. Depletion of cellular cholesterol reduced cell surface accumulation of PIV glycoproteins and inhibited virus assembly and release. In contrast, depletion of cellular cholesterol did not decrease IAV and RSV surface glycoproteins accumulation, and virus particles were efficiently released from the cells. However, the released virus particles were less stable due to abnormal virion density and decreased cholesterol content in the viral membrane. Replenishing the virus released from the treated cells with cholesterol rescued virus stability and infectivity. Collectively, our findings suggest that cholesterol is critical for PIV assembly, and maintaining the stability of infectious IAV and RSV particles. Our data suggests that cholesterol is an attractive target for antiviral agents against various clinically important respiratory viruses.

131. Inhibition of Nuclear Export by Verdinexor May Enhance a Broad Therapeutic Window in Mouse Models of Influenza

Sharon Tamir, PhD[1], Shelton Cochran, BS[1], Patricia Jorquera, PhD[2], Jennifer Pickens, PhD[2], Sharon Shacham, PhD[1], Margaret Lee, PhD[1], Ralph Tripp, PhD[2]
[1]*Karyopharm Therapeutics, Newton, Massachusetts, United States of America;* [2]*Department of Infectious Diseases, University of Georgia, Athens, Georgia, United States of America*

Oseltamivir is the standard of care for treating influenza and it is recommended that treatment is initiated within 48 hours of symptom onset. However, with symptoms similar to those of other viral and bacterial infections, in some cases a positive diagnosis can take more than 48 hours, so there is a significant unmet need for treatments with a prolonged therapeutic window. One such promising new treatment is verdinexor (KPT-335), a well-tolerated, orally-bioavailable inhibitor of the nuclear export protein, exportin 1 (XPO1). The two drugs' different mechanisms of action may explain their different therapeutic windows: oseltamivir, a neuraminidase inhibitor, prevents release of viral particles and promotes aggregation; verdinexor inhibits the nuclear export of important influenza cargos like viral ribonucleoprotein and is thus efficacious even after significant viral release. We evaluated the efficacy of verdinexor and oseltamivir at different time points post-inoculation to compare the drugs' therapeutic windows. Female BALB/c mice were infected with influenza A virus and divided into control groups or treatment groups, which received 10 mg/kg oseltamivir BID beginning 1 day after inoculation or 20 mg/kg verdinexor QoD beginning up to 4 days post-inoculation. Both verdinexor and oseltamivir showed strong efficacy when treatment was initiated 1 day post-inoculation. When initiation of dosing was delayed by up to 4 days post-inoculation, verdinexor demonstrated antiviral activity (a reduction in lung viral titer) as good as or better than dosing only 1 day post-inoculation. These results suggest that verdinexor has potential to be an effective antiviral with a prolonged therapeutic window.

132. Apilimod, a PIKfyve Inhibitor with Antiviral Activity against Ebola and Marburg Viruses

Julie Dyall, PhD[1], Elizabeth Nelson, PhD[2], Thomas Hoenen, PhD[3], Alyson Barnes, PhD[2], Huanying Zhou, MS[1], Janie Liang, MS[1], Julia Michelotti, PhD[1], William Dewey, MS[1], Lisa Evans Dewald, PhD[1], Richard Bennett, PhD[1], Patrick Morris, PhD[4], Rajarshi Guha, PhD[4], Carleen Klumpp-Thomas, PhD[4], Crystal McKnight, PhD[4], Yu-Chi Chen, PhD[4], Craig Thomas, PhD[4], Scott Martin, PhD[4], Peter Jahrling, PhD[5], Lisa Hensley, PhD[1], Gene Olinger, PhD[1], Judith White, PhD[2]

[1]Integrated Research Facility, NIAID, NIH, Frederick, Maryland, United States of America; [2]University of Virginia, Charlottesville, Virginia, United States of America; [3]Laboratory of Virology, Division of Intramural Research, NIH, Hamilton, Montana, United States of America; [4]National Center for Advancing Translational Sciences, National Institutes of Health, Bethesda, Maryland, United States of America; [5]Integrated Research Facility and Emerging Viral Pathogens Section, NIAID, NIH, Frederick, Maryland, United States of America

The recent Ebola virus disease epidemic in Western Africa has demonstrated that there remains a pressing need to develop therapeutics to treat patients infected with filoviruses. Apilimod, a potent inhibitor of IL-12 and IL-23 production, was developed for the treatment of autoimmune diseases (e.g., Crohn's disease, rheumatoid arthritis, psoriasis) and was deemed well-tolerated in humans in Phase 2 trials. The molecular target of apilimod is phosphatidylinositol-3-phosphate 5-kinase (PIKfyve), which is involved in endosomal maturation and trafficking and is required for EBOV infection. In this study, we tested the potential utility of apilimod as an antiviral agent. We found that apilimod blocks Ebola virus (EBOV) and Marburg virus (MARV) infection in Huh 7, Vero E6, and primary human macrophage cells (IC_{50} ~10 nm). Mechanistic studies with EBOV glycoprotein bearing-virus like particles (VLPs) and transcription-replication competent VLPs suggest that apilimod is primarily an entry inhibitor, which prevents release of the viral genome into the cytoplasm to initiate replication. Our findings further indicate that apilimod blocks EBOV entry by blocking particle delivery into endolysosomes that contain Niemann-Pick C1, the intracellular receptor for EBOV. Concurrently, apilimod caused VLPs to accumulate in early endosome antigen 1-positive endosomes, which appeared larger than those observed in mock-treated cells. In summary, we introduce apilimod, a small molecule PIKfyve inhibitor that has proven safe in Phase 2 clinical trials, as a potential candidate for repurposing as part of a therapeutic regimen to treat filoviral infections.

133. Modified Tetrahydropteridines as Potent Anti-HIV NNRTIs with Improved Resistance Profile and Solubility

Ondřej Baszczyňski, PhD[1], Petr Šimon, PhD[1], David Šaman, PhD[1], Eric Hu, PhD[2], Eric Lansdon, PhD[2], Petr Jansa, PhD[2], Zlatko Janeba, PhD[1]

[1]IOCB Prague, Prague, Czech Republic; [2]Gilead Sciences Inc., Foster City, California, United States of America

Non-nucleoside reverse transcriptase inhibitors (NNRTIs) are generally the preferred agents in currently recommended combinations for the treatment of HIV-1 infection. A particular issue with NNRTIs is poor solubility, which can cause low bioavailability or difficulties during formulation. Another limitation of NNRTIs is the developemnt of resitant viral strains and thus compounds with higher genetic barrier to resistance developement are of high interest[1].

Here we disclose our discovery of new series of NNRTIs bearing the 5,6,7,8-tetrahydropteridine core. The most potent compound within the series exhibits promising anti-HIV activity against the wild type virus (EC_{50} = 2.6 nM) and low nanomolar activity against two highly prevalent HIV mutants (K103N and Y181C). Moreover, for the most active 5,6,7,8-tetrahydropteridine we observed improvement of the PBS solubility compared to all FDA approved NNRTIs.

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LITERATURE:

1. *Antiviral Res.* 2010; 85(1): 75-90; *Chem Soc Rev.* 2012; 41(13): 4657-70. *Antiviral Res.* 2010 85(1): 25-33. *J Int AIDS Soc.* 2013; 16: 1-14.

134. Identification of Novel Amodiaquine Derivatives as Anti-EbolaVirus Compounds

Yasuteru Sakurai, PhD[1], Norikazu Sakakibara, PhD[2], Masaaki Toyama, PhD[3], Masanori Baba, MD[3], Robert Davey, PhD[1]

[1]Texas Biomedical Research Institute, San Antonio, Texas, United States of America; [2]Tokushima Bunri University, Sanuki, Tokushima, Japan; [3]Kagoshima University, Kagoshima, Kagoshima, Japan

Ebola virus disease is a highly lethal and rapidly progressing disease caused by Ebola virus infection. The recent outbreak in West Africa killed 11,310 people and spread to USA and several European countries, generating a global public health threat. Although a few therapeutic agents are under development, there is currently no approved therapy. Recently, it was reported that Ebola virus disease patients, who were prescribed an anti-malaria drug, amodiaquine, had a significantly lower risk of death compared to other patients. In order to improve the antiviral activity of amodiaquine, we synthesized a series of derivatives and tested their anti-Ebola virus activity in our BSL4 laboratory. We found 14 compounds that were more potent than amodiaquine against replication competent Ebola virus. Several of them had selectivity indexes being more than 130. The screening also revealed a well-defined structure-activity relationship. Hydrophobicity of the aminophenol-moiety and a halogen group bonded to the quinoline ring were key to increase the antiviral activity without increasing cytotoxicity. Importantly, these features are independent each other and can be combined into one molecule. For mechanistic analyses using pseudotype viruses and a minigenome system, we found that the potent compounds worked by blocking host cell entry of Ebola virus, but not genome replication. Taken together, this study found multiple amodiaquine compounds that potently inhibited Ebola virus infection by targeting the entry step and they are good lead compounds for the future drug development.

135. Activation of STING Mediates Antiviral Effects in a Mouse Model of Chronic Hepatitis B

Emily Thi, PhD, Luying Pei, BS, Hui Huang, MD, PhD, Xin Ye, PhD, Agnes Jarosz, BS, Joseph Wasney, BS, Xiaowei Teng, PhD, Megan Fowler, BS, Shannon Tang, BS, Laurèn Bailey, PhD, Chris Moore, PhD, Rene Rijnbrand, PhD, Amy Lee, MS, Michael Sofia, PhD

Arbutus Biopharma, Burnaby, British Columbia, Canada

STING (stimulator of interferon genes) is a critical modulator of innate immune responses, and its activation has been shown to reduce tumor burden in cancer treatment. Many viruses such as Dengue, Hepatitis C and Hepatitis B virus (HBV) actively inhibit STING, and it is hypothesized that stimulation of STING signaling may be effective for potentiating host immune responses against viral infections.

Here we demonstrate through the use of two compounds in distinct chemical classes that STING activation in a mouse model of chronic HBV results in substantial anti-viral effects. A single dose of the small molecule drug DMXAA at 25 mg/kg was able to immediately suppress HBV DNA, resulting in up to 1 $\log_{10}$ reductions in both serum and liver compartments in an adeno-associated virus mouse model of HBV. Repeat treatment with either DMXAA or a chemically modified variant of the natural STING agonist 2'3'-cGAMP resulted in cumulative reductions of HBV DNA, in some cases exceeding the level of viral control mediated by the current standard of care agent, IFN . These effects correlated with activation of STING and upregulation of interferon stimulated genes in the liver.

In summary, treatment with two distinct drug modalities which activate STING resulted in anti-viral effects against HBV. The molecules assessed were chemically dissimilar yet engaged the same target and mediated similar viral control. Thus, the effects observed are not arising from off-target effects and demonstrate that STING may be a druggable target for chronic HBV infection.

136. Tricyclic Matrinic Derivative DXC-10 Inhibits Influenza A Virus Replication Before the Nucleoprotein Nucleus Entry

Jinqiu Yin, BS, Haiyan Yan, MS, Huiqiang Wang, MS, Shuo Wu, PhD, Danqing Song, PhD, Yuhuan Li, PhD

Institute of Medicinal Biotechnology, Chinese Academy of Medical Sciences & Peking Union Medical College, Beijing 100050, China

Given the emergence of drug-resistant influenza A virus (IAV) strains and the time-dependent effectiveness of drug or vaccine, there remains a need for novel anti-influenza drugs. We demonstrated the antiviral activity of a novel compound DXC-10, which is the chemical structure modification of Tricyclic Matrinic. We found that DXC-10 exhibited a broad-spectrum antiviral activity against influenza virus in vitro, including the oseltamivir or amantadine-resistant strains. Pharmacokinetic and toxicological studies of DXC-10 demonstrated favorable drugable properties and an encouraging safety property. The in vivo efficacy study showed that both the simultaneous administration and intragastric administration of DXC-10 protected 40% of animals from death and reduced the viral load by about 2 logs in IAV-infected mice. Mechanistically, DXC-10 had no effect on the viral membrane protein hemagglutinin (HA) and neuraminidase (NA). A time-ofaddition study showed that DXC-10 plays a role in very early stage of IAV replication circle after viral infection. We further used the confocal immunofluorescence microscopy to test the colocalization of the nucleoprotein (NP) and cell nucleus. Interestingly, DXC-10 treatment inhibits the viral NP entry of the cell nucleus. However, DXC-10 had no effect on viral adsorption, viral RNA-dependent RNA polymerase (RdRp) and the endosomal pH. Further mechanisms study and in vivo efficacy and pharmacological studies will identify the potential preclinical candidates for development of the Tricyclic Matrinic derivatives as therapeutics of influenza infections in the future.

137. Dual Activation of IFN and Pro-Inflammatory Responses by TLR-Agonists Leads to a Strong Inhibition of HBV Replication

Julie Lucifora, PhD[1], Marc Bonnin, PhD[1], Sarah Maadadi, BS[1], Floriane Fusil, PhD[2], Laura Dimier, BS[1], Maud Michelet, MS[1], Océane Floriot, PhD[1], Anna Salvetti, PhD[1], Michel Rivoir, MD, PhD[3], Stephane Daffis, PhD[4], Simon Fletcher, PhD[4], François-Loïc Cosset, PhD[2], Fabien Zoulim, MD, PhD[5], **David Durantel, PhD**[1]

[1]Cancer Research Center of Lyon (CRCL), Inserm U1052, Lyon, France; [2]CIRI, INSERM U1111, Lyon, France; [3]CLB hospital, Inserm U1032, Lyon, France; [4]Gilead Sciences, Foster city, California, United States of America; [5]Hospices Civils de Lyon, CRCL, Inserm U1052, Lyon, France

Recent studies highlighted the therapeutic potential of agonists of innate immunity receptors/sensors. Our aims were to explore the anti-HBV potential of TLR agonists. Primary human hepatocytes or differentiated HepaRG cells (dHepaRG) were treated with TLR agonists once HBV replication was established. Antiviral activity was evaluated by quantification of viral parametres with standard aproaches. Among the different tested molecules, four ligands induced sustained reduction in all HBV parameters, including levels of cccDNA in HBV-infected dHepaRG and PHH. The maximal antiviral effect was obtained after Pam3CSK4 (TLR1/2-ligand) or poly(I:C) (TLR3-ligand) treatments without any viral rebound being observed after treatment arrest. Importantly, we also identified a novel TLR3 ligand, called riboxxol, presenting, as compared to poly(I:C), improved TLR3 specificity and biochemical characteristics allowing *in vivo* injection. This agonist demonstrated a strong anti-HBV effect in HBV-infected dHepaRG and PHH. Whereas, Pam3CSK4 and poly(I:C) respectively induced the expression of classical genes from the interferon or NF-kB pathway, riboxxol interestingly induced both. Moreover and contrary to TLR7-ligands that have been clinically studied, our ligands could theoretically activate responses in both hepatocytes and immune cells. We indeed observed an additional antiviral effect when hepatocytes where treated with riboxxol and conditioned medium generated by stimulation of PBMCs with the same ligand. Finally, we are currently investigating the antiviral effect of Pam3CS4K or riboxxol in HBV-infected HuHep mice or AAV-HBV transduced mice. In conclusion, our data highlight the potential of innate immunity activation in the control of HBV replication, and support the development of PRR-based antiviral strategies against HBV.

138. Two Birds with One Stone: Nucleoside Polymerase Inhibitors that Block the Replication of Both Noro- and Rotaviruses, the Two Main Etiological Agents of Viral Diarrhea

Jana Van Dycke, MS[1], Justine Vandepoele, MS[1], Guido Papa, MS[2], Francesca Arnoldi, PhD[2], Oscar Burrone, PhD[2], Johan Neyts, PhD[1], **Joana Rocha-Pereira, PhD**[1]

[1]Rega Institute for Medical Research, KU Leuven, Leuven, Belgium; [2]International Centre for Genetic Engineering and Biotechnology, Trieste, Italy

Noroviruses and rotaviruses are the two most common causes of severe childhood diarrhea, being responsible for ~800.000 deaths in children <5 years, each year. These occur mostly in developing countries, where children have multiple episodes of diarrhea/ year, leading to nutritional deficits and long-term consequences such as growth stunting. Having one single highly efficient antiviral treatment to rapidly treat such patients is highly desirable.

We here report that one single antiviral molecule can inhibit the replication of both noro- and rotavirus by targeting the viral polymerase. 7-deaza-2'-*C*-methyladenosine (7DMA) inhibits the *in vitro* replication of human norovirus and rotavirus with an EC_{50} of ~7µM and ~1µM, respectively; 2'-*C*-methylcytidine (2CMC) also has dual activity. The antiviral effect was confirmed by qRT-PCR and staining of viral antigens. Furthermore, the 5'-triphosphates of 2CMC and 2'-*C*-methyladenosine (2CMA) inhibited the transcription activity of purified rotavirus SA11 double-layered particles (EC_{50} 4 and 9µM, respectively) showing that these molecules act by inhibiting the rotavirus polymerase activity. We could earlier demonstrate the protective effect of 2-'C-methyl nucleosides in norovirus-infected mice and are now evaluating the potential *in vivo* efficacy of 7DMA against rotavirus infections in $IFNLR1^{-/-}$ C57/BL6 mice.

This is, to the best of our knowledge, the first time that a nucleoside analogue is shown to be active against a highly pathogenic dsRNA virus. Given the dual antiviral activity noro- and rotaviruses, this study opens the door for the development of a truly broad-spectrum antiviral to treat and prevent viral diarrhea in children.

139. Polo-Like-Kinase 1 is a Proviral Host-Factor for Hepatitis B Virus Replication and a Potential Target for Combined Antiviral Strategies

Adrien Foca, MS[1], Ahmed Diab, PhD[2], Floriane Fusil, PhD[3], Pascal Jalaguier, PhD[1], Nathalie Isorce, PhD[1], François-Loïc Cosset, PhD[3], Fabien Zoulim, MD, PhD[1], Ourania Andrisani, PhD[2], **David Durantel, PhD**[1]

[1]CRCL, Inserm U1052, Lyon, France; [2]Purdue University, West Lafayette, Indiana, United States of America; [3]CIRI, Inserm U1111, Lyon, France

A carcinogenetic link between HBV and PLK1 was previously established, but whether PLK1 has a role in HBV replication remained to be determined.

Primary human hepatocytes and differentiated HepaRG cells were used in this study as HBV infection models. Liver-humanized HBV-infected FRG mice were also used to demonstrate the efficacy/safety of PLK1 inhibitors in vivo.

First, we showed the early activation of PLK1 by phosphorylation in HBV-infected hepatocytes. Second, gain- and loss-of-function studies demonstrated PLK1 is a proviral factor in differentiated-hepatocytes. Next, the BI-2536 antiviral property was investigated and EC_{50} on the accumulation of HBV rcDNA were established at 5.0 and 50 nM respectively in dHepaRG and PHH. BI-2536 antiviral activity was further established in liver-humanized HBV-infected FRG mice. A mean reduction of 1.5-log_{10} in viremia was observed after 4 weeks of treatment in this model. Beside the potent inhibition of rcDNA synthesis, no effect on other viral parameters was observed, indicating a post-transcriptional mechanism. In fact BI-2536 prevented the formation of nucleocapsid, as revealed by immunofluorescence and capsid migration assays. Finally, *in vitro* PLK1 kinase assays demonstrated PLK1 could phosphorylate HBc, secondary to phosphorylation by primary kinases.

In this study, we demonstrated that PLK1 is a proviral host factor for HBV replication. In particular PLK1 activity seems to be important for the formation/stability of nucleocapsid and subsequent rcDNA synthesis. PLK1 inhibition represents a novel antiviral therapeutic strategy for suppressing HBV replication, which could be combined to nucleoside analogues and/or core inhibitors to further inhibit rcDNA synthesis and capsid recycling.

140. Recombinant, Fully-Human mAbs Have Potent Therapeutic Activity in Murine Models of Chikungunya Virus Disease

Marie Mandron, PhD[1], Jonathan Rothblatt, PhD[2], Pierre Cortez, PhD[1], Xavier Marniquet, PhD[1], Heather Hughes, PhD[3], Jooyun Lee, MD[4], Nicole Haese, PhD[5], Skot Neilson, PhD[6], Rebecca Broeckel, PhD[5], Gopal Sapparapu, PhD[7], Anna Park, PhD[3], Julie Bird, PhD[3], Cendrine Lemoine, PhD[1], Catherine Devaud, PhD[8], Anne Caron, PhD[8], Soila Sukupolvi, PhD[9], Julie Fox, PhD[9], Daniel Streblow, PhD[5], Justin Julander, PhD[6], Michael Diamond, MD, PhD[9], James Crowe, MD[7], **Kara Carter, PhD**[2]
[1]Sanofi, Marcy L'Etoile, France; [2]Sanofi, Cambridge, Massachusetts, United States of America; [3]Sanofi, Framingham, Massachusetts, United States of America; [4]Sanofi, Bridgewater, New Jersey, United States of America; [5]Oregon Health Sciences University, Beaverton, Oregon, United States of America; [6]Utah State University, Logan, Utah, United States of America; [7]Vanderbilt University, Nashville, Tennessee, United States of America; [8]Sanofi, Paris, France; [9]Washington University in St Louis, St Louis, Missouri, United States of America

Chikungunya virus (CHIKV) is an Old World Alphavirus that is currently undergoing an intense re-emergence in the Caribbean region, South America and Central America, also recently appeared in the US and Europe. CHIKV disease elicits incapacitating arthralgia that persists from months to years in over half of infected patients. Unfortunately, there are no FDA approved vaccines or antiviral treatments for CHIKV.. Previously, highly potent neutralizing monoclonal antibodies from a convalescent patient were identified. In the current study, recombinant versions of the most potent of these antibodies were generated to further the hypothesis that highly potent recombinant neutralizing antibodies can reduce viral burden at sites of infection and thus mitigate disease. In two independent murine models of CHIKV disease, the recombinant antibodies were able to significantly reduce viral titer in the joints of mice following establishment of disseminated infection in a dose dependent fashion. Developability assessment of these antibodies identified motifs potentially affecting stability which were then optimized to reduce this liability and maintain binding. Two lead antibodies were shown to be antigen specific with no human tissue binding. In vitro efforts to identify neutralization resistant viral isolates generated such mutants to one of the two lead antibodies, but failed to identify resistant mutants to the second lead antibody after three independent attempts. Given the pharmacological activity, antigen specificity, and favorable resistance profile of these antibodies, they are strong candidates to move forward to clinical therapeutic evaluation and further assessment of prophylactic activity in preclinical models.

141. A Novel Agonist of the TRIF Pathway Induces a Cellular State Refractory to Replication of Zika, Chikungunya, and Dengue Viruses

Kara Pryke, BS[1], Jinu Abraham, PhD[1], Tina Sali, PhD[1], Bryan Gall, PhD[1], Daniel Streblow, PhD[1], Alec Hirsch, PhD[1], Marita Chakhtoura, PhD[2], Elias Haddad, PhD[2], Jessica Smith, PhD[1],
Victor DeFilippis, PhD[1]
[1]Oregon Health and Science University, Portland, Oregon, United States of America; [2]Drexel University, Philadelphia, Pennsylvania, United States of America

The ongoing concurrent outbreaks of Zika, Chikungunya, and Dengue viruses in Latin America and the Caribbean highlight the need for development of broad-spectrum antiviral treatments. The type I interferon system has evolved in vertebrates to generate cellular and tissue responses that actively block replication of multiple known and potentially zoonotic viruses. As such its activation and control through pharmacologic agents may represent a novel therapeutic strategy for simultaneously impairing growth of multiple virus types and rendering host populations resistant to virus spread. In light of this we undertook a molecular screen to identify interferon-activating small molecules and now describe a compound we term "AV-C". Treatment of human cells with AV-C activates interferon-associated responses that strongly inhibit replication of Zika, Chikungunya, and Dengue viruses. Utilizing genome editing we investigated the host proteins essential to AV-C-induced cellular activity. This revealed that AV-C requires a TRIF-dependent signaling cascade that culminates in IRF3-dependent expression and secretion of type I interferon to elicit antiviral responses. The other canonical IRF3-terminal adaptor proteins STING and IPS-1/MAVS were dispensable for AV-C-induced phenotypes. Interestingly, this work also revealed an important biological role for IPS-1/MAVS, but not TRIF, in flavivirus replication in human cells implying that TRIF-directed viral evasion may not occur. Additionally, we show that human peripheral blood mononuclear cells treated with AV-C secrete proinflammatory cytokines that are linked with establishment of adaptive immune responses. Ultimately synthetic innate immune activators such as AV-C may serve multiple therapeutic purposes including direct antimicrobial responses and indirect facilitation of microbe-directed adaptive immunity.

142. Structural and Enzymatic Studies on Zika Virus NS2B-NS3 Protease

Linlin Zhang, PhD, Yasmin Gül, MS, Jian Lei, PhD, Rolf Hilgenfeld, PhD
Institute of Biochemistry, University of Lübeck, Lübeck, Germany

In addition to causing similar symptoms (rash, fever, and myalgia) as other flaviviruses, Zika virus (ZIKV) infection is related to serious neurological disorders including Guillain-Barré syndrome [1] and a significant number of microcephaly cases in fetuses and newborns [2]. In addition, mouse-model studies have shown that ZIKV can lead to testicular infection and epididymal damage, resulting in destruction of the seminiferous tubules and cell death [3,4].

Due to its essential role for processing the polyprotein, ZIKV NS2B-NS3 protease (NS2B-NS3^{pro}) is considered an attractive drug target. We have determined the crystal structure of ZIKV NS2B-NS3^{pro} in complex with a capped-dipeptide boronate inhibitor (K_i = 42 ± 5 nM), thereby providing a basis for the design of new inhibitors [5]. In addition, we investigated the kinetics of various recombinant constructs of ZIKV NS2B-NS3^{pro} against the substrate Bz-Nle-KKR-AMC and a series of octapeptide FRET substrates corresponding to the putative cleavage sites of the protease in the polyprotein. These studies demonstrate that ZIKV protease is hyperactive compared to the proteases of the related flaviviruses WNV and DENV-2.

REFERENCES

[1] Fontanet A., et al. 2016. Lancet 387, 2600.

[2] Rasmussen S. A., et al. 2016. N. Engl. J. Med. 374, 1981–1987.

[3] Govero J., et al. 2016. Nature 540, 438-442.

[4] Ma W., et al. 2016. Cell 167, 1511-1524.

[5] Lei J., et al. 2016. Science 353, 503-505.

143. Cyclin G Associated Kinase (GAK) Inhibition as a Strategy for the Discovery of Broad Spectrum Antivirals

Steven De Jonghe, PhD[1], Stefan Knapp, PhD[2], Piet Herdewijn, PhD[1], Shirit Einav, MD, PhD[3]
[1]Medicinal Chemistry, Rega Institute for Medical Research, KU Leuven, Leuven, Belgium; [2]Goethe-University Frankfurt, Institute of Pharmaceutical Chemistry, Frankfurt-am-Main, Germany; [3]Department of Microbiology and Immunology, Stanford University School of Medicine, Stanford, California, United States of America

Most of the currently available antiviral drugs target viral factors which are essential for the virus. However, targeting viral enzymes by monotherapy is often associated with the emergence of viral resistance. In addition, these drugs are limited by a narrow-spectrum of antiviral coverage (the so-called 'one drug-one bug' approach). An alternative strategy, which is less explored, is the development of compounds that target cellular host factors. This approach has the great advantage of offering a higher genetic barrier to resistance. In addition, different classes of viruses can depend on the same cellular factor for their replication. Hence, targeting such a common host factor with small molecules may lead to the discovery of broad-spectrum antivirals.

Recently, cyclin G associated kinase (GAK) emerged as a promising target for the treatment of hepatitis C (HCV) virus and other RNA virus infections. GAK is a host cell kinase known to regulate interactions between clathrin adaptor complexes and host cargo proteins. It has been shown that depletion of GAK by siRNA is dispensable for HCV RNA replication, but significantly inhibits two temporally distinct steps of the HCV life cycle: viral entry and infectious virus assembly. This prompted us to set up a program focusing on the identification of small-molecule GAK inhibitors. In this poster, the hit-to-lead optimization campaign and the antiviral activity against HCV, dengue virus and Ebolavirus of isothiazolo[4,3-b]pyridines as the first selective GAK inhibitors will be discussed.

144. ADAR1 Function Regulates Innate Immune Activation and HIV-1 Susceptibility in Primary Macrophages

Maria Pujantell, MS, Eva Riveira-Muñoz, PhD, Roger Badia, PhD, Bonaventura Clotet, MD, PhD, José Esté, PhD, Ester Ballana, PhD
AIDS Research Institute – IrsiCaixa, Badalona, Barcelona, Spain

HIV-1 infection induces innate intracellular antiviral defenses, aimed at restricting virus replication and spread. Controversial data exists regarding the capacity of HIV-1 infection to induce type I IFN and evade immune recognition in macrophages. Therefore, understanding the role and function of innate immune effectors and modulators can help to establish novel strategies for HIV-1 control. *ADAR1* knockdown (siADAR1) in primary monocyte-derived macrophages (MDM) led to a significant increase in *IFNB1* mRNA (7.5-fold, p=0.03) and CXCL10 gene (1000-fold, p=0.02) and protein expression (250-fold, p=0.01) compared to mock-transfected MDM, indicative of innate immune activation. siADAR1 MDM showed a significant reduction in HIV-1 infection either with a single cycle, VSV-pseudotyped NL4-3 GFP expressing virus (75% inhibition, p<0.0001) or a fully replicative R5 HIV-1 strain (BaL) (80% inhibition, p<0.0001). Proviral DNA formation or integration were not affected in siADAR1 MDM; however, a significant reduction in viral transcription was detected (75% reduction, p<0.0007). Although ADAR1 deaminase activity was detected in cellular genes, direct modification of viral RNAs by ADAR1 was not observed. siADAR1 MDM also showed upregulation of MDA5 (*IFIH1*), the cytoplasmic sensor of ADAR1-edited RNAs. Further characterization of innate immune pathways in siADAR1 MDM showed enhanced expression of the innate immune RNA sensor RIG-I, increased STAT1-phosphorylation and IRF7 expression, comparable to that observed after LPS or polyI:C treatment in mock-transfected MDM. *ADAR1* knockdown induces innate immune activation that renders macrophages resistant to HIV-1 infection, suggesting ADAR1 as a potential target to boost HIV-1 immune response.

145. Discovery of 2-Pyridinone Aminals: A Prodrug Strategy to Advance a Second Generation of HIV-1 Integrase Strand Transfer Inhibitors

Izzat Raheem, PhD
Merck, West Point, Pennsylvania

The search for new molecular constructs that resemble the critical 2-metal binding pharmacophore required for HIV integrase strand transfer inhibition represents a vibrant area of research within drug discovery. Efforts are focused on the development of inhibitors that display broad resistance profiles and are amenable to once-daily or less frequent dosing. Herein we present the discovery of a novel class of HIV integrase strand transfer inhibitors (InSTI) based on the 2-pyridinone core of MK-0536. These efforts led to the identification of two lead compounds with excellent antiviral activity and preclinical pharmacokinetics to support a once-daily human dose prediction. Dose escalating PK studies in dog revealed significant issues with limited oral absorption and required an innovative prodrug strategy to enhance the high-dose plasma exposures of the parent molecules. Our strategy relied on functionalizing two different accessible hydroxyl groups on the leading compounds to introduce promoieties, with prodrugs designed to independently address the limited aqueous solubility and limited permeability of parent compounds. Our efforts culminated in a unique phosphate/carbonate acetal "double–prodrug" that achieved high parent plasma exposures and provided a path to further advance this new class of InSTI.

146. Lability of the Favipiravir Ribonucleoside and First Mechanistic Details

Johanna Huchting, PhD, Matthias Winkler, MS, Hiba Nasser, MS, Chris Meier, PhD
Hamburg University, Hamburg, Germany

The antiviral agent T-705 (favipiravir) has been extensively studied with regard to its activity profile and mechanism-of-action. As a nucleobase analogue, it is first phosphoribosylated intracellularly by the host cell enzyme *hypoxanthine guanine phosphoribosyl transferase*. Further phosphorylation yields the active compound, T-705-ribonucleoside triphosphate. Albeit reports on the synthesis of the T-705-ribonucleoside and -tide can be found in the (patent-) literature, our attempts to reproduce the reported protocols revealed considerable problems in the synthesis.

Therefore, after developing a reliable synthetic strategy to obtain this ribonucleoside analogue, its remarkable low stability was studied in detail. This revealed the high tendency of T-705-ribonucleoside towards nucleophilic displacement of the fluorine. Ultimately, this leads to a breakdown of the aromatic system as will be presented.

To gain a deeper understanding of the underlying processes, this degradation was studied in different buffers as well as in non-buffered aqueous solution, reflecting the simplest and mildest conditions as they're also applied for biochemical studies. T-705-ribonucleoside proved labile under all studied conditions. However, an increase of the pH strongly accelerated the observed decomposition. The application of different spectroscopic and spectrometric techniques enabled us to propose mechanistic details of this decomposition which will be discussed.

The results presented here are of huge relevancy especially for those using T-705 as an antiviral agent or in synthetic chemistry.

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147. Tropolones Powerfully Suppress Herpesvirus Replication: Preliminary Structure-Activity Relationship and Inhibition of Acyclovir-Resistant Viruses

Bindi Patel, BS[1], Aswin Garimallaprabhakaran, PhD[2], Alex Berkowitz, BS[2], Nana Agyemang, MS[2], Andreu Gazquez, BS[1], Peter Ireland, MD[1], Mark Cadiz, BS[1], John Tavis, PhD[1], Ryan Murelli, PhD[2], **Lynda Morrison, PhD**[1]
[1]*Saint Louis University School of Medicine, St. Louis, Missouri;* [2]*The Graduate Center, City University of New York, New York, New York*

Herpesvirus DNA replication requires several enzymes in the nucleotidyl transferase superfamily (NTS) that have recombinase and nuclease activities. Our previous data showed that compounds which block NTS enzymes efficiently inhibit replication of herpes simplex virus (HSV)-1 and HSV-2 clinical isolates, and human cytomegalovirus. We previously identified 15 tropolones that reduced HSV replication by 3 to 6 $\log_{10}$ at 5 μM in Vero cells and human foreskin fibroblasts. Tropolones also profoundly inhibited replication of acyclovir (ACV)-resistant mutants. The most effective synthetic tropolone had a 50% effective concentration (EC_{50}) of 80 nM and a selectivity index >1235. Preliminary structure-function analysis suggested that an intact -hydroxytropolone moiety on the cycloheptatriene ring was advantageous for potency. To gain further structure-function insight, 43 new ahydroxytropolones were synthesized and tested for HSV replication inhibition to probe the nature and positioning of effective appendages. The most potent of these molecules shared a common 4-biphenyl appendage at the R^2 position of the ring. Most of these new molecules suppressed HSV replication better than ACV at 5 μM, and viral resistance to ahydroxytropolones evolves much more slowly than against ACV. We have defined a preliminary structure-activity relationship for the tropolones, and determined that select -hydroxytropolones apply a higher barrier to evolution of viral resistance than ACV. This property, and their different mechanism of action than the nucleoside analogs, suggests that NTS enzyme inhibitors are promising candidates for development into highly effective treatments for herpesvirus infections as mono- or combination therapies, or as salvage therapies for patients with ACV-resistant infections.

148. Post-Exposure Administration of USC-087 Protects Immunosuppressed Syrian Hamsters Against Lethal Challenge with Human Species C Adenoviruses

Karoly Toth, DVM[1], Jacqueline Spencer, BS[1], Baoling Ying, MD[1], Ann Tollefson, PhD[1], Caroll Hartline, PhD[2], Jiajun Fan, BS[3], Jinglei Lyu, BS[3], Boris Kashemirov, PhD[3], Mark Prichard, PhD[2], William Wold, PhD[1], Charles McKenna, PhD[3]

[1]Saint Louis University School of Medicine, St. Louis, Missouri, United States of America; [2]University of Alabama at Birmingham, Birmingham, Alabama, United States of America; [3]University of Southern California, Los Angeles, California, United States of America

Human adenoviruses (AdV) cause generally mild infections of the respiratory and GI tracts as well as some other tissues. However, certain types (serotypes) can cause serious acute respiratory disease and epidemic keratoconjunctivitis. Also, AdV can cause serious infection in severely immunosuppressed individuals, especially pediatric patients undergoing allogeneic hematopoietic stem cell transplantation, where mortality rates are up to 80% with disseminated disease. Despite the seriousness of AdV disease, there are no drugs approved specifically to treat AdV infections. Cidofovir, an analogue of cytidine phosphate which functions as an AdV DNA chain terminator, is used in many clinics, but controlled studies have not been done, and cidofovir can be toxic to the kidney. We report here that USC-087, an *N*-alkyl tyrosinamide ester prodrug of HPMPA, the adenine analog of cidofovir, is effective against multiple AdV types in cell culture. USC-087 is also effective against AdV-C6 in our immunosuppressed permissive Syrian hamster model. In this model, hamsters are immunosuppressed by treatment with high dose cyclophosphamide. Injection of AdV-C6 (or AdV-C5) intravenously leads to a disseminated infection that resembles the disease seen in humans, including death. We have tested the efficacy of orally-administered USC087 against the median lethal dose of intravenously administered AdV-C6. USC087 completely prevented or significantly decreased mortality when administered up to 4 days post challenge. USC-087 also prevented or significantly decreased liver damage caused by AdV-C6 infection, and inhibited virus replication even when administered 4 days post challenge.

149. Inhibitors of Emerging Flaviviruses

Radim Nencka, PhD[1], Hubert H ebabecký, PhD[1], Michal Šála, PhD[1], Milan Dejmek, PhD[1], Evzen Boura, PhD[1], Kamil Hercik, PhD[1], Daniel R žek, PhD[2], Lud k Eyer, PhD[2]

[1]IOCB Prague, Prague, Czech Republic; [2]Veterinary Research Institute, Brno, Czech Republic

Flavivirus, a genus of *Flaviviridae* family, comprise numerous important human pathogens, which are transmitted by arthropods such as mosquitoes and ticks. Zika virus (ZIKV), a mosquito-borne flavivirus, has recently gained a significant attention mainly due to its outbreak in South America. ZIKV infections are usually asymptomatic or accompanied only by mild symptoms, e.g. rash and arthralgia. In some cases, however, the infection results in encephalitis, myelitis or Guillain-Barre syndrome. Most importantly, ZIKV infection during pregnancy seems to be associated with fetus malformations resulting in congenital microcephaly. West Nile virus (WNV) also belongs to mosquito-borne flaviviruses with similar occurrence of symptomatic manifestation of the disease as in the case of ZIKV. The symptoms include fever with arthralgia, vomiting or diarrhea. Approximately 1% of infected people develop neurologic symptoms that lead to death in up to 10% of cases. Finally, tick-borne encephalitis virus (TBEV), unlike its two forenamed siblings, is transmitted solely by ticks. The TBEV infection is characterized by encephalitis, meningitis or meningoencephalitis with 1-2% mortality. Our team has been interested in the development of novel nucleoside and nucleotide derivatives against these emerging arboviruses and we have recently evaluated a number of such compounds for their efficacy against ZIKV, WNV and TBEV. In addition, we have prepared triphosphate derivatives of selected known nucleosides and examined their inhibitory activity on the ZIKV RNA-dependent RNA polymerase. Based on these data, we have designed and synthesized novel nucleoside derivatives with potential activity against these dangerous human pathogens.

150. HBV RNaseH Inhibitors: Lack of Sensitivity to Viral Genetic Variation, Synergy with Approved and Experimental Drugs, and *In Vivo* Efficacy in FRG Chimeric Mice

Elena Lomonosova, PhD[1], Kelly Long, MS[2], Qilan Li, PhD[1], Nathan Ponzar, BS[1], Juan Villa, PhD[1], Ryan Murelli, PhD[3], John Bial, PhD[4], John Sagartz, DVM, PhD[2], **John Tavis, PhD**[1]

[1]Saint Louis University School of Medicine, St. Louis, Missouri, United States of America; [2]Seventh Wave Laboratories, St. Louis, Missouri, United States of America; [3]City University of New York, New York, New York, United States of America; [4]Yecuris, Inc., Tualatin, Oregon, United States of America

We identified all 77 known inhibitors of the Hepatitis B Virus (HBV) ribonuclease H (RNaseH), primarily among the -Hydroxytropolones (HT), N-Hydroxyisoquinolinediones (HID), and N-Hydroxypyridindiones (HPD). The best inhibitors include the HTs #110 and 46, an HID (#1) and an N-Hydroxypyridinedione (#208). Here, we evaluated key parameters that would impact RNaseH inhibitors as drug candidates.

Effects of HBV's high genetic variation on inhibitor efficacy were evaluated by screening compounds #1 and 46 against RNaseH variants from genotypes B, C, and D. There was a large range of basal RNaseH activity, but all variants were equivalently sensitive to compounds #1 and 46. Synergy against viral replication in cells was evaluated with compounds #1 and #46 against Lamivudine, the capsid assembly modifier HAP12, and each other. The RNaseH inhibitors were synergistic with Lamivudine and each other. Both RNaseH inhibitors were additive with HAP12, and cytotoxicity was not enhanced by any compound combination. Finally, we tested efficacy of #110 and #208 against HBV replication in FRG chimeric mice. Compound #208 inhibited HBV viremia by 1.3 log ($p = 0.00003$) after two weeks of treatment, and #110 suppressed viremia by 0.4 log ($p=0.001$). Weight loss, partially due to the vehicle, was substantial on treatment.

These studies indicate that anti-RNaseH drug discovery is unlikely to be hampered by differential sensitivity of HBV strains to the inhibitors and that RNaseH antagonists could be productively employed in combination with nucleos(t)ide analogs or capsid assembly modifiers. The *in vivo* studies directly validate the RNaseH as a drug target.

151. KPT-335, a Selective Inhibitor of Nuclear Export (SINE) Compound, Modulates Respiratory Syncytial Virus (RSV) Matrix Protein Nuclear Trafficking and Immune Responses

Jennifer Pickens, PhD[1], Sharon Tamar, PhD[2], Margaret Lee, PhD[2], Ralph Tripp, PhD[1]

[1]University of Georgia, Athens, Georgia, United States of America; [2]Karyopharm Therapeutics, Newton, Massachusetts, United States of America

Respiratory syncytial virus (RSV) is the a primary cause for hospitalization of children in the US. Children born prematurely are at high risk for infection, resulting in subtantial morbity and mortality . There is no licensed RSV vaccine or effective treatment available. There is a need for safe and effective antiviral therapies to prevent and/or reduce RSV infections. A novel class of orally available small molecule inhibitors (XPO1;Exportin 1) have recently been developed and one compound, verdinexor KPT-335, has shown to be efficacious at inhibiting influenza A/B strains and providing protection against disease pathology *in vivo*. In the current studies, we are examining the mechanism of action of KPT-335 against RSV. Our focus is to better understand the RSV matrix (M)-host interaction within the nuclear compartment. RSV M is a critical mediator of RSV replication and viral egress, and is the only viral protein trafficked within the nucleus through the action of Importin b1 and XPO1, respectively. Treatment of RSV with KPT-335 reduced XPO1 expression while increasing p53 expression, but interestly, there was no induction of the caspase 3/7 apoptotic pathway. Furthermore, KPT-335 treatment resulted in the nuclear accumulation of the RSV M protein and the NF-kB p65 subunit in RSV infected cells, resulting in varying pro-inflammatory cytokine expression. This work provides preliminary edvidence outlining the mechanism of action of KPT-335 against RSV, where nuclear trapping of the RSV M reduces viral replication and egress, while modulating the host immune response independent of the p53 apoptic pathway.

152. Engineering Approaches to Combat Infectious Diseases: An Example of Broad-Spectrum Antiviral Peptides

Nam-Joon Cho, PhD
School of Materials Science and Engineering, Nanyang Technological University, Singapore

Infectious diseases represent one of the leading causes of worldwide morbidity and mortality, with their emergence, re-emergence, and potential application as bio-terror agents all serious public health and security concerns. While there have been important advances in antiviral drug development over the past few decades, there remains an urgent need to develop new classes of antiviral agents. One promising antiviral target is the lipid envelope surrounding a wide range of medically important viruses, although its selective targeting is difficult to achieve. By utilizing engineering approaches, we have addressed this need by developing a broad-spectrum antiviral peptide that acts by selectively destabilizing lipid membranes with high-curvature membrane architectures, including small, spherical particles (*e.g.*, dengue) and filamentous particles (*e.g.*, Ebola). The peptide exhibits highly potent *in vitro* antiviral activity against multiple virus families and has a therapeutic index around 1,000. *In vivo* experiments in a humanized dengue mouse model demonstrate that treatment with the peptide significantly reduces viral titer in the bloodstream. The findings support that viral membrane targeting holds excellent potential for the treatment and prevention of virus infections and highlight the potential of engineering approaches to combat infectious diseases.

153. Design, Synthesis and Anti-RNA Virus Activity of Novel Fluorocarbocyclic Nucleosides

Lak Shin Jeong, PhD[1], Jiseong Yoon, MS[1], Young Sup Shin, BS[1], Dnyandev Jarhad, PhD[1], Kristina Kovacikova, PhD[2], Clara Posthuma, MS[2], Eric Snijder, MD, PhD[2], Martijn van Hemert, MD, PhD[2]
[1]Seoul National University, Seoul, Korea, Republic of; [2]Leiden University

Chikungunya virus (CHIKV) is a mosquito-borne alphavirus that causes an acute febrile illness and a severe arthralgia that may persist for months. CHIKV re-emerged in 2004 and has since spread in more than 60 countries worldwide, most recently in the Americas. Zika virus (ZIKV) is also a mosquito-borne virus and spread by daytime-active Aedes mosquitoes, leading to the 2015-2016 Zika virus epidemic. However, the current lack of specific treatment stresses the importance of developing inhibitors with the potential to advance into clinical development. Thus, in order to develop novel antiviral agents targeting a chikungunya virus, we designed the novel fluorocarbocyclic nucleosides based on the potent anti-RNA virus activity of aristeromycin and assyed them for antiviral activity. Among compounds tested using cytopathic effect (CPE) reduction assays, several compounds inhibited CHIKV and ZIKV in the sub-micromolar range (EC_{50} 0.2 – 0.3 μM) with high selective indices.

To elucidate the mode of action of these compounds, we also assayed them for the inhibitory activity of cellular S-adenosylhomocysteine hydrolase. As expected, most of active compounds exhibited potent inhibitory activity in the sub-micromolar range, demonstrating that the antiviral activity is correlated with the inhibitory activity of S-adenosylhomocysteine hydrolase. In addition, these compounds seem to have a direct inhibitory effects on CHIVK and ZIKV RNA polymerases.

Synthesis and anti-RNA virus activity of novel nucleoside analogues will be presented in detail.

154. Inhibiting Viral RNA Replication and Enhancing Host Cellular Cytokine Response: Unique Dual Effects of a Benzodiazepine Yellow Fever Virus (YFV) NS4B Inhibitor

Xuexiang Zhang, MS, Shuo Wu, PhD, Julia Ma, BS, Fang Guo, MD, PhD, Yanming Du, PhD, Timothy Block, PhD, Ju-Tao Guo, MD, **Jinhong Chang, MD, PhD**
Baruch S. Blumberg Institute

We reported previously a benzodiazepine compound BDAA that specifically inhibited YFV replication and significantly protected the lethality of YFV-infected hamsters (Guo F. et al. J. Virol 90:10774, 2016). Genetic mapping of BDAA-resistant mutation at proline 219 of nonstructure protein 4B (NS4B) strongly suggests that NS4B protein is the molecular target of BDAA. In our continuing efforts toward understanding the antiviral mechanism, we obtained evidence showing that BDAA specifically enhanced the host cellular cytokine response induced by YFV, but not other viruses. Interestingly, a time-of-addition experiment clearly indicated that BDAA enhancement of the cytokine response occurred only after the onset of YFV RNA replication. In order to investigate the relationship between BDAA targeting of NS4B and its enhancement of cytokine response in YFV infected cells, HEK293 cells were infected with wild-type or BDAA-resistant YFV bearing NS4B P219S, P219T or P219A mutation and followed by treatment with a serial concentration of BDAA for 48 h. The study revealed that the BDAA concentration for maximal enhancement of the cytokine response was always around its EC_{50} of inhibition of viral RNA replication, that is, higher concentrations were required in cells infected with BDAA-resistant YFVs. Hence, the dual effects of BDAA on YFV RNA replication and induction of cytokine response are most likely resulted from a single action of BDAA on YFV NS4B protein, which may not only disrupt the integrity of membrane-associated YFV RNA replication complex and suppress viral replication, but also increase the exposure of viral RNA for enhanced innate cytokine response.

155. The Nucleoside Prodrug GS-5734 Inhibits Multiple Coronaviruses and Selects for Resistance Mutations in the RNA-Dependent RNA Polymerase that are Associated with a Decrease in Viral Replication Fitness

Maria Agostini, BS[1], Erica Andres, BS[1], Xiaotao Lu, BS[1], Amy Sims, PhD[2], Rachel Graham, PhD[2], Timothy Sheahan, PhD[2], Everett Smith, PhD[3], James Case, BS[1], Joy Feng, PhD[4], Robert Jordan, PhD[4], Adrian Ray, PhD[4], Tomas Cihlar, PhD[4], Dustin Siegel, PhD[4], Richard Mackman, PhD[4], Michael Clarke, PhD[4], Ralph Baric, PhD[2], Mark Denison, MD[1]
[1]Vanderbilt University Medical Center, Nashville, Tennessee; [2]University of North Carolina at Chapel Hill, Chapel Hill, North Carolina; [3]The University of the South, Sewanee, Tennessee; [4]Gilead Sciences, Inc., Foster City, California

The emergence of SARS-coronavirus (CoV) in 2002 and the continued circulation of MERS-CoV emphasize the capacity of CoVs to cause new zoonotic infections with pandemic potential. Despite the high mortality rates of these infections, no therapeutics or vaccines against CoVs are currently available. GS-5734, a prodrug of an adenine C-nucleoside analog GS-441524 with known activity against filoviruses, is currently undergoing clinical development for the treatment of Ebola virus infection. Here, we demonstrate that GS-5734 is also highly active (EC_{50} 0.02-0.07 mM) against multiple CoVs, including SARS, MERS, and murine hepatitis virus (MHV), and exhibits selectivity indexes (SI) of 135-1600 in human and murine cell lines. The nucleoside GS-441524 also exhibits anti-CoV activity, but is ~3-19-fold less potent. Passage of MHV in the presence of increasing concentrations of GS-441524 selected two nonsynonymous mutations (F476L and V553L) within the predicted fingers domain of RNA-dependent RNA polymerase (nsp12-RdRp). These mutations emerged over 23 passages, confer 4-6 fold resistance to GS-5734, and confer reduced fitness compared to WT in direct competition experiments. Our results are consistent with a mechanism of action of GS-5734 targeting the RdRp, potentially through the incorporation of active nucleoside triphosphate into viral RNA. In summary, GS-5734 is broadly active against diverse CoVs, exhibits a high barrier to development of resistance in vitro, and has a fitness cost associated with resistance mutations. These features support the development of GS-5734 as an effective antiviral therapeutic against known and emerging CoVs.

156. Repurposing Kinase Inhibitors Against Influenza – The Clinically Approved MEK Inhibitor Trametinib Efficiently Blocks IAV Propagation and Limits Hyperexpression of Cytokines

Tobias Schräder, PhD[1], Sabine Dudek, PhD[1], Christina Ehrhardt, MD, PhD[1], Oliver Planz, PhD[2], Stephan Ludwig, PhD[1]

[1]Institute of Virology (IVM) Westfaelische Wilhelms-University Muenster, Muenster, Germany; [2]Interfaculty Institute for Cell Biology, University of Tuebingen, Germany, Tuebingen

Influenza A virus (IAV) infections are still a major global threat for humans. The currently licensed anti-viral drugs target viral factors and are prone to provoke viral resistance. Thus there is an urgent need for effective antivirals to fight IAV. In infected host cells IAV induces various cellular signaling cascades. We have shown previously that the Raf/MEK/ERK signaling cascade is indispensable for IAV replication because it triggers the nuclear export of newly assembled viral ribonucleoproteins (vRNPs). Inhibition of this cascade limits viral replication. Thus, next to their potential in anti-tumor therapy, inhibitors targeting the Raf/MEK/ERK signaling cascade came into focus as potential anti-viral drugs. The first licensed MEK inhibitor Trametinib (GSK-1120212) is used for treatment of malignant melanoma, being highly selective and having a very promising side effect profile. Since Trametinib may be qualified for a repurposing approach that would significantly shorten development time for an anti-flu use, we evaluated its anti-viral potency and mode of action. In this study, we describe that Trametinib efficiently blocks replication of different influenza subtypes *in vitro* and *in vivo*. The broad anti-viral activity against various IAV strains was due to its ability to interfere with export of progeny vRNPs from the nucleus. The compound also limited hyper-expression of several cytokines. Thus, we show for the first time that a clinically approved MEK inhibitor acts as a potent anti-influenza agent.

157. Discovery and Mechanistic Study of Benzamide Derivatives that Modulate Hepatitis B Virus Capsid Assembly

Shuo Wu, PhD, Qiong Zhao, PhD, John Kulp, PhD, Timothy Block, PhD, Yanming Du, PhD, Jinhong Chang, PhD, **Ju-Tao Guo, MD**

Baruch S. Blumberg Institute

Chronic hepatitis B virus (HBV) infection is a global public health problem. Although the currently approved medications can reliably reduce the viral load and prevent the progression of liver diseases, they fail to cure the viral infection. In an effort toward discovery of novel antiviral agents against HBV, we identified a group of benzamide (BA) derivatives that significantly reduced the amount of cytoplasmic HBV DNA. Our initial lead optimization efforts identified two BA derivatives with improved antiviral activity for further mechanistic studies. Interestingly, similar to our previously reported sulfamoylbenzamide derivatives (SBAs), the BAs promote the formation of empty capsids through specific interaction with HBV core protein, but not other viral and host cellular components. We also obtained genetic evidence suggesting that both SBAs and BAs inhibited HBV nucleocapsid assembly by binding to the "HAP" pocket between core protein dimer-dimer interfaces. However, unlike SBAs, BA compounds uniquely induce the formation of empty capsids that migrate slower in native agarose gel electrophoresis from A36V mutant core protein. Moreover, we showed that wild-type core protein confers susceptibility of chimeric capsid assembly with drug resistant core proteins to multiple capsid assembly modulators. Hence HBV core protein is a dominant antiviral target that may suppress the selection of drug resistant viruses during antiviral therapy. Our studies thus indicate that BAs are a chemically and mechanistically unique type of HBV capsid assembly modulators and warranted for further development as antiviral agents against HBV.

158. Transchromosomatic Bovine Immunoglobulin – A Novel Platform for the Rapid Response to Emerging Pathogens with Demonstrated Efficacy in Nonhuman Primates Against Ebola Virus

Richard Bennett, PhD[1], Dawn Gerhardt, MS[1], Sri Yellayi, DVM, PhD[1], Nick Oberlander, BS[1], Hua Wu, PhD[2], Jin-An Jiao, PhD[2], Gene Olinger, PhD[1], Tom Luke, MD, PhD[3], Gale Smith, PhD[4], Greg Glenn, PhD[4], David Flyer, PhD[4], Anna Honko, PhD[1], Lisa Hensley, PhD[1], Eddie Sullivan, PhD[2], Peter Jahrling, PhD[1]

[1]NIAID/NIH, Frederick, Maryland, United States of America; [2]SAB, Sioux Falls, South Dakota, United States of America; [3]NMRC, Silver Spring, Maryland, United States of America; [4]Novavax, Inc, Gaithersburg, Maryland, United States of America

The size of the recent Ebola epidemic in West Africa was unprecedented and highlights the enormous challenges in responding to a new and/or emerging infectious diseases with sufficient quantities of vaccines and therapeutics. Passive immunotherapy can effectively prevent and/or treat multiple bacterial, toxin mediated, and viral diseases such as Ebola. However, current passive immunotherapeutic options can be limited in availability and therapeutic benefit due to cultural, scientific, and production issues which can negatively impact the ability to rapidly respond to outbreaks of disease.

Here we describe the *in vitro* and *in vivo* assessment of fully-human polyclonal antibodies produced in transchromosomic bovines vaccinated with an Ebola virus Makona recombinant glycoprotein nanoparticle vaccine. Treatment of rhesus macaques with a low dose (57 mg/kg) immunoglobulin 1 day after inoculation with a lethal dose of Ebola virus (1000 PFU) resulted in 33% survival (2 of 6). By increasing the dose to 150 mg/kg and starting treatment 1 or 3 days after inoculation, 100% of the treated animals survived and did not develop signs of EVD (6 of 6 in both groups). Treating animals with a high dose during late stage disease (day 5) did not prevent EVD (0 of 6 survived), suggesting either additional immunoglobulin is required at this time point or a synergistic therapy could be required during late stage disease. This proof of concept study demonstrates the potential of this anti-Ebola therapeutic, and also suggests the applicability of this platform to rapidly produce potent passive immunotherapeutics for other emerging infectious diseases.

159. Can Antiviral Drug-Resistant Chikungunya Viruses be Transmitted by Mosquitoes?

Leen Delang, PhD[1], Pei-Shi Yen, MS[2], Marie Vazeille, PhD[2], Anna-Bella Failloux, PhD[2]

[1]Rega Institute for Medical Research, KU Leuven, Leuven, Belgium; [2]URE Arboviruses and Insect vectors, Institut Pasteur, Paris, Paris, France

The chikungunya virus (CHIKV) is transmitted by female *Aedes aegypti* and *albopictus* mosquitoes, mostly present in (sub)tropical regions. No antivirals are available to treat CHIKV infections. Several molecules with anti-CHIKV activity were recently discovered and for some of these, resistant CHIKV variants could be selected in cell culture. However, no information is available about the replication and transmission abilities of these antiviral-resistant viruses in mosquitoes. Therefore, we orally infected *Aedes aegypti* Paea mosquitoes with an artificial blood meal containing either WT CHIKV or resistant CHIKV variants (i.e. MADTP^{res} CHIKV: mutation in nsP1 gene, and favipiravir^{res} CHIKV: mutation in RdRp gene). At different days post-infection (pi), viral loads were quantified in bodies (infection), heads (dissemination) and saliva (transmission) of individual mosquitoes.

The infection rate of the resistant viruses was mostly similar to that of WT. In contrast, the dissemination of favipiravir^{res} CHIKV was markedly decreased as compared to WT and MADTP^{res} CHIKV. Furthermore, favipiravir^{res} CHIKV was only transmitted in the saliva at day 20 pi in contrast to WT (transmission starting at d3 pi). Similar results were obtained in field-collected species (Turkey and France). In body samples of mosquitoes infected with the resistant variants, the virus was still resistant to the antiviral, even at day 20 pi (fold resistance favipiravir^{res}=12, MADTP^{res}=5-40). Deep sequencing of saliva samples is ongoing to determine the presence of resistance mutations. Our results thus indicate that some antiviral-resistant arboviruses can efficiently replicate and disseminate in mosquitoes and that these viruses retain their resistant phenotype in the mosquito.

160. Development of ALS-8112/ALS-8176 as an Effective Replication Inhibitor of Human Metapneumovirus

Jia Meng, PhD, Jerome Deval, PhD, Andreas Jekle, PhD, Julian Symons, PhD
Alios BioPharma., Inc, part of the Janssen Pharmaceutical Companies, South San Francisco, California, United States of America

Human metapneumovirus (hMPV) is the second leading cause of pediatric lower respiratory tract infection. ALS-8112 is the parent molecule of ALS8176 (JNJ64041575), a first-in-class nucleoside prodrug inhibitor of human respiratory syncytial virus (RSV) replication currently under clinical evaluation. When tested in tissue culture against other RSV-related respiratory viruses, ALS-8112 also demonstrates potent activity against various strains of hMPV with assay dependent EC_{50} values ranging from 0.035-0.5 μM. The nucleoside triphosphate form of ALS-8112 (ALS-8112-TP) was recognized as an efficient substrate by the hMPV polymerase complex. The level of discrimination of ALS-8112-TP against natural CTP is within 2-fold between hMPV and RSV polymerases. Once incorporated into the viral genome, ALS-8112 caused chain termination of viral RNA synthesis. In order to understand the resistance profile of hMPV polymerase upon exposure to ALS-8112, CAN97-83 and TN/93-32 (representing A2 and B2 subtypes respectively) were passaged in the LLC-MK2 cell line either in the presence of ALS-8112 or DMSO alone as a control. A luciferase-based hMPV mini-genome system was developed using published sequence from the CAN97-83 strain. Codon-optimized N, P, L, and M2-1 plasmids were co-transfected with the firefly luciferase plasmid flanked by the hMPV regulatory sequences into HEp-2 cells co-infected with MVA-T7. The signal-to-noise ratio was about 200-fold. When tested in the mini-genome system, ALS-8112 exhibits similar potency against hMPV to RSV with an EC_{50} of 0.2±0.03 μM. In summary, the results of these studies support the further evaluation of ALS-8176 for the treatment of hMPV infection in both pediatric and adult patients.

161. Antifungal Azoles that Target an Early Stage of the Parechovirus A3 Life Cycle

Eric Rhoden, BS[1], Allan Nix, BS[1], William Weldon, PhD[1], Laurence Briesach, MS[2], Rangaraj Selvarangan, PhD[3]
[1]Centers for Disease Control and Prevention; [2]IHRC, contracting agency to the Centers for Disease Control and Prevention; [3]Children's Mercy Hospital

Parechovirus A3 (Par-A3, formerly human parechovirus 3) is known to cause severe illness in young infants, including sepsis, meningitis and encephalitis. We identified two FDA-approved drugs, itraconazole (ITZ) and posaconazole (PSZ) as potent broad-spectrum inhibitors of enterovirus (EV) and Par-A3. Currently, there is no approved antiviral treatment for Parechovirus species A (Par-A) infection, highlighting the need for antiviral therapy.

We performed *in vitro* mechanism of action studies to identify the stage in the Par-A3 life cycle at which these drugs exert their antiviral effect. Antiviral activity was assessed in 96-well homogeneous cross-titration and synchronized infection cell-based assays that measured inhibition of viral cytopathic effect (CPE) in Vero cells. These assays utilized differing compound addition, dilution and temperature shifts to evaluate antiviral effect on pretreatment, coaddition, postinfection, inactivation, attachment and entry. Additionally, antiviral compounds enviroxime (PI4KIIIβ inhibitor) and 25-hydroxycholesterol (25HC; oxysterol-binding protein [OSBP] inhibitor) were evaluated against Par-A1 through A6 and strains representing EV species A-D.

ITZ, PSZ, enviroxime and 25HC had broad-spectrum EV A-D antiviral activity, consistent with a similar mechanism of action against EV. Only ITZ and PSZ were identified as specific inhibitors of Par-A3 activity, suggesting a different and yet unknown mechanism for Par-A3 antiviral activity. ITZ and PSZ target an early stage of the Par-A3 life cycle, exerting antiviral effects during compound pretreatment, coaddition, cell-free virus-compound preincubation, inactivation and attachment. These results support the further evaluation of the use of ITZ and PSZ as antivirals against Par-A3.

162. Experience of a Translational Research Platform for the Evaluation of Human Cytomegalovirus (HCMV) Drug-Resistance in Belgium

Graciela Andrei, PhD, Sarah Gillemot, MS, Robert Snoeck, MD, PhD
Rega Institute – KU Leuven, Leuven, Belgium

Drug-resistance in HCMV is virtually not observed in immunocompetent individuals but it is a well-recognized problem among immunocompromised patients. Therefore, in 2009 a Reference and Service Center, RegaVir [**Re**search **G**roup for **A**nti**vi**ral **R**esistance, (www.regavir.org)], for the diagnosis and typing of drug-resistant herpesviruses was established in Belgium.

Genotyping [PCR amplification of the HCMV genes UL97 (protein kinase, PK) and UL54 (DNA polymerase, DP) followed by capillary (Sanger) sequencing] are used to diagnose resistance to the anti-HCMV drugs ganciclovir, cidofovir and foscavir. To date, we have analyzed 628 clinical samples recovered from patients with refractory HCMV disease. 536 out of 628 samples could be genotyped, 370 being wild-type. Mutations known to be linked to drug-resistance were found in 27.6% of the genotyped samples: 105 samples had mutations in the PK, 24 in the DP and 12 samples bore mutations both in the PK and DP genes. Novel mutations of unknown significance in the PK and/or DP genes were found in 35 samples.

Our data showed a) the usefulness of rapid HCMV genotyping for antiviral therapy adjustment, b) emergence of multiple drug-resistance due to infection with a virus having a single mutation conferring resistance to ganciclovir, cidofovir and foscavir (e.g. DNA pol mutations A834P and del 981-982) or caused by co-infection with viruses having distinct genotypes, c) viral compartmentalization, d) advantage of next generation sequencing for detecting minor populations of drug-resistant viruses, e) emergence of resistance to the investigational drug maribavir, f) urgent need for development of novel anti-HCMV agents.

163. Antiviral Activity and Mechanism of Action of Site-1 Protease (S1P) Inhibitor on Crimean-Congo Hemorrhagic Fever Virus.

Éric Bergeron, PhD, Stephen Welch, PhD, Mike Flint, PhD, Stuart Nichol, PhD, Christina Spiropoulou, PhD
Centers for Disease Control and Prevention

Crimean-Congo Hemorrhagic Fever virus (CCHFV) is a nairovirus (family *Bunyaviridae*) that causes a lethal hemorrhagic disease in humans. Specific and effective therapeutics and vaccines are currently lacking. The CCHFV glycoprotein precursor (GPC) is processed via multiple cleavage events to yield the virus surface structural glycoproteins Gn and Gc. The site-1 protease (S1P) is a host proprotein convertase essential for the production of infectious CCHFV. Although, Gn precursor (PreGn) of CCHFV is cleaved by S1P, the mechanism of action is currently not fully understood. Treating cells with low micromolar concentrations of an S1P inhibitor (PF-429242) blocked live CCHFV and virus-like particles (VLP) infectivity. In contrast, replication and transcription of RNA minigenome reporter activity was unaffected by S1P inhibition. As expected, PF-429242 inhibited Gn maturation. However, S1P inhibition also effectively inhibited Gc maturation suggesting that the Gc precursor (PreGc) is also cleaved by S1P, or that Gn maturation indirectly required for PreGc cleavage. Mutagenesis of PreGn and PreGc cleavage motifs confirmed that the PreGc cleavage motif (RKPL) is recognized by S1P. Infectivity of VLPs was abolished by a mutation blocking PreGc cleavage, whereas mutations blocking PreGn resulted in a ~5 fold reduction. Therefore, S1P direct cleavage of PreGc is the most critical cleavage explaining the loss in CCHFV infectivity. In conclusion, this study provide mechanistic details on how blocking of S1P cleavage leads to a dramatic block in CCHFV infectivity and the relevance of further development of S1P inhibitors as putative treatments for CCHF.

164. In Search of a Cure: A Scientist-Entrepreneur's Journey in Biotech

Michael Sofia, PhD
Arbutus Biopharma, Doylestown, Pennsylvania, United States of America

165. Historical Context and Biological Enigma of Rhinovirus C

Ann Palmenberg, PhD
Institute for Molecular Virology, University of Wisconsin-Madison

167. Antivirals at the Interface with Public Health: a Case Study of Polio

Mark Pallansch, PhD
Centers for Disease Control and Prevention, Atlanta, Georgia, United States of America

169. Top Ten Steps to Start Up a Small Bio-Business and Sources of Funding

Isaac R. Rodriguez-Chavez, PhD, MHS, MS
Science Consultant, Rockville, Maryland, United States of America

In this era of global markets in the bio-health space dominated by large multi-million dollar corporate partnerships, it is paramount to understand and define the most critical steps needed to initiate a small business and how to ensure its success. Considering that entrepreneurship is the backbone of wealthy and robust economies driven by prosperity, it is essential for governments and all sectors of the economies of countries to enable the creation and sustainability of small companies. This presentation will discuss the main strategic elements that entrepreneurs in science and medicine should consider when initiating a business. This presentation will also address statistical data about the success and failure rates for small businesses and the factors that influence making it or not during the first years after setting up small companies. An important segment will be focused on obtaining funds to finance scientific ideas and turn them into bio-health, marketable products. This presentation will close with an overview of the different types of corporate partnerships and a discussion on strategic alliances, outsourcing and licensing as key management tools that enhance logistical feasibility of businesses in the marketplace.

170. Small Molecule Inhibitors of Viral Entry: Pharmacological Mimicry of the Humoral Immune Response to Viral Infection

Priscilla Yang, PhD
Harvard Medical School, Boston, Massachusetts, United States of America

171. Catch me if you can – Using HBV Immunity for Therapy

Ulrike Protzer, MD
Institute of Virology, Technical University of Munich / Helmholtz Zentrum München, Munich, Bavaria, Germany

172. Broad-Spectrum Antivirals to Prevent Emerging Coronavirus Pandemic Disease

Timothy Sheahan, PhD
University of North Carolina at Chapel Hill, Chapel Hill, North Carolina, United States of America

Emerging viral infections are difficult to control as heterogeneous members periodically cycle in and out of humans and zoonotic hosts, complicating the development of specific antiviral therapies and vaccines. Coronavirus (CoV) have a proclivity to spread rapidly into new host species causing new disease. SARS-CoV and MERS-CoV successively emerged causing severe epidemic respiratory disease in immunologically naïve human populations throughout the globe. Broad-spectrum therapies capable of inhibiting CoV infections would address an immediate unmet medical need and could be invaluable in the treatment of emerging CoV infections. Here we show that a nucleotide prodrug GS-5734, currently in development to treat Ebola virus disease, can inhibit SARS-CoV and MERS-CoV replication in multiple in vitro systems including primary human airway epithelial cell cultures with submicromolar IC_{50} values (therapeutic index >100). GS-5734 was also effective against bat-CoVs, prepandemic bat-CoVs and circulating contemporary human CoV in primary human lung cells, thus demonstrating broad-spectrum anti-CoV activity. In a mouse model of SARS-CoV pathogenesis, prophylactic and early therapeutic administration of GS-5734 significantly reduced lung viral load and improved clinical signs of disease as well as respiratory functions in both adult and aged mice. For the first time, we demonstrate that a small molecule inhibitor has potent in vitro and in vivo antiviral efficacy against multiple zoonotic, epidemic and contemporary human CoV. These data provide substantive evidence that GS-5734 may prove effective against endemic MERS-CoV in the Middle East, circulating human CoV, and emerging CoV of the future.

173. From basic science to promising antivirals for hemorrhagic fever viruses

Christina Spiropoulou, PhD
Centers for Disease Control and Prevention, Atlanta, Georgia, United States of America

174. Flaviviral Translation[2]

Mariano Garcia-Blanco, MD, PhD
University of Texas Medical Branch

The *Flavivirus* genus contains several arthropod-borne viruses that pose global health threats, including dengue viruses (DENV), yellow fever virus (YFV) and Zika virus (ZIKV). We previously conducted genome-scale RNAi screens to identify candidate host factors and among these identified ribosomal proteins RPLP1 and RPLP2 (RPLP1/2) to be among the most crucial host factors required for DENV and YFV infection. RPLP1/2 are phosphoproteins that bind the ribosome through interaction with another ribosomal protein, RPLP0, to form a structure termed the ribosomal stalk. RPLP1/2 were validated as essential host factors for DENV, YFV, and ZIKV infection in human cell lines. RPLP1/2 knockdown strongly reduced early DENV protein accumulation, indicating a requirement for RPLP1/2 in viral translation.

Currently there are no specific therapies to treat flaviviruses. We interrogated a library of FDA-approved drugs for their ability to block infection of human HuH-7 cells by a newly isolated ZIKV strain (ZIKV MEX_I_7). More than 20 out of 774 tested compounds decreased ZIKV infection in our *in vitro* screening assay. Selected compounds were further validated for inhibition of ZIKV infection in human cervical, placental and neural stem cell lines, as well as primary human amnion cells. Established anti-flaviviral drugs (e.g., bortezomib and mycophenolic acid) and others that had no previously known anti-viral activity (e.g., daptomycin) were identified as inhibitors of ZIKV infection. This study identifies drugs that could be tested in clinical studies of ZIKV infection and provides a resource of small molecules to study ZIKV pathogenesis.

175. Nucleosides: A Rich Source of Antiviral Agents

Chung K (David) Chu, PhD
University of Georgia, Athens, Georgia, United States of America

176. Collaborating in Drug Discovery: Challenges and Solutions

Maaike Everts, PhD[1]
[1]*University of Alabama at Birmingham, Birmingham, Alabama, United States of America*

177. Trials and Tribulations of Starting a Biotech Business – Entrepreneurship 101

Robert Buckheit, Jr., PhD
ImQuest BioSciences, Frederick, Maryland, United States of America

178. Creating Value by Protecting Intellectual Property

Rebecca Kaufman, JD
King & Spalding, Atlanta, Georgia, United States of America

179. Impact of Transmitted HIV Phenotype on Host-Virus Interactions and Disease Progression

Eric Hunter, PhD
Emory University, Atlanta, Georgia, United States of America

180. Targeting HIV Reservoirs for Stimulation and Elimination

Jeff Murry, PhD
Gilead Sciences, Foster City, California, United States of America

181. Host Factors as Potential Targets for Influenza Virus Antivirals

Adolfo Garcia-Sastre, PhD
Icahn School of Medicine at Mount Sinai, New York, New York, United States of America

182. Zika Antiviral and Vaccine Development

Pei-Yong Shi, PhD
University of Texas Medical Branch, Galveston, Texas, United States of America

184. Treatment and Prevention of Emerging Rodent-Borne Viruses

David Safronetz, PhD
University of Manitoba, Winnipeg, Manitoba, Canada

185. In Search of a Cure: A Scientist-Entrepreneur's Journey in Biotech

Michael Sofia, PhD
Arbutus Biopharma, Doylestown, Pennsylvania, United States of America

186. Predicting ADME and PK Properties of Antivirals for Ebola

Mary A. Lingerfelt[1], Kimberley M Zorn[1], Joel S. Freundlich[2], Manu Anantpadma[3], Gauri Rao[4], John Diep[4], Robert A. Davey[3], Peter B. Madrid[5] and Sean Ekins[1]
[1]Collaborations Pharmaceuticals Inc., Fuquay-Varina, NC 27526; [2]Departments of Pharmacology, Physiology & Neuroscience and Medicine, Center for Emerging and Reemerging Pathogens, Rutgers University, Newark NJ, 07103; [3]Texas Biomedical Research Institute, San Antonio, TX 78227, USA; [4]Eshelman School of Pharmacy, University of North Carolina, Chapel Hill, NC 27599; [5]SRI International, Menlo Park, CA, 94025, USA

The West African Ebola outbreak of 2013-2016 resulted in over 11,000 deaths. To date there is no FDA approved antiviral or vaccine for the Ebola virus. We have previously detailed the creation of in silico Bayesian models using pseudotype viral entry assay and Ebola virus replication assay data for over 800 compounds. These resulting models were then validated both internally and externally and used to score a library of drugs available from MicroSource. Quinacrine, pyronaridine and tilorone, three of the highest scoring molecules that were not in either of the model training sets, were tested in vitro and had EC50 values of 350, 420 and 230 nM, respectively. All three compounds have been moved forward into in vivo efficacy testing in the infected mouse model.

In preparation for the murine in vivo testing, absorption, distribution, metabolism, excretion (ADME) and pharmacokinetic (PK) experimental data for pyronaridine and tilorone was obtained. The values pertaining to mouse liver microsomal stability, plasma protein binding, Caco-2 permeability and CYP inhibition (against CYP1A2, CYP2C9, CYP2C19, CYP2D6 and CYP3A4) were determined. We then used this in vitro data as a "real life" comparison to the computational predictions for microsomal stability, Caco-2 permeability and CYP inhibition using our previously described Bayesian models for these properties. Pyronaridine and tilorone had good ADME and PK properties (demonstrating adequate exposure) as would be expected for potential clinical candidates, and therefore further justified in vivo efficacy testing. Our combined in vitro and in silico results suggest a strategy that could be used for identifying additional antivirals.

We kindly acknowledge NIH funding: R21TR001718 from NCATS.

30TH International Conference on Antiviral Research (ICAR)

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Hosted by the International Society for Antiviral Research (ISAR)

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