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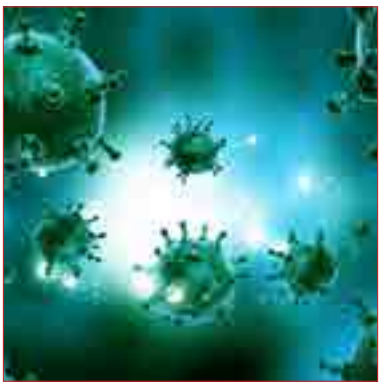
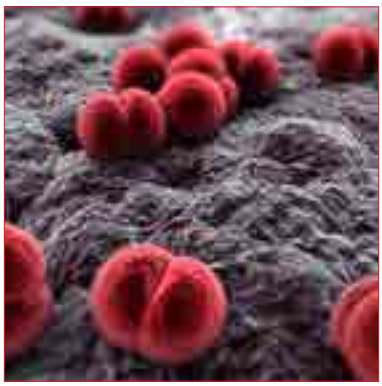
Workshop
August 17, 2017

Biotechnology 2017





ANNUAL BIOTECHNOLOGY CONGRESS

August 17-18, 2017 | Holiday Inn Toronto International Airport
Toronto, Canada

Azad K Kaushik, Ann Biol Sci, 2017, 5:3
DOI: 10.21767/2348-1927-C1-002

ANNUAL BIOTECHNOLOGY CONGRESS

August 17-18, 2017 | Toronto, Canada

Azad K Kaushik

University of Guelph, Canada

Developing novel vaccines for neonatal immunization

My laboratory discovered that some bovine antibodies are the largest known to exist in a species. This is because of generation of an exceptionally long CDR3H (up to 61 amino acids) which is encoded by an unusually long germline IGHD genes together with an unique insertion of "A" nucleotide rich conserved short nucleotide sequence at the IGHV-IGHD junction. The atypical CDR3H provides a "knob and stalk" structure capable of creating configurational diversity via variable intra-CDR3H disulfide bridges within the knob. The knob is held by the solvent exposed stalk formed by anti-parallel beta strands. I have exploited these structural features of the bovine antibody for the development of new therapeutics and vaccines. In this context, I will provide insights into the structural optimization of anti-viral bovine scFv to enhance their potency, apart from discussing influence of framework residues on viral neutralization function. I will provide 'proof of concept' for developing new vaccines by antigenizing bovine antibody with exceptionally long CDR3H that induce specific immune response. First, I successfully developed functional bovine scFv with an exceptionally long CDR3H followed by grafting of a viral B-epitope into the CDR3H. The grafted B-epitope in the exceptionally long CDR3H of bovine scFv sustained its native configuration and induced desired specific antibody response. Thus, antigenization of bovine scFv with an exceptionally long CDR3H provides a novel approach to developing new vaccines against infectious disease.

Speaker Biography

Azad K Kaushik has published two books *Molecular Immunobiology of Self-Reactivity* (1992) and *Comparative Immunoglobulin Genetics* (2014) and over 87 research articles. He is on the editorial boards of several immunology journals and is a Consultant to various international organizations. He was recognized as The Esther Z Greenberg Honors Chair in Biomedical Research, and Visiting Professor, Oklahoma Medical Research Foundation, USA, in 1998. He received BVSc&AH (Honors) in 1976 and MVSc (1978) from the Faculty of Veterinary Science, Hisar, Haryana, India; followed by Docteur es Science (DSc) in Immunology (1987) from the Pasteur Institute (University of Paris VII), Paris, France. He has been teaching Immunology at the University of Guelph since 1991.

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Notes:

Goetz RA Ehrhardt, Ann Biol Sci, 2017, 5:3
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Goetz RA Ehrhardt

University of Toronto, Canada

Biomarker discovery using monoclonal VLR antibodies of the evolutionarily distinct sea lamprey

Monoclonal antibodies are widely used reagents in biomedical research as well as in clinical applications. However, tolerogenic and structural constraints limit the antibody repertoire. In contrast to conventional antibodies, the recently identified variable lymphocyte receptor (VLR) antibodies of the evolutionarily distant jawless vertebrates utilize the **β**-sheet forming leucine-rich repeat (LRR) as basic structural unit. We hypothesize that the unique origins and radically distinct protein architecture will allow VLR antibodies to bind antigens, which conventional antibodies cannot readily recognize for tolerogenic or structural constraints. Memory B cells (Bmem) and plasma cells (PC) are tasked with providing long lasting humoral protection to re-encountered pathogens. However, no conventional antibodies exist that specifically detect these cell populations. In an effort to identify novel biomarkers uniquely expressed on Bmem and PC, we developed a method to generate monoclonal VLR antibodies to cell surface antigens. We isolated panels of monoclonal VLR antibodies binding specifically to human Bmem and PC. Flow cytometric analysis of VLR antibody binding to cell lines and primary human cells from blood, tonsil, spleen and bone marrow revealed binding patterns that are inconsistent with those of any conventional antibody, suggesting that the monoclonal VLR antibodies recognize novel antigens. Interestingly, we observed greatly increased VLR antibody binding to memory B cell populations in blood of individuals diagnosed with the autoimmune disorders Systemic Lupus Erythematosus (SLE) and Multiple Sclerosis (MS). Our data indicate that monoclonal VLR antibodies hold promise as novel reagents with a wide range of application potential in basic and clinical research.

Speaker Biography

Goetz RA Ehrhardt has completed his PhD at the University of British Columbia and continued his training as Post-doctoral fellow in the laboratory of Dr. Max D Cooper at Emory University in Atlanta, GA. In 2011, he was recruited to the Department of Immunology at the University of Toronto. His laboratory focuses on mechanisms governing the regulation of human memory B cell responses and on the use of the non-conventional VLR antibody system of jawless vertebrates for biomarker discovery purposes.

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Notes:

Ashok Kumar, Ann Biol Sci, 2017, 5:3
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Ashok Kumar

Children's Hospital of Eastern Ontario, Canada

Inhibition of the inhibitors of apoptosis (IAP) genes and selective apoptosis of HIV-infected human macrophages

To eradicate HIV in individuals on ART (antiretroviral therapy), it is imperative to eliminate both CD4+ T cells and myeloid cell reservoirs. Most research to-date has focused on eliminating the latent reservoir of CD4+ T cells. However, information regarding the potential elimination of HIV-infected macrophages is lacking. We have previously demonstrated that resistance of monocyte-derived macrophages (MDMs) to Vpr-induced effects was attributed to inhibitors of apoptosis (c-IAP-2) expression. These results suggest that strategies based on suppressing IAP-1/2 by its specific inhibitor, smac mimetics (SM) may be useful in killing HIV-infected macrophages. Our results show that SM do not affect uninfected normal MDMs but selectively kill *in vitro* HIV-infected macrophages, and macrophages derived from the HIV-infected patients. We also show that by using HIV-GFP strain, SM induced apoptosis of HIV- infected macrophages. In addition, HIV-chronically infected U1 cells were highly susceptible to SM-induced apoptosis in contrast to its corresponding U937 cells. We have investigated the molecular mechanism governing selective apoptosis of HIV-infected macrophages by SM. Our results show that SM induced macrophage cell death through apoptosis and not through necroptosis. Furthermore, SM-induced apoptosis in HIV-infected macrophages is not mediated through TNF-α. In addition, *in vitro* HIV infection inhibits expression of RIP-1, RIP-3 and TRAF-1 and bid. Inhibition of Rip-1 by its inhibitor, necrostatin, induced cell death in normal macrophages following treatment with SM suggesting a key role for Rip-1 in SM-induced killing of HIV-infected macrophages. Overall, the results suggest that inhibitions of IAPs could be a potential strategy to selectively kill HIV-infected macrophages.

Speaker Biography

Ashok Kumar is a Professor in the Department of Pathology and Laboratory Medicine and in the Department of Biochemistry, Microbiology and Immunology at the University of Ottawa, Ontario, Canada. He is also a Senior Scientist at the Children's Hospital of Eastern Ontario, Research Institute, Ottawa, ON, Canada. His main research interests are in the field of HIV immunopathogenesis, induction and regulation of apoptosis in HIV-infected human macrophages, regulation of T helper cytokines in health and disease and Toll-like receptor signaling in healthy and HIV-infected innate immune cells such as human macrophages and macrophage subsets.

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Scientific Tracks & Abstracts
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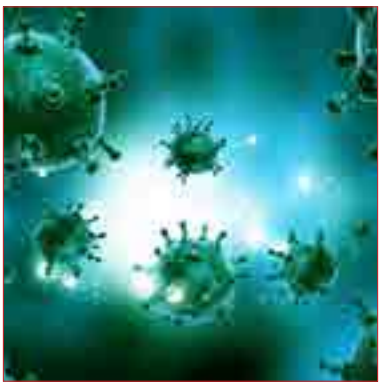

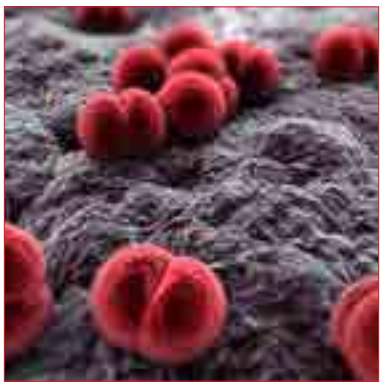

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Biochemistry | Molecular Biology | Biomedical Engineering | Pharmaceutical Biotechnology | Industrial Biotechnology | Nano Biotechnology | Environmental Biotechnology

Chair
Guido Krupp
AmpTec GmbH, Germany

Co-chair
Ricardo Hugo Lira
CIQA, Mexico

Session Introduction

Title: New insight into the functional switching of 2-Cys Peroxiredoxin revealed by high-speed atomic force microscopy
Hiroki Konno, Kanazawa University, Japan

Title: Hypoglycemic Mechanism of Guava Leaf Extract: Slow Inactivation of Protein Tyrosine Phosphatase 1B (PTP1B)?
Henry Tsai, Asia University, Taiwan

Title: RBBP6 isoform 3 plays a role in cell cycle regulation and carcinogenesis in cervical cancer
Dlamini Zodwa, Mangosuthu University of Technology, South Africa

Title: Application of Microarrays to Develop an In vitro – *In Vivo* Correlation Screening Toolbox
Craig Russell, Aston University, UK

Title: Metallic and carbon nanoparticles differentially impact physiological traits of four agricultural plant species
Ricardo Hugo Lira , CIQA, Mexico

Title: Quantitative characterization of RNA fitness landscapes
Ramon Xulvi-Brunet, National Polytechnic School, Ecuador

Title: Engineering biomaterials for medical imaging of cancer
Naomi Matsuura University of Toronto, Canada

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Hiroki Konno et al., Ann Biol Sci, 2017, 5:3
DOI: 10.21767/2348-1927-C1-002

New insight into the functional switching of 2-cys peroxiredoxin revealed by high-speed atomic force microscopy

Hiroki Konno, Takamitsu Haruyama, Takayuki Uchihashi, Yutaro Yamada, Noriyuki Kodera and **Toshio Ando**
Kanazawa University, Japan

Peroxiredoxin (Prx) is an ubiquitous antioxidant enzyme that reduces reactive oxygen species (ROS) such as hydrogen peroxide, organic peroxide and peroxynitrite. Prxs are classified into typical 2-Cys Prx, atypical 2-Cys Prx and 1-Cys Prx based on the number of cysteine residues and the catalytic mechanisms for their peroxidase activity. The function of 2-Cys peroxiredoxins (2Cys-Prxs) can be converted alternatively from peroxidases to molecular chaperones. This conversion has been reported to occur by the formation of high molecular weight (HMW) complexes upon overoxidation of or ATP/ADP binding to 2-Cys Prxs that appear in electron micrographs as spheres, decameric rings, double-stacked decamers or further stacked filaments. However, the entity responsible for the chaperone function is under debate. We employed the high-speed atomic force microscopy (HS-AFM) to investigate correlation between structure of HMW complex of human PrxII (hPrxII) and its chaperone activity. By the HS-AFM observation, we found that upon binding to phospholipids dimeric human 2-Cys PrxII (hPrxII) is assembled to small oligomers with full chaperone and null peroxidase activities. Spherical HMW complexes are formed, only when phospholipids is bound to overoxidized or ATP/ADP-bound hPrxII. The spherical HMW complexes are lipid vesicles covered with hPrxII oligomers arranged in a hexagonal lattice pattern. Thus, these lipids can be supplied by increased membrane trafficking under oxidative stress, are essential for the structural and functional switch of hPrxII and possibly most 2-Cys Prxs.

Speaker Biography

Hiroki Konno has completed his PhD from Tokyo Institute of Technology (Tokyo Tech) in 2002 and Post-doctoral studies from Tokyo Tech. In 2006, he joined chemical resources laboratory, Tokyo Tech, as an Assistant Professor. In the above period, he has studied regulation mechanism of rotary motor, ATP synthase, with biochemical and biophysical methods. Since November 2011, he has been with the Imaging Research Division of Bio-AFM Frontier Research Center, Kanazawa University, where he is currently an Associate Professor. His current research interests include observing protein molecule in dynamic action with HS-AFM.

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Henry J Tsai, Ann Biol Sci, 2017, 5:3
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ANNUAL BIOTECHNOLOGY CONGRESS

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Hypoglycemic mechanism of guava leaf extract: slow inactivation of protein tyrosine phosphatase 1B (PTP1B)

Henry J Tsai
Asia University, Taiwan

Guava leaf tea, has been used as a folk medicine for treating hyperglycemic conditions in Asia and Africa. The hypoglycemic efficacy of guava leaf has been documented by many scientists in these regions, but the hypoglycemic mechanism is poorly understood. Guava leaves were extracted with methanol and the crude extract was partitioned against hexane, ethyl acetate, and butanol in sequence. The leftover in water is defined as the aqueous partition. A second smaller batch was extracted with hot water directly. Our study confirmed the hypoglycemic efficacy on healthy mice and found the most effective molecules reside in the aqueous partition which is also less cytotoxic to Chinese hamster ovary cells when compared to other less polar partitions. Therefore, the guava leaf tea can serve as a functional hypoglycemic drink that is suitable for either healthy or diabetic subjects. Coincidentally the aqueous partition possesses a potent inhibitory effect on protein tyrosine phosphatase 1b (PTP1B) enzymatic activity and this PTP1B inhibition is through a slow oxidative inactivation on the enzyme.

Speaker Biography

Henry J Tsai obtained his PhD in Biochemistry (1996) and MS in Nutritional Science (1990) from the Michigan State University. He is currently an Associate Professor at the Department of Health and Nutrition, Asia University, Taiwan.

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Zodwa Dlamini et al., Ann Biol Sci, 2017, 5:3
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ANNUAL BIOTECHNOLOGY CONGRESS

August 17-18, 2017 | Toronto, Canada

RBBP6 isoform 3 plays a role in cell cycle regulation and carcinogenesis in cervical cancer

Zodwa Dlamini and **Zukile Mbita**
Mangosuthu University of Technology, South Africa

Cervical cancer is rated the second most common malignant tumor globally, and is etiologically highly linked to the human papillomavirus. South Africa is reported to have the highest incidence of cervical cancer in the world. It is the most common cancer in Black (31.2%) and Colored (22.9%) South African women. The DWNN domain is a novel ubiquitin-like cell death-related protein, that only makes up RBBP6 isoform 3 and RBBP6 isoform 1 and 2 also contain a zinc finger, RING finger, Rb-binding domain, p53- binding domain and Nuclear Localization Signal (NLS) downstream of the DWNN domain. This research was aimed at determining a consensus of gene expression pattern of the RBBP6 in cervical cancer both at mRNA and protein levels using ISH/FISH; ICC, RT-PCR and Western blotting. It was also of interest to determine the involvement of RBBP6 in cell cycle regulation and apoptosis. In this study, there was no detectable localization of the RBBP6 mRNAs in the tumor islands. The normal tissue showed few labeled cells. These results are in agreement with prior studies, which reported that cervix expresses low levels of RBBP6. In the cervical tumors, although tumor cells lacked RBBP6 mRNA, some cells in tissue located between the tumor islands were RBBP6 positive. It was found that cervical cancer cells (HeLa) do express the DWNN domain-containing RBBP6 gene products, at least at the mRNA level as demonstrated by FISH and RT-PCR. In this cell line RBBP6 exhibited both nuclear and cytoplasmic localization in mitotic cells (rearrangement of chromosomes as a marker for mitosis, with visible metaphase and anaphase) showing up-regulation of the RBBP6. Localization of DWNN-containing proteins in HeLa cells showed RBBP6 proteins *in situ* in HeLa cells and mitotic Hela cells at telophase showing increased lRBBP6 levels. RBBP6 isoform 3 was also shown to cause cell cycle arrest at G2/M and its diminished expression resulted in cell cycle progression. We have also shown that RBBP6 isoform 3 plays a role in cell cycle regulation and carcinogenesis in cervical cancer. These studies have shown that RBBP6 isoform 3 has great potential as a therapeutic target for cervical cancer biomarker and drug development.

Speaker Biography

Professor Zodwa Dlamini is the Deputy Vice Chancellor Research, Innovation & Engagements at Mangosuthu University of Technology and a Professor of Molecular and Functional Genomics. She was previously the Deputy Executive Dean at UNISA. She is also the current Vice-Chairperson of the South African Medical Research Council Board. She obtained her BSc and BSc.Hons. in Biochemistry from the University of the Western Cape, MSc from the University of Natal and PhD from the University of Natal. Her research interests include the "omics" technologies including the use of bioinformatics to provide unprecedented possibilities to identify the underlying molecular basis of cancer.

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ANNUAL BIOTECHNOLOGY CONGRESS

August 17-18, 2017 | Toronto, Canada

Application of microarrays to develop an *in vitro–in vivo* correlation screening toolbox

Craig Russell[1], Sumyra Begum[2], Yasar Hussain[2], Majad Hussain[2], David Huen[3], Ayesha S Rahman[3], Yvonne Perrie[4] and **Afzal R Mohammed[1]**
[1]Aston University, UK
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[3]University of Wolverhampton, UK
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Microarrays are powerful tools utilised in genomics allowing high throughput analysis of mRNA abundance. They have found application in many areas of drug discovery and development including comparative assessment of normal and diseased state tissues, transcription and expression profiling, side-effect profiling, pharmacogenomics and identification of biomarkers. In this application they were utilised to examine Caco-2 cells used in transport studies, investigating potential correlations between expression flux of genes coding for transpoter proteins known to interact with model drugs, and *in vitro* and *in vivo* permeability of the drugs, with a view towards developing a tool for predicting drug bioavailability early in the drug development pipeline. Lisinopril, Ramipril and Spironolactone formulations developed in house were used as model formulations. *In vitro* and *in vivo* uptake data was gathered for each formulation and focus was on genes coding transporters ABCB1, SLC15A1, SLC15A2, ABCC2 and SLCO1A2 following microarray analysis. Shortlisted genes of interest, all exhibited non-significant flux in expression levels in Caco-2 following analysis after transport studies using model formulations. There were however numerous SLC and ABC genes for which the expression had changed significantly. These were subsequently investigated using the Koyoto Encyclopaedia of Genes and Genomes (KEGG) to identify their function and seek clarity about the findings. Athough no clear cut revelations were derived from this study, the data strongly suggested that further research is warranted in this area, where future work intends to utilise a much larger formulation repertoire in conjunction with novel computational approaches currently in development to illucidate trends.

Speaker Biography

Craig Russell has completed his BSc (Hons) in Human Biology at the University of Huddersfield in 2010 and later obtained his PhD from Aston University. After which, he carried out his Post-doctoral research in the same institution. Upon completion of this work, he then became a Lecturer in Physiology and Pharmacology at Coventry University College before recently returing to Aston University in his current position as Lecturer in Pharmacy. He has multiple publications in reputable journals and serves as an Editorial Board Member for the *Journal of Research and Reviews: Drug delivery*.

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Notes:

Lira-Saldivar R H et al., Ann Biol Sci, 2017, 5:3
DOI: 10.21767/2348-1927-C1-002

Metallic and carbon nanoparticles differentially impact physiological traits of four agricultural plant species

Lira-Saldivar R H[1], Méndez-Argüello B[1], Vera-Reyes I[1, 3] and **De los Santos-Villareal G[2]**
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Recently, scientific community dedicated to the development of sustainable agricultural techniques have focused their attention and concern towards the application of engineered nanoparticles (ENPs), since the use of metal oxide nanoparticles could result in their accumulation in soil, threatening higher terrestrial plants. ENPs are able to interact with biomolecules, creating functional nanosystems for transportation within cells, and leading to the study of their potential applications in the field of Plant Biotechnology. On the other hand, the physical and chemical features of carbon nanomaterials such as multi-walled carbon nanotubes (MWCN) and graphite oxide (GO) NPs, had been used to promote plant's growth, and seeds germination. Therefore, this report focus on the application of copper nanoparticles (Cu NPs), iron oxide (Fe_2O_3 NPs), MWCN and GO NPs, to seeds and plants of *Solanum lycopersicum, Capsicum annuum, Cucumis melo* and *Rhapanus sativus,* to evaluate germination and plant growth characteristics. Imbibition of tomato seeds in Cu NPs significantly improved germination (14.3%), seedlings vigour (69%), plumule and radicle length increased by 20% and 95% respectively compared to control. In pepper seeds, Cu NPs also promoted vigour (118%) and seeds germination (10.2%); improved seedlings growth was reflected by longer plumule and radicle length (8% and 15% correspondingly). For *C. melo* Fe_2O_3 NPs also enhanced vigor (30%), germination (16.5%), plumule and radicle length by 20% and 95% respectively compared to control plants. On *R. Sativus* MWCN and GO NPs reduced plants growth and vegetative development, suggesting a phytotoxic effect by these carbon NPs.

Speaker Biography

Lira-Saldivar R H has completed his PhD from University of California, Davis. He is a Senior Researcher at the Centro de Investigación en Química Aplicada (CIQA) belonging to the Mexican Federal Government, located in Saltillo, Coahuila, Mexico. He has published more than 35 papers in reputed journals and has been serving as an Editorial Board Member of several journals.

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Ramon Xulvi-Brunet, Ann Biol Sci, 2017, 5:3
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ANNUAL BIOTECHNOLOGY CONGRESS

August 17-18, 2017 | Toronto, Canada

Quantitative characterization of RNA fitness landscapes

Ramon Xulvi-Brunet
National Polytechnic School, Ecuador

Experimental evolution of RNA (or DNA) is a powerful method to isolate sequences with useful function (e.g., catalytic RNA), discover fundamental features of the sequence-activity relationship (i.e., the fitness landscape), and map evolutionary pathways or functional optimization strategies. However, the limitations of current sequencing technology create a significant undersampling problem which impedes our ability to measure the true distribution of unique sequences. In addition, synthetic sequence pools contain a non-random distribution of nucleotides. We present and analyze simple models to approximate the true sequence distribution. We also provide tools that compensate for sequencing errors and other biases that occur during sample processing.

Speaker Biography

Ramon Xulvi-Brunet has completed his PhD in Theoretical Physics from Humboldt Universitaet zu Berlin, Post-doctoral Position in Applied Mathematics from University of Sydney, Post-doctoral Position in Biostatistics from University of Pennsylvania and Post-doctoral Position in Modelization of Biological Systems from Harvard University. He is a Research Scientist at University of California Santa Barbara and also serves as Physics Professor.

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Naomi Matsuura, Ann Biol Sci, 2017, 5:3
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ANNUAL BIOTECHNOLOGY CONGRESS

August 17-18, 2017 | Toronto, Canada

Engineering biomaterials for medical imaging of cancer

Naomi Matsuura
University of Toronto, Canada

Imaging is a fundamental tool in the practice of medicine. The interaction of medical imaging radiation with new materials has long been exploited to develop new and improved imaging systems and techniques. In parallel with these advances, there is increasing interest in developing new contrast agents for the diagnosis of disease. Exogenous contrast agents are non-native sources of contrast that differentially scatter, absorb, or emit medical imaging radiation (e.g., sound waves for ultrasound imaging, radiofrequency waves for magnetic resonance imaging, near IR light for photoacoustic imaging, and x-rays for computed tomography and mammography) as compared to surrounding tissues and inherent background noise such that their location can be tracked upon introduction into a patient. At the forefront of new contrast agent development are new, clinically-relevant, materials that can be activated by medical imaging radiation external to the patient and under image guidance, to characterize and treat cancer. Since the contrast agents' *in-vivo* distribution and interaction with radiation are strongly size- and material-dependent, a new opportunity in engineering is the creation of new nanoscale systems that can be tailored for specific contrast imaging and with therapeutic properties. This talk will focus on the development of new perfluorocarbon agents that can facilitate more focused and targeted delivery of cancer therapies to tumours for higher therapeutic ratios, and can permit the treatment of hard-to-access organs like the brain in a minimally-invasive manner

Speaker Biography

Dr. Naomi Matsuura, PhD, P.Eng., is currently an Associate Professor in Materials Science & Engineering and the Institute of Biomaterials & Biomedical Engineering (IBBME) with a cross-appointment in Medical Imaging at the University of Toronto. Dr. Matsuura leads a research program at the intersection of nanoengineering and medicine, focusing on the design of new contrast agents to guide the imaging and treatment of disease. Awards and recognitions include the John C. Polanyi Prize in Physiology/Medicine and Physics, an NSERC Discovery Accelerator Award, and the Early Researcher Award from the Ontario Ministry of Research and Innovation.

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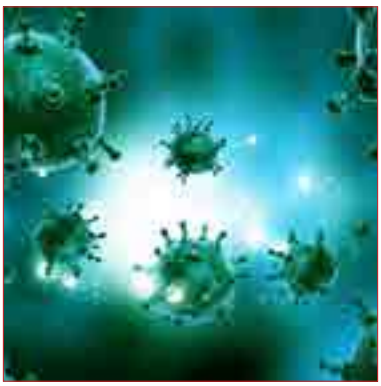
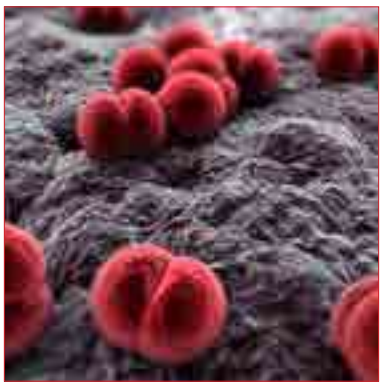
Young Researchers Forum
August 18, 2017

Biotechnology 2017





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Animal Biotechnology | Agricultural Biotechnology | Biomedical Engineering | Nano Biotechnology | Environmental Biotechnology | Bioinformatics

YRF Judge
Kiminobu Sugaya
University of Central Florida, USA

Session Introduction

Title: Efficiency of genetic screening for identification of lactic acid bacteria for their nutritional properties
Williams Turpin, University of Toronto-Mount Sinai Hospital, Canada

Title: The impact of epidermal growth factor (EGF) supernatant on pig performance and ileal microbiota
Nadeem Akthar, University of Guelph, Canada

Title: Improvement of Rice Variety PAU 201 Through Marker Assisted Selection For Grain Colour And Bacterial Blight Resistance
Kaur Rupinder, Punjab Agricultural University, India

Title: Omp31 plays an important role on outer membrane properties and intracellular survival of Brucella melitensis in murine macrophages and HeLa cells
Lázaro Verdiguel Fernández, Universidad Nacional Autónoma de México, Mexico

Title: Antagonistic effect of metallic nanoparticles on phytopathogenic fungi and bacteria
Ileana Verareyes, CONACYT-CIQA, Mexico

Title: Production of protegrin-1 with a matrix metalloproteinase/elastase cleavage site and its therapeutic potential for skin wound infections
Emily K. Hill, University of Guelph, Canada

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ANNUAL BIOTECHNOLOGY CONGRESS

August 17-18, 2017 | Toronto, Canada

Efficiency of genetic screening for identification of lactic acid bacteria for their nutritional properties

Williams Turpin
University of Toronto-Mount Sinai Hospital, Canada

The relationship between the lactic acid bacteria composing the microbiota of tropical starchy fermented foods and humans has been poorly investigated. Most of the studies focus on a combination of phenotypical (cells models, animals) and clinical trials. However, the increasing number of genomic data allows new strategies. Lactic acid bacteria (LAB) can synthesize molecules of interest during fermentation of food. The objective of this work was to screen the presence of around 50 genes involved in probiotic functions in a collection of 152 lactic acid bacteria isolated from an African fermented cereal based food called ben-saalga, and in the metagenome of various starchy fermented foods. In this study, several primers have been designed allowing the detection of genes of interest by PCR. The genetic screening is efficient in determining the potential linked to simple functions (B vitamins and carotenoids synthesis, starch metabolism, tannin degradation), as in most cases it allows to limit the number of phenotypical tests to the strain harbouring the genes of interest. On the contrary, more complex functions such as cell binding or bacterial survival, estimated *in vitro* (low pH, bile salts, cell models, surface plasmonic resonance imagery) revealed the limit of the approach. The genetic screening of the metagenomes shows that the traditional starchy fermented foods harbour a promising probiotic and nutritional potential.

Speaker Biography

Williams Turpin has completed his PhD in Microbiology from Montpellier II University (France), and Post-doctoral studies from University of Toronto (Canada). He is now a Research Associate at University of Toronto/Mount Sinai Hospital, currently working in the field of Human Genetics and its microbiome relationship in the context of inflammatory bowel diseases. He published six papers related to the field of Food Microbiology. His current work was acknowledged by five publications, with two of them published in high impact factor journals. He recently received one national (CDDW2015) and three international awards (UEGW2015, DDW2015-DDW2016).

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ANNUAL BIOTECHNOLOGY CONGRESS

August 17-18, 2017 | Toronto, Canada

The impact of epidermal growth factor (EGF) supernatant on pig performance and ileal microbiota

Nadeem Akhtar[1], Crystal Levesque[2], Evanna Huynh[1], Carrie Walk[3], Pete Wilcock[3], Zhengxiao Zhang[4], Paul Dyce[4], Cornelis F M De Lange[1], Ehsan Khafipour[4], and Julang Li[1]

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Early weaning of pigs can lead to low feed intake resulting in a lag in growth, performance and intestine infection. Epidermal growth factor (EGF), the most abundant growth factor in milk, increased weaned pig body weight gain and feed efficiency in our previous work. It is believed that intestinal microbiota plays an important role in pig growth but data is limited on the impact of feed additives on intestinal microbiota. The objective of the study is to investigate, if the positive influence on weight gain, and intestinal health with dietary EGF supplementation is related to differences in intestinal microbiota. *Pichia pastoris* were engineered to secrete porcine EGF using codon-optimized sequence. To examine the efficacy of EGF, an animal trial was performed using 72 pigs (2 equal blocks of 36 pigs with 3 barrows and 3 gilts/pen). The animals were assigned to one of two dietary treatments at weaning (20±2 d of age; n=6 pens/treatment) balancing across treatment for litter, gender and initial BW. Supernatant with EGF at 120 µg/kg BW/d and without EGF (control) was added to the feed for 21 d, followed by a common diet for 7 d. Animal performance was monitored on a weekly basis and ileal digesta samples were collected for microbiome analysis after 21 d of treatment. Pigs fed diets containing EGF fermentation supernatant had a greater (P=0.01) overall daily gain which is consistent to our previous finding. No difference in alpha-diversity (Chao1, Shanon, and Simpson indices) and beta-diversity (weighted and unweighted UniFrac distances) of ileal digesta microbiota between EGF supplemented and control pigs were observed. The relative abundances of bacterial taxa did not differ among treatment groups at the phylum level; however, the abundances of *Corynebacterium* (0.0 vs 0.9%), *Blautia* (0.003 vs 0.26%), and *Coprococcus* (0.0 vs 0.05%) genera and *Rumminococcaceae* family (0.001 vs 0.08%) were decreased (P<0.05) in EGF group compared to control, which might positively influence intestinal health.

Speaker Biography

Nadeem Akhtar has completed his PhD in Biotechnology from Thapar University, India and is currently is a Post-doctoral fellow in the Department of Animal Biosciences, University of Guelph, Canada. He has published more than 15 papers in reputed journals and filed a patent in India. He has also been serving as a Reviewer for few reputed journals such as *Environmental Progress & Sustainable Energy, Journal of Taiwan Institute of Chemical Engineers, Bioinfo Publications* etc.

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Notes:

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Kaur Rupinder et al., Ann Biol Sci, 2017, 5:3
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ANNUAL BIOTECHNOLOGY CONGRESS

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Improvement of rice variety PAU 201 through marker assisted selection for grain color and bacterial blight resistance

Kaur Rupinder, Mangat GS and **Singh Kuldeep**
Punjab Agricultural University, India

PAU 201, a very high yielding medium duration rice variety was released for cultivation in Punjab state in 2007, however, due to red pericarp color; the variety did not meet the specification of Food Corporation of India and was officially withdrawn in the year 2010 for cultivation in Punjab. Due to high yield potential, this variety covered more than 20% area in Punjab in two years after its release. In addition to red pericarp, this variety had only one bacterial blight resistance gene xa13. The red aleurone is due to single gene, designated as Rc7 and is located on chromosome 7. Both these genes are cloned and gene based primers have been designed. Due to consistent demand from farmers for improvement of this variety, we have improved this variety through MAS by replacing Rc7 allele with recessive allele *rc*7, and additional bacterial blight resistance gene *Xa*21. A set of BC_2F_4 progenies selected for white grain color and bacterial blight resistance having more than 90% of the recurrent-parent genome were evaluated for yield and yield components. Lines that significantly out-yielded the recurrent parent and the check cultivars in station trials are evaluated at multiple locations in national-level nurseries for identifying the lines that could be released as varieties. These lines, in addition to being released as cultivars, can also be used as immediate donors for further improvement of rice cultivars.

Speaker Biography

Ms. Rupinder Kaur is a Project Fellow with Department of Fruit Science, Punjab Agricultural University, Ludhiana 141 004, India. She is presently working with the objective of Standardization of high density planting and canopy architecture for high productivity and better fruit quality in low chill peach. Previously her research was concerned with the Varietal development for biotic stress resistance and yield improvement in rice through marker assisted selection. Ms. Rupinder attended the conferences and presented posters. She has published three scientific papers, five abstracts and one review paper in national and international journals of repute.

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Omp31 plays an important role on outer membrane properties and intracellular survival of *Brucella melitensis* in murine macrophages and HeLa cells

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Brucellosis is an infectious disease that affects practically all species of mammals, including humans, and is a major zoonosis worldwide. *Brucella sp*. are facultative intracellular pathogens that have the ability to survive and multiply in phagocytic and non-phagocytic cells such as trophoblast and epithelial cells. Among the six recognized species of the genus Brucella, *Brucella melitensis* is the main etiological agent involved in goat brucellosis and is also the most pathogenic for human. It causes significant losses in livestock production as a result of abortions, metritis, infertility, and birth of weak animals. Outer membrane proteins (OMPs) are exposed on the bacterial surface and are in contact with cells and effectors of the host immune response, whereby they could be important virulence factors of *Brucella* species. To evaluate this hypothesis, the gene encoding for the major outer membrane protein Omp31 was amplified, cloned into pUC18 plasmid, and inactivated by inserting a kanamycin cassette, rendering pLVM31 plasmid which was transformed into B. melitensis wild-type strain to obtain LVM31 mutant strain. The outer membrane (OM) properties of the mutant strain were compared with *B. melitensi*s Bm133 wild-type and *B. melitensis* Rev1 vaccine strains, in assessing its susceptibility to polymyxin *B, sodium deoxycholate*, and nonimmune serum. The mutant strain was assessed *in vitro* with survival assays in murine macrophages J774.A1 and HeLa cells. Our results demonstrate that LVM31 mutant is more susceptible to polymyxin B, sodium deoxycholate, and nonimmune serum than control strains. Moreover, Omp31 mutation caused a decrease in the internalization and a significant decrease in the intracellular survival compared with the reference strains in both cell lines. These results allow us to conclude that Omp31 is important for maintaining OM integrity, but also it is necessary for bacterial internalization, establishment and development of an optimal replication niche, and essential for survival and intracellular multiplication.

Speaker Biography

Lázaro Verdiguel-Fernández has completed his Master's degree from National Autonomous University of Mexico. Currently, he is a PhD student. He is Professor of Veterinary Immunology and Applied Molecular Microbiology and directed five undergraduate thesis. He has published one paper in *Archives of Microbiology Journal*. He has participated in the International Brucellosis Research Conference including the "69th Annual Brucellosis Research Meeting", New Delhi, India, 2016. He is a member of the Biotechnology Committee of the National Technical Advisory Council on Animal Health of Mexico.

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Antagonistic effect of metallic nanoparticles on phyto-pathogenic fungi and bacteria

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In this study, we investigated the antimicrobial properties of pure zinc oxide nanoparticles (ZnO NPs), Cu NPs and Fe_2O_3 NPs, against two plant pathogenic fungi: *Fusarium oxysporum*, *Alternaria solani* and one bacterial strain: *Clavibacter michiganensis,* which are the main microorganisms responsible for severe diseases of a large number of agricultural crops. Metal nanoparticles were applied at various concentrations (0, 250, 500 and 1000 mg L-1) to determine their *in vitro* antimicrobial activity. We synthesized ZnO NPs at room temperature by using a mechanically assisted metathesis reaction that permitted the formation of spherical NPs with mean sizes of around 20-30 nm. NPs characterization was accomplished by X-ray diffraction; particles size and shape were determined by TEM. We compared the effect of engineered NPs against commercial Cu and Fe_2O_3 NPs of similar size and shape. Antifungal activity of NPs was evaluated on PDA media, and King´s B medium was used for bacteria. ANOVA and Tukey multiple range tests were employed to analyse data. Cu NPs showed the greatest antimicrobial activity against both fungal strains, followed for ZnO NPs. In contrast, ZnO produced maximum growth inhibition against the bacteria *C. michiganensis*. On the other hand, Fe_2O_3 NPs did not exhibit antimicrobial activity with these phytopathogenic strains. Based on these results, it is viable that tested ZnO and Cu NPs could be used in programs of sustainable agriculture, since they are required in minute quantities by comparison to conventional pesticides.

Speaker Biography

Vera-Reyes I has completed her PhD from the Centro de Investigación en Estudios Avanzados del Instituto Politecnico Nacional, Mexico D F. She is a CONACYT Research Fellow.

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Production of protegrin-1 with a matrix metalloproteinase/elastase cleavage site and its therapeutic potential for skin wound infections

Emily K Hill and **Julang Li**
University of Guelph, Canada

Clinically-relevant pathogens are rapidly developing resistance to conventional antibiotics making infection control difficult. Methicillin-resistant *Staphylococcus aureus* (MRSA) is a gram-positive bacterium commonly found to be multidrug-resistant. As infections incited by MRSA are associated with increased rates of mortality compared to drug-sensitive strains, their presence in healthcare settings and emergence in the community is of growing concern. Protegrin-1 (PG-1) is a broad spectrum antimicrobial peptide effective against bacteria and antibiotic-resistant bacteria. PG-1 may be a potential alternative to conventional antibiotics and its promising therapeutic abilities have been previously demonstrated in infection models and in the reduction of inflammation in chemically-induced murine colitis. In the current study, proform PG-1 (ProPG) was designed to include a novel cleavage site and recombinantly generated for more efficient activation at sites of tissue inflammation. A widely selective matrix metalloproteinase (MMP) cleavage site was inserted into ProPG 5' to the native neutrophil elastase site to allow for the more efficient release of mature PG-1 at skin inflammation sites where MMPs are abundantly expressed. *Pichia pastoris* served as the expression host for the constructed expression vector and fermentation parameters were adjusted for optimal ProPG expression and secretion. Cleavage studies of the introduced MMP site performed with recombinant mouse MMP-3, demonstrated the functionality of the inserted MMP-3 site. Future studies will verify the selectivity of the MMP site and the therapeutic potential of ProPG against MRSA infected skin wounds.

Speaker Biography

Emily K Hill is in the process of completing her MSc (Animal Biosciences) from the University of Guelph in Dr. Julang Li's laboratory. She is scheduled to defend in August 2017 and has one publication so far. An aspiring veterinarian, she will begin her DVM degree at the Ontario Veterinary College in the Fall Semester.

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