Abstracts

As ordered in program

July 24th, 2019
Keynote Presentation

Label-free flux assay technologies in drug discovery and preclinical development

Since its first description (Terstappen, Anal. Biochem. 1999, 272, 149-155) the non-radioactive rubidium efflux assay - which employs Rb+ as tracer for K+ in conjunction with atomic absorption spectroscopy as detection technology - has found wide-spread application in drug discovery and the preclinical development of novel therapies. In addition to the original focus on ion channel drug discovery the basic concept of this technology has also been adapted for the study of transporters which is another important target class. On the occasion of its 20th anniversary I will present an overview of the development of this non-radioactive label-free technology and the various applications in drug discovery and preclinical development

Georg Terstappen
Chief Scientific Officer
OxStem

July 24th, 2019
Keynote Presentation

Synergistic activation of anti-tumor immunity by an oncolytic virus VG161 armed with multiple immune stimulating genes

Oncolytic viruses (OVs) are among the most powerful approaches in cancer immunotherapy. OVs not only cause cancer cell lysis but more importantly, their infection in tumors induces anti-tumor immune response from the host, resulting in lasting anti-tumor immunity. It has been recognized that anti-tumor immune response requires multiple immune regulatory factors that act synergistically and tumor microenvironment is critical for tumor to grow. Herpes simplex virus type-1 (HSV-1) has been approved by FDA as an oncolytic viral drug to treat melanoma. One advantage of HSV-1 is its large genomic capacity for carrying multiple exogenous genes.

A HSV-1 oncolytic viral vector (VG161) was constructed to simultaneously express IL12, IL15 with its receptor alpha unit and a PDL-1 blocking peptide. Anti-tumor activity of VG161 was tested in both immune competent mice (CT26 and A20 tumor models) and nude mice for human tumor models (LNCaP and U87). Since CT26 and A20 are poorly permissive for HSV-1 replication, the mouse tumor models were able to demonstrate the anti-tumor immune response induced by VG161 while oncolytic activity of VG161 was demonstrated in LNCaP and U87 models since the immune system is compromised in those models.

VG161 completely inhibited tumors in all the models tested and the animals survived tumor-free for many months till sacrificed. VG161 induced tumor oncolysis in both LNCaP and U87 tumors. In the CT26 model, animals were protected from the second challenging with CT26 cells following previous virally induced tumor regression. Furthermore, in a A20 double tumor model, intratumoral injection into the tumor on one side caused tumor regression on both sides. Transcriptome analysis showed significant change in tumor microenvironment. Finally, tumor specific memory T-cells were evident in the treated animals. The anti-tumor immune response by VG161 was significantly stronger than similar viruses that did not express any immune stimulating gene or only express GM-CSF. These results showed that Intratumorally expressed multiple immune regulatory factors by an oncolytic virus may significantly change the tumor immune microenvironment to enhance efficacy of the oncolytic virus.

William Jia
Chief Scientific Officer & Co-Founder
Virogen Biotech Ltd.

July 24th, 2019
Peptide Research in Drug Discovery - Precision Medicine

The role of NaV1.6 in sensory neurons: insight from subtype-selective venom peptides

Peripheral sensory neurons express multiple voltage-gated sodium channels (NaV) critical for the initiation and propagation of action potentials and transmission of sensory input. Emerging evidence suggests NaV1.6 channels are an important isoform in pain sensing. Using the potent and NaV1.6-selective β-scorpion toxin Cn2, we defined the contribution of NaV1.6 to sensory neuron function using Ca2+ imaging, whole-cell patch-clamp recordings, skin-nerve preparations and in vivo behavioural assessment. Enhanced NaV1.6 early channel opening, increased persistent and resurgent currents facilitated by Cn2 led to enhanced excitatory drive and tonic action potential firing predominantly in mechanosensitive A-fibers and emergence of nocifensive responses *in vivo*. In conclusion, sensory neurons expressing NaV1.6 are important for the transduction of mechanical information in sensory afferents.

Irina Vetter
Co-Director & Principal Research Fellow, Institute for Molecular Bioscience
University of Queensland

July 24th, 2019
Peptide Research in Drug Discovery - Precision Medicine

Peptide array platform for studying protein-protein interaction and potential development of novel drugs

The SPOT synthesis of peptides on cellulose membranes was first developed in early 1990s, and its application is only pioneered in few laboratories worldwide. With this technology, it becomes possible for peptide manipulation including addition (D-type amino acids, unnatural amino acids, or even other non-amino acid motifs), substitution/mutation, and amino acid combination. Peptide array technology can be used to study most of protein-protein interactions in areas such as vaccine development and drug discovery.

William Jia
Chief Scientific Officer & Co-Founder
Virogen Biotech Ltd.

July 24th, 2019
Peptide Research in Drug Discovery - Precision Medicine

Construction and Screening of a Cyclic Peptide Library

A cyclic peptide library was constructed based on the Peptide Information Compression Technology (PICT). The library contains all the possible pentapeptide sequences, and therefore all the possible tripeptide and tetrapeptide sequences. The library also contains millions of longer peptide (up to 80 amino acids) sequences. The initial test screening of 400 cyclic peptides randomly picked from the library was performed using CisBio PD1/PD-L1 Binding Assay Kits, which gave satisfactory results, and 4 of the samples showed IC50 values lower than 100 nM.

Zhuyin Wang
Professor
Lanzhou University

NaV1.6 selective and NaV1.2/NaV1.6 dual Inhibitors Reduce Action Potential Firing in Mouse Cortical Pyramidal Neurons While Sparing Inhibitory Interneuron Firing

An ideal antiepileptic drug (AED) would inhibit hyper-excitability associated with seizures in excitatory circuits while sparing inhibitory circuits and other unwanted targets. Non-selective inhibitors of voltage-gated sodium channel (NaV) inhibitors, like carbamazepine, are effective in some epilepsy patients but many patients remain refractory to treatment. These drugs inhibit the sodium channels that drive action potential (AP) firing in excitatory neurons (NaV1.2 and NaV1.6 channels) as well as those that drive inhibitory interneuron firing (NaV1.1). Gain of function mutations in both SCN8A (encoding NaV1.6 channels) and SCN2A (encoding NaV1.2 channels) cause early infantile epileptic encephalopathy in humans (EIEE13 and EIEE11 respectively). Conversely, loss of function mutations in SCN1A (encoding NaV1.1) cause Dravet Syndrome (EIEE6) and generalized epilepsy with febrile seizures plus (GEFS+). Non-selective sodium channel inhibitors are known to exacerbate seizures in Dravet patients, further highlighting the liability non-selective Nav inhibitors may have because of modulation of targets in inhibitory interneurons as well as at other sites (eg cardiac and muscle). Selective sodium channel inhibitors that reduce AP firing in excitatory neurons, while sparing inhibitory interneurons and non-CNS Nav targets, may therefore provide a better pharmacologic profile and therapeutic index compared with older non-selective Nav modulators such as carbamazepine, lamotrigine and lacosamide.

We created novel small molecule inhibitors that target NaV1.6 selectively (XPC'7224) or NaV1.2 and NaV1.6 in combination (XPC'5462) while sparing NaV1.1 and other voltage gated sodium channels. We then tested the relative efficacy of these compounds to inhibit action potential firing in current clamped mouse brain slice neurons. Activity on excitatory pyramidal neurons and inhibitory interneurons of these selective inhibitors was compared to that of carbamazepine.

Selective inhibition of NaV1.6 with 500 nM XPC-7224 (3X IC50 for NaV1.6) inhibits firing of pyramidal neurons in cortical layer 2/3 pyramidal neurons in brain slices from adult mice. However, the same concentration of XPC-7224 had markedly less impact on AP firing in inhibitory interneurons. Similarly, inhibition of NaV1.2/1.6 with 150 nM XPC-5462 (3X NaV1.6 IC50) impairs AP firing in cortical pyramidal neurons with only modest effect on AP firing in inhibitory interneurons. In contrast, 100 µM carbamazepine (3X NaV1.6 IC50), impairs AP firing in both pyramidal neurons and interneurons .

JP Johnson
Vice President, Biology
Xenon Pharmaceuticals

July 24th, 2019
Ion Channels in Epilepsy - Ion Channel

Development and validation of selective and potent small-molecule probes for the CLC-2 chloride channel

CLC-2 is the most broadly expressed voltage-gated chloride channel in the mammalian brain. Although its presence has been known for decades, the neurophysiological function of CLC-2 remains controversial, in part due to the absence of potent and selective small-molecule tools to acutely and reversibly inhibit its function. To develop small-molecule inhibitors of CLC-2, we first screened a small library of FDA-approved compounds and identified several low-micromolar hit compounds. Through four rounds of Structure-Activity-Relationship studies on one of these hit compounds, we Identified a first-in-class CLC-2 inhibitor with low-nanomolar potency and no significant off-target effects on other CLC channels or on a panel of CNS channels, transporters, and receptors. We demonstrated the efficacy of this inhibitor in blocking CLC-2 currents in CA1 hippocampal pyramidal neurons in mouse brain slices, using slices from knock-out animals as a control. This inhibitor thus presents itself as a novel tool for studying the function of CLC-2 in the brain.

Merritt Maduke
Associate Professor, Molecular & Cellular Physiology
Stanford University

P/Q-type Calcium Channel Dysfunction: an Underlying Mechanism in Sudden Unexpected Death in Epilepsy

Sudden Unexpected Death in Epilepsy (SUDEP) is strongly linked to seizure activity yet ~30% of people with epilepsy do not adequately respond to current treatments and little is known concerning the underlying mechanism of SUDEP. We have discovered that a mouse model of Familial Hemiplegic Migraine Type-1 (P/Q-type calcium channel defect) is also associated with sudden death. Combining DW-MRI to examine spreading depolarization (SD) with high spatiotemporal visualization and EEG recording to correlate seizure activity, we identify subcortical regions susceptible to seizures and concomitant SD that together result in SUDEP. Optogenetic and biophysical experiments confirm that selected subcortical regions are sensitive to SD propagation into the brainstem likely due to underlying calcium-channel mediated enhanced neuronal excitability.

Terry Snutch
Professor, Canada Research Chair, Biotechnology & Genomics-Neurobiology
University of British Columbia

July 24th, 2019
Neurodegenerative Diseases - Ion Channel

Patient-derived organoid as a model to study Alzheimer's Disease and drug development.

Alzheimer's Disease (AD) is an incurable disorder that causes a heavy burden not only to the afflicted patient, but also to associated family, caregivers, and loved ones. It is estimated that overall costs can reach over $150 billion per year. Currently, there is not an effective treatment for this disease, and many pharma companies have discontinued active pre-clinical research in this field. To further complicate therapeutic development, in vivo model that has been developed to study the disease and used to test therapeutic, do not comprehensively recapitulate the human disease. It is, therefore, paramount to develop a model system where human brain cells are used to study brain disorders. Induced Pluripotent Stem cells can be obtained from patients blood sample, and they have the ability to differentiate into all cell type including neurons. In addition, when cells are differentiated into a three-dimensional structure, they can recapitulate human brain development. Here we show that AD derived organoids, accumulate phosphor Tau, and beta-amyloids providing evidence that this is a good in vitro system to use for drug development.

Michelina Iacovino
Assistant Professor, Pediatrics, Division of Medical Genetics
David Geffen School of Medicine UC Los Angeles

July 24th, 2019
Neurodegenerative Diseases - Ion Channel

The Role of Ion Channels in Dementia and Other Neurodegenerative Disorders

The prevalence of neurodegenerative disease is rising rapidly as the median age of the world population increases. Alzheimer's disease (AD), Lewy body dementia (LBD), and Frontotemporal dementia (FTD) spectrum disorders represent most of the disease burden and none currently have disease-modifying treatments. This talk will provide an overview of the presentation, pathophysiology and the known role of ion channels in these disorders.

In AD, memory and cognition progressively decline as amyloid and tau proteins accumulate in the brain. The role of these proteins in neurotoxicity is unknown and the ion channel hypothesis has been proposed. Targeting the proteins directly has been ineffective. The only approved therapies instead aim to correct a cholinergic deficit and decrease glutamate-triggered neurotoxicity.

In LBD, movement, cognition, behavior, autonomic function and sleep are all progressively impaired, attributed to alpha synuclein's prion-like spread through the central and peripheral nervous system. Potassium channels have been implicated in the pathogenesis of Parkinson's disease as have other ion channels in the activation of microglia, causing loss of dopaminergic neurons in the substantia nigra.

FTD spectrum disease may have variable presentations, with behavior, language, and movement dysfunction. Tau is the most common pathogenic protein, likely spreading in a prion-like fashion and suspected to cause neuronal degeneration by modulating NMDA receptor response to glutamate resulting in potentially toxic intracellular influx of calcium.

AD, LBD, and FTD spectrum disorders are devastating conditions with a dismal prognosis. Ongoing discoveries suggest underlying similarities in their molecular mechanisms that may pave the way for novel therapeutics.

Oleg Yerstein
Postdoctoral Fellow, Behavioral Neurology and Neuropsychiatry
David Geffen School of Medicine UC Los Angeles

July 24th, 2019
Neurodegenerative Diseases - Ion Channel

Precision Medicine in the Treatment of Neurodegenerative Diseases

The prevalence and impact of neurodegenerative conditions are increasing as population demographics shift towards a greater proportion of elderly members of society. Precision medicine based treatment strategies will be a key component of successfully answering the healthcare challenges posed by neurodegenerative disorders. The goal is to identify innovative and sustainable treatment approaches that optimize development of safer and more effective therapies, as well as identify at risk individuals at pre-clinical or early stages of neurodegenerative disorder when treatments may be most effective. Individualized preventive strategies may be an effective and feasible way to reduce disease burden for this group of disorders, a group of disorders that is considered very difficult to treat because disease modifying therapies are not currently available.

Aaron McMurtray
Associate Clinical Professor of Neurology
UC Los Angeles Health

The Massively Parallel Sequencing (MPS) Revolution

The development of automated DNA sequencers utilizing Sanger sequencing and capillary electrophoresis made it possible to develop the first draft sequences of the human genome. The cost of doing this was hundreds of millions of dollars, which makes it totally impractical in the clinical setting. However, the advent of technologies which could generate sequences for extremely large numbers of DNA fragments simultaneously based up MPS heralded a revolution in sequence capability. All first generation MPS platforms utilize one of three approaches to amplify individual DNA molecules to a high copy number followed by sequence interrogation of the original short DNA molecules. The most successful platform for MPS was developed by the company Illumina and they have increased sequence output from 1 gigabase (Gb) to over 6 terabases (Tbs) in less than 13 years. Currently the cost for generating sufficient DNA sequence for whole genome sequencing (WGS) of an individual human is now just $375. However, the total cost for WGS is considerably higher when one factors in library preparation, sequencing, assembling and interpreting that genome sequence, and data storage. There is an alternative platform developed by Complete Genomics based upon a non-PCR based technology to amplify DNA templates. This platform is now being utilized by BGI and they have a machine which is capable of generating 7 Tbs of sequence data per run. On this platform the total cost for WGS is now just $600 and BGI is developing larger machines in the hope of bringing WGS total costs down to just $100. First generation MPS can now be utilized for WGS, but also for whole exome sequencing, targeted genome sequencing, transcriptome sequencing, methylation sequencing as well as metagenomic sequencing. All of these will completely transform research and its' clinical translation. Second generation MPS is based upon the analysis of single unamplified DNA molecules and can generate DNA sequences that can be 100 kilobases in length or greater. In my talk I will discuss the history of first generation MPS and how this revolution represents an important technological singularity. I will also discuss the strengths and weaknesses of short read sequence technologies.

David Smith
Professor, Department of Laboratory Medicine and Pathology
Mayo Clinic

Oxford Nanopore long-read sequencing to refine somatic variant identification

Third generation long read sequencing technologies, including Oxford Nanopore (ONT) sequencing, directly sequence DNA and RNA molecules, enabling the sequencing of extremely long reads that is not possible with short read sequencing by synthesis technologies. This direct sequencing enables accurate characterization of haplotypes, DNA methylation, structural variants, and fusion transcripts. However, ONT sequencing has a very high error rate (~15%), so it is necessary to either sequence to a high depth, which is currently cost prohibitive for the human genome, or to correct the ONT sequencing errors with highly accurate short read sequencing. In the case of the low-affinity Fcγ receptor gene FCGR2B, which is implicated in treatment resistance in non-Hodgkin lymphoma, short read sequencing technologies fail to distinguish it from its highly similar paralog FCGR2C. Through ONT sequencing of four distinct haplotypes from two human bacterial artificial chromosome libraries and cell line-derived transcripts, we were able to accurately place sequence variants that distinguish the two paralogs, improving the detection of somatic variants affecting FCGR2B. Several methods for targeted long read sequencing have been developed, which will facilitate accurate somatic variant identification in regions that are difficult to parse with short read sequencing and increase the utility of this technology in clinical applications.

Laura Hilton
Postdoctoral Fellow
Simon Fraser University

July 24th, 2019
Application & Advancement of Next Generation Sequencing - Precision Medicine

High-Resolution Views of the Human Genome and Transcriptome with PacBio SMRT Sequencing

The lofty ambitions of precision medicine will require that both the research and medical communities have access to highly accurate genomic and transcriptomic information for individuals. While the short-read sequencing era has ushered in an impressive understanding of single nucleotide variants and how these contribute to Mendelian or de novo disorders, many of the underlying differences between humans come in more complex forms, such as structural variants, allele-specific gene regulation and expansions of repeat regions. Long-read SMRT sequencing can now illuminate much of this human biology previously inaccessible with short reads. In this talk, I will discuss how accurate long read sequencing has uncovered new features of the genome and the underlying cause of some genetic diseases, with several examples highlighting the impact on genes encoding ion channels. I will also elaborate on how long reads enable isoform-level transcriptomes, with particular emphasis to genes such as neuronal ion channels and receptors that exhibit extensive alternative splicing.

Jason Underwood
Principal Scientist
Pacific Biosciences

July 24th, 2019
Cancer Diagnosis & Treatment - Precision Medicine

BRAvE at BC Children's Hospital: a precision medicine research initiative for bone marrow-involved childhood cancers

Almost 1000 children under the age of 14 are diagnosed with cancer each year in Canada. Although current treatments will achieve cure in about 80% of these patients, cancer remains the leading disease-related cause of death for children and adolescents. For childhood cancers that are initially refractory or that relapse following first-line treatments, the survival rates are still dismal. Personalized medicine clinical trials are currently ongoing and early results show that the majority of profiled tumors have mutations that may be specifically targeted with a new therapy. Unfortunately, only about 8% of patients are currently receiving these new molecule-targeted treatments. This is a critical current roadblock for the adoption of personalized medicine.

The BRAvE (Better Responses with AVatomics Evidence) Initiative at BC Children's Hospital (BCCH) aims to tackle this roadblock. By extending the application of precision medicine approaches and initiating studies when the disease is first diagnosed, the team plans to use the remission phase to prepare for a possible relapse. Established in 2016, BRAvE at BCCH team has addressed significant hurdles to this approach by: evaluating the first NGS assay specifically designed for childhood cancers; assessing the persistence of predicted tumor variant-agent pairings from diagnosis through relapsed disease; establishing and incorporating a proteomic workflow for target ID; and, validating predicted tumor variant-agent pairings. This work represents a collaboration between the Lange, Lim, Maxwell, and Reid labs at BCCH with philanthropic support from the BCCH Foundation, Michael Cuccione Foundation and Team4Hope Foundation.

Chris Maxwell
Co-Lead, Childhood Cancer & Blood Research Group
BC Children's Hospital

July 24th, 2019
Cancer Diagnosis & Treatment - Precision Medicine

Xenotransplantation of minimal residual disease reveals early enrichment of immunophenotypic subpopulations during leukemia therapy

The presence of minimal residual disease (MRD) at the end of induction therapy is the strongest predictor of leukemia relapse, for which outcomes remain poor. Despite this prognostic value, MRD+ samples have been under-utilized for the investigation of early therapy resistance and disease progression because of the low numbers of leukemic blasts present. Here we show that xenotransplanting MRD+ bone marrow from children with acute lymphoblastic leukemia expands the treatment-selected blasts with patient-specific kinetics. The absence of normal human hematopoiesis in MRD xenografts provides clarity on the broader leukemia immunophenotype, revealing previously unappreciated subpopulation heterogeneity and phenotypic shifts that diverged from diagnostic xenografts. By incorporating unbiased dimensionality-reduction algorithms into the analytical platform, we demonstrate that distinct leukemic subpopulations, already present at MRD-levels at diagnosis, are enriched in patients by induction therapy. This novel approach could refine clinical treatment-response assessment and provide a platform for identifying vulnerabilities in early chemotherapy-resistant blasts.

Gregor Reid
Investigator & Scientist, Michael Cuccione Childhood Cancer Research Program
BC Children's Hospital

July 24th, 2019
Cancer Diagnosis & Treatment - Precision Medicine

Microfluidic Single-Cell Technology for Liquid Biopsy

Expression of ABC transporters, such as P-gp, on cancer cells is a hallmark of multidrug resistance (MDR), which is one of the major obstacles in cancer drug delivery. MDR may be overcome by using inhibitors that block the cell-surface transporters. We employed the microfluidic single cell biochip (SCB) to measure cancer cells (leukemia, prostate, ovarian, lung, and breast) for their drug uptake, which would be enhanced by using MDR inhibitors. We further made use of the microfluidic technology to select rare cancer cells from among blood cell samples, referred by many researchers as the liquid biopsy. Enhancement of drug uptake in a circulating tumor cell by MDR inhibitors may indicate a good prognosis for patient treatment in precision medicine.

Paul Li
Professor, Department of Chemistry
Simon Fraser University

July 24th, 2019
Ion Channels & Immune System - Ion Channel

Identification of novel ion channels regulating T cell-mediated immunity

Ion channels and transporters (ICTs) control ion fluxes across lipid membranes and play pivotal roles in a multitude of cell functions. While ICTs have been extensively investigated in excitable cells, there is a surprising lack of knowledge with respect to their function in immune cells and immunity. Of the more than 500 known ICTs only 10-15 are well established to play a role in immune responses. This includes Ca2+ channels such as CRAC (encoded by ORAI and STIM genes), TRPM2 and TRPM7, the Na+ channel TRPM4, the Mg2+ transporter MAGT1, the K+ channels KV1.3 and KCa3.1, the Cl- channel LRRC8A and the Zn2+ transporter ZIP7. Overall, our knowledge of ICTs in immune function is very limited. In order to fill this gap, our lab has developed in vivo shRNA screening approaches to identify novel ion channels and regulators that are required for T cell mediated immune responses to viral infection, tumors and in autoimmune diseases. This talk will provide an overview of ICTs regulating immune function, immune ion channelopathies and discuss new insights into the role of ion channels in T cell mediated immune function.

Stefan Feske
Jeffrey Bergstein Professor of Medicine & Director, Ion Channel & Immunity Program
New York University, Langone Medical Center

July 24th, 2019
Ion Channels & Immune System - Ion Channel

Targeting KCa1.1 in fibroblast-like synoviocytes for the treatment of rheumatoid arthritis

Rheumatoid arthritis (RA) is a chronic systemic autoimmune disease that leads to joint destruction, pain, and decreased mobility. Fibroblast-like synoviocytes (FLS) are resident joint cells. During RA, FLS develop a pathogenic phenotype involving invasiveness and the production of pro-inflammatory cytokines, chemokines, angiogenic factors, and proteases. FLS contributes to cartilage and bone degradation within the joint and to synovial hyperplasia. No RA therapy specifically targets FLS.

We have found that FLS from patients with RA and from rats with models of RA have increased expression of KCa1.1 at their plasma membrane when compared to FLS from patients with osteoarthritis or from healthy rats, respectively. The presence and activity of KCa1.1 at the plasma membrane of RA-FLS is both necessary and sufficient to regulate the in vitro invasiveness of RA-FLS. Blocking KCa1.1 with the small molecule paxilline or the scorpion venom peptide iberiotoxin reduces many of the pathogenic aspects of RA-FLS, including proliferation and release of cytokines, chemokines, angiogenic factors, and proteases.

Blocking KCa1.1 after onset of clinical signs significantly reduces joint inflammation, bone and cartilage damage, and synovial hyperplasia in two rat models of RA. FLS from blocker-treated animals also have reduced ex vivo invasiveness and proliferation. While paxilline induces severe side effects, including tremors, iberiotoxin appears not only efficacious but also safe for systemic administration.

These studies indicate the importance of KCa1.1 as a novel target for RA and emphasize the potential efficacy of directly inhibiting FLS with iberiotoxin to reduce the severity of this debilitating disease.

Christine Beeton
Graduate Program Director & Associate Professor, Biomedical Sciences & Molecular Physiology
Baylor College of Medicine

TRPM7 at the Crossroads of Tissue Homeostasis and Inflammation

Efficient and non-inflammatory clearance of apoptotic cells, also known as efferocytosis, is a fundamental biological process of major significance to developmental biology, organ physiology, and immunology. Understanding the cell biology of efferocytosis holds the biomedical potential to advance novel therapies in a wide range of diseases, including inflammatory diseases, cancer and neurodegenerative diseases. Much is known about how macrophages find and engulf apoptotic cells, but very little is known about how the engulfed apoptotic cells are digested (phagosome maturation). In this study, we reveal that the ion channel TRPM7 is a crucial mediator of Ca^{2+}-signaling that is essential for phagosome maturation during efferocytosis. Using genetically-encoded Ca^{2+}-sensors, we show that rapid, periphagosomal (surrounding the phagosome) Ca^{2+}-signals are required for phagosome maturation and that deletion of TRPM7 in myeloid cells diminishes *ex vivo* and *in vivo* clearance of apoptotic cells. Overall, our study has identified a crucial pharmacologically tractable ion channel drug target that regulates phagosome maturation through electrical activity at the phagosomal membrane. These studies now set the stage for understanding the implications of defective phagosome maturation in a wide array of physiological processes that lie at the crossroads of inflammation and tissue homeostasis.

Bimal Desai
Associate Professor, Pharmacology
University of Virginia

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Role of ERG1 channel in lymphocyte development and in leukemic clonal evolution

It is well established that different types of ion channels, play a relevant role both in the normal process of T- and B- cell development and function, as well as during lymphoblast's neoplastic transformation. In our laboratory we have provided evidence that ERG1b, an isoform of the ether-a-gò-gò-related gene 1, encoding for a K+ channel, is overexpressed in leukemias, and in myeloma, and its expression correlates with a worse prognosis both in AML and ALL. The current hypothesis is that these facts can be traced back to a relevant role exerted by ERG1B at specific stages of lymphopoiesis. In this scenario our aim is to study how ERG1B affects B and T lymphocytes development, and to determine the mechanisms underlying hERG1b over-expression and dysregulation in order to define the role of the channel in the leukemogenic process.

Mice carrying a selective deletion of the Erg1b gene show a block in the lymphocyte development both in the B and T lineages respectively at the DN and proB stages. BM cells of Erg1b KO mice display a reduced capacity to develop in vitro colonies, in particular, CFU-GEMM, CFU-GM and CFU-M colonies were reduced. On the contrary, erythroid precursors were increased both at the CFU-E and BFU-E level. The signaling underpinning the lympho- erythro- poiesis differentiative mechanisms was found affected. Particularly we noticed a reduction in the level of AKT phosphorylation both in the erythroid compartment and in the bone marrow.

Our results indicate a physiological role of ERG1B during the regulation of self-renewal, proliferation, differentiation and migration of HSCs and HSC-derived multipotent progenitor cells. The fact that the Erg1b turned out to be highly and selectively expressed in the staminal Lin-/Sca+ HSC compartment suggests a role of ERG1B in the differentiative programs. The evidenced block in the development might represent a first step in the leukemic transformation.

Cesare Sala
Research Associate
University of Florence

July 25th, 2019
Keynote Presenation - Precision Medicine

Liquid biopsy approaches from genomics and beyond

A multitude of approaches are currently available for liquid biopsy applications from cancer screening and early detection to tumor molecular profiling and monitoring. Nucleic acid approaches include tumor mutational burden, DNA methylation and analysis of coding and non-coding RNA. Other approaches include proteomics, metabolomics and immunomics and analysis of circulating extra-cellular vesicles. While the application of tumor mutational burden to cancer assessment has advanced significantly, other applications are in the early stage. The current state of liquid biopsy applications to cancer detection and the progress made will be reviewed.

Samir Hanash
Director, Red and Charline McCombs Institute for the Early Detection and Treatment of Cancer
The University of Texas MD Anderson Cancer Center

July 25th, 2019
Keynote Presenation - Ion Channel

Novel Treatments for Pain, Cough and Itch by Selective Targeting of Sodium Channel Modulators into Nociceptors

Nav channel blockers like lidocaine are effective anesthetic agents that have shown clinical utility in the treatment of pain, itch and cough. Local anesthetics have several drawbacks however which limit their utility. These include a short duration of action, effects on non-nociceptor nerves including mechanoreceptors and motor neurons as well as the potential for systemic CNS and cardiac toxicity.

In order to selectively silence nociceptors involved in cough, pain and itch we have developed charged sodium channel blockers that use large pore channels (e.g. TRPV1, TRPA1 and P2X) expressed in nociceptors to specifically enter nociceptors where they block sodium channels and thus excitability. This approach allows for the highly-selective targeting of cough or pain-sensing neurons while sparing motor neurons and sympathetic neurons.

This presentation will focus on our ongoing efforts to develop novel topically applied charged Nav channel blockers in order to selectively block nociceptive reflexes involved in chronic pain, cough and itch conditions without causing numbness or altering defensive reflexes like the gag and swallow reflexes.

Jim Ellis
Chief Scientific Officer
Nocion Therapeutics

Targeting the Nav1.6 channel with protein:protein interaction based allosteric modulators for pain management

Pain is most commonly initiated by firing of peripheral sensory neurons that transduce painful stimulation into electrical signals through activity of voltage-gated Na+ (Nav) channels. Nav1.6 channels amplify response to painful stimuli (hyperalgesia) through sustained firing of sensory neurons. Studies have shown that two splice isoforms of fibroblast growth factor 13 (FGF13), an accessory protein that binds to the Nav1.6 intracellular C-terminal tail differentially regulate channel function with FGF13-1a reducing pain while FGF13-1b exacerbating it. To develop new effective therapeutics against hyperalgesia, we used a minimal functional domain (MFD) approach to rationally design compounds targeting the protein-protein interaction (PPI) interface between the Nav1.6 channel and FGF13. This approach led to the discovery of PW164, a novel lead that inhibits FGF13-1b and acts as a FGF13-1a mimetic. Using a bioluminescence primary assay to reconstitute the FGF13:Nav1.6 C-tail complex in cells, cell-free orthogonal screens, patch-clamp electrophysiology and in vivo behavioral models of pain, we provide evidence for efficacy of PW164 as a novel non-opioid lead with anti-hyperalgesic properties. Strikingly, unlike common local anesthetics, PW164 does not affect normal sensory function, but rather exclusively inhibits enhanced pain (hyperalgesia). Our goal is to build on these results and optimize PW164 toward future clinical applications as an alternative to opioids for pain management.

Fernanda Laezza
Graduate Program Director & Associate Professor, Department of Pharmacology and Toxicology
University of Texas (Medical Branch)

Functional antibodies against human Nav1.7 and their use in modulating pain

In the last decade, genetic studies have shown that mutations in the SCN9A gene, which encodes for the subtype 1.7 of the voltage gated sodium channels family (Nav), produce familial pain disorders. These mutations can be described as gain of function mutations (increase of Nav1.7 activity) which cause paroxysmal extreme pain disorder (PEPD) or loss of function mutations (reduction of Nav1.7 activity) which are linked to complete insensitivity to pain (CIP). Importantly, people totally lacking Nav1.7 have minimal cognitive, cardiac, motor and sensory deficits, supporting Nav1.7 as a valid and indeed attractive target for development of drugs against pain. Small molecules targeting Nav1.7 have demonstrated to be ineffective therapeutics due to their lack of specificity and serious side effects, thus at NRC we decided to develop biologics (mouse monoclonal antibodies -mAbs- and single domain antibodies – llama VHHs) against hNav1.7, which have the great advantage of being extremely selective. However, the majority of antibodies developed against ion channels so far are binders lacking functional efficacy. Using bioengineering analysis we selected a 70 aa peptide from the hNav1.7 protein sequence and produced a recombinant protein to keep this sequence in a 3D conformation. This was then used as antigen for the production of mAbs and VHHs. Next, we used patch-clamp whole-cell recordings of sodium currents on HEK293 cells overexpressing hNav1.7 channels to verify the ability of these anti-Nav1.7 antibodies to inhibit/block the hNav1.7 currents. We found that 4 mAbs and 1 VHH were able to reduce the hNav1.7 currents. To evaluate if the functional in vitro effect of the anti-hNav1.7 antibodies translated into a functional in vivo effect against pain, we tested the 4 mAbs and 1 VHH using a rat Hargreaves model of hyperalgesia. All selected antibodies successfully promoted the reversal of hyperalgesia in this chronic pain model.

Martina Marzia
Senior Research Officer & Team Leader, Electrophysiology in Human Health Therapeutics
National Research Council of Canada

Cultured human dorsal root ganglia (DRG) neurons for preclinical pain research

The translation from preclinical animal pain models to human outcomes continues to be unreliable at best. Cross-species difference in pharmacological and toxicological responses are well documented and extremely common. Over the last several years, AnaBios has pioneered a novel preclinical discovery strategy, which relies on the utilization of primary cells and tissues ethically recovered from organ donors. By combining novel technologies and reagents, which minimize donor organ ischemia-reperfusion damage, with innovative cell- and tissue- interrogation methods, it is now possible to measure, at the preclinical stage, drug effects in human ex vivo preparations. This approach can provide data highly predictive of clinical outcomes and avoids cross species extrapolation risks in development. An additional benefit is the ability to test drug activity in human cells and tissue recovered from patient donors in the relevant pathological state. Finally, the effects measured in the human ex vivo preparations, provide a quantitative assessment of drug potency and therapeutic window and furthermore, can be used to guide dosing during the first-in-human clinical studies. We have applied these methodologies to the advancement of discovery programs in several therapeutic areas. This presentation will include data from AnaBios' unique in vitro imaging and electrophysiological assays with human DRG neurons.

Chris Mathes
Chief Commerical Officer
AnaBios Corporation Inc.

Potent and selective state-dependent Cav2.2 calcium channel modulators with efficacy in preclinical pain models and human tissue

Traditional pain treatments such as opioids are effective at managing pain but often associated with side-effects as well as societal costs related to opioid abuse and addiction. An alternative class of pain targets is ion channels, which regulate cell activity associated with chronic and spontaneous pain without the risk of addiction. In the clinic, non-selective or poorly state-dependent (SD) ion channel modulators (e.g. Nav1.x anti-convulsants, Cav gabapentinoids) exhibit side-effects and poor efficacy, revealing the need for more effective, non-opioid analgesics. We present an overview of an 8 year drug discovery project that identified potent, SD, and selective Cav2.2 ion channel inhibitors with efficacy in native rodent and human tissue. The exemplar lead series compound exhibited potencies of 400 nM and >30 μM for inactivated and resting states of the human Cav2.2 channel, respectively (>75 fold SD), and >30 fold selectivity over Cav gene family members and cardiac ion channels. Compounds from multiple lead series inhibited Cav2.2 currents in native rodent and human DRG neurons with a similar profile to that seen in heterologous cell lines. Promising ADME and PK properties allowed testing in in vivo pain models, where significant reversal of mechanical allodynia was seen after dosing at 3-10 mpk, without side-effects. Thus, achieving the desired in vitro profile of selective and SD Cav2.2 modulators translates into efficacious in vivo analgesics without the side-effects seen with other ion channel modulators.

Marc Rogers
Director & Chief Scientific Officer
Metrion BioSciences

Targeting TRPA1 for Development of Novel Analgesic Drugs

The selective block of transient receptor potential ankyrin 1 (TRPA1) receptor-channels holds great therapeutic promise for the development of a novel analgesic drug for treating pain that is devoid of addictive and abuse potential side effects. TRPA1 is a validated analgesic target, consistent with a human TRPA1 channelopathy pain syndrome. Small molecule TRPA1 antagonists have shown robust and marked efficacy in many rodent models of inflammatory and chronic pain. We have pursued the discovery of peripherally acting TRPA1 antagonists as an effective therapeutic option for treating inflammatory and neuropathic pain. Employing rational drug discovery approach, Algomedix has discovered a novel structural class of small molecules with potent cellular IC50 values that are effective antagonists of both resting and open TRPA1 channels. Antagonist activity in a human recombinant assay was confirmed in a native human TRPA1 cell line. These potent antagonists are highly selective for hTRPA1 among a family of related TRP channels. Importantly, these drugs do not interact with other established pain targets, which include opioid receptor subtypes, cannabinoid receptors, or COX isoforms, other GPCRs, kinases or ion channel safety targets. In addition, these antagonists have similar potency for human and rodent TRP channels. One of the major challenges that has limited TRPA1 drug development has been the need to identify molecules with suitable PK characteristics that will enable either once or twice daily oral dosing in man. Metabolic profiling of compounds in the TRPA1 series identified a principal site of metabolism. A variety of analogs that were created to metabolically block the vulnerable site produced compounds with low clearance and long half-lives. In pharmacokinetic studies, the lead compound demonstrated a long half-life consistent with once daily dosing in man. Based on excellent pharmacokinetic profiles and strong preclinical data, Algomedix is advancing development of a clinical candidate which may provide a first-in-class pharmacologic therapy for multiple pain conditions.

Jeffrey Herz
President & Chief Executive Officer
Algomedix Inc

July 25th, 2019
Ion Channels as Pain Targets II - Ion Channel

Discovering Glycine Receptor Allosteric Modulators for Treatment of Chronic Pain

The α3-subunit-containing glycine receptors (α3GlyR) mediate the inhibitory currents in the spinal dorsal horn, where peripheral nociceptive signals first enter the central nervous system to convey the sensation of pain to the brain. Positive allosteric modulation of glycine receptors enhances the net inhibition in nociceptive signal transduction, ultimately leading to analgesia. We recently demonstrated that this glycinergic mechanism is responsible for the analgesic action of marijuana independent of its negative psychoactive effects. High-resolution structure determination and drug binding analysis by NMR revealed a novel transmembrane domain drug binding site for Δ9-tetrahydrocannabinol (THC) and cannabidiol, two major active components of marijuana. This high-resolution site allows for structure-based discovery of novel compounds as the next generation of analgesics. Combining *in silico* and high-throughput electrophysiology screening, we discovered and tested a group of compounds that were predicted to bind to the THC site in α3GlyR. In vivo measurements in mice with inflammatory pain induced by complete Freund's adjuvant and in rats with neuropathic pain induced by chronic constriction injury showed that a subset of the top-ranked compounds were highly potent and efficacious in alleviating inflammatory and neuropathic pain. We also examined for drug tolerance with repeated doses and found no tolerance or decrease in analgesia for two lead compounds. Both compounds were also tested for mutagenicity via the Ames test and showed negligible activity. In conclusion, we have discovered a novel class of positive allosteric modulators acting at the THC-binding site in GlyRs that produce analgesia without tolerance, mutagenicity, or psychoactive effects. The analgesic action of these novel compounds may have a significant impact on future chronic pain management and help address the current crisis of opioid misuse disorders.

Yan Xu
Peter M. Winter Professor & Vice Chair, Anesthesiology and Perioperative Medicine
University of Pittsburgh School of Medicine

July 25th, 2019
Ion Channels as Pain Targets II - Ion Channel

Structural basis of toxin binding to voltage-gated sodium channels and what it teaches us about voltage-gating mechanisms

Voltage-gated sodium (Nav) channels are responsible for the generation and propagation of action potentials and thereby play an essential role in the functioning of electrically-excitable cells. They also represent important drug targets since modulating them in specific and selective ways could be effective in several neurological disorders including pain, cardiovascular disease, epilepsy, schizophrenia, Alzheimer's disease, etc. Unfortunately, selective Nav channel modulators have been difficult to identify, in part due to an incomplete molecular understanding of binding sites that could enable molecularly selective pharmacology. Using a combination of X-ray crystallography and cryo-electron microscopy techniques, we have recently solved structures of the spider toxin binding site on the second voltage-sensing domain (VSD2) (Xu et al., Cell 2019) as well as the scorpion toxin binding site on VSD4 (Clairfeuille et al., Science 2019). These structures not only reveal the molecular details of these pharmacologically important binding sites, but also reveal details about how voltage-sensors move across the membrane electric field and how these movements couple to the activation and inactivation machinery.

David Hackos
Senior Scientist, Neuroscience
Genentech

July 25th, 2019
Ion Channels as Pain Targets II - Ion Channel

An *in silico* approach towards solving ligand-host interactions in TRPV1 ion channels

Identification of atomic interactions is key for understanding ligand-host interactions towards drug developments. Yet, due to their dynamic nature, bound ligands are often difficult to resolve at atomic resolutions by cryo-EM or other modern structural methods. We have applied an in silico method based on computational structural modeling to capsaicin binding of TRPV1, a classic case of ligand-host interactions and emerging pain target for novel analgesic drugs. Predictions by this method have successfully guided the identification of atomic interactions between TRPV1 and capsaicin, ginger vanilloids compounds, and piperine in black peppers. Initial applications to other ligand-host interactions yield promising results. The approach should be powerful for studying natural and synthetic compounds interacting with ion channels and other membrane receptors.

Jie Zheng
Professor, Physiology and Membrane Biology
UC Davis School of Medicine

July 25th, 2019
Cardiac Research, CiPA & the Impact on Drug Discovery - Ion Channel

High Throughput Electrophysiology: Lessons from CiPA

Recent efforts in standardizing drug development and safety testing have brought many insights into experimental design and analysis, in order to obtain data of the highest quality, reproducibility and reliability. Ion channels have long been targets for various testing of this kind. Here, we focus primarily on the development, applications and proposed guidelines developed from such efforts, by focusing on automated electrophysiology technologies in cardiac safety testing, as done during the Comprehensive in Vitro Proarrhythmia Assay (CiPA) initiative introduced by the FDA. This initiative is focused on proarrhythmia to improve specificity compared to *in vitro* hERG and *in vivo* QT studies. Here, we will focus on combining automated patch clamp (APC), impedance and extracellular field potential (EFP) measurements in order to study cardiac ion channels in cell lines and hiPSC-derived cardiomyocytes (hiPSC-CMs). We will present gathered data emphasizing protocols, ease-of-use and results obtained in this initiative, by providing automated high-throughput systems with cross-site and cross-cell stable recordings. Special focus will be put on discussing various crucial protocol developments for the recording and analysis of hERG ion channels, as well as the other targets of the CiPA panel. We will also summarize views on lessons learned from our comprehensive investigation approaches, including multiple strategies and technologies, which gave us valuable information on possible ways forward.

Elena Dragicevic
Alliance Manager
Nanion Technologies GmbH

July 25th, 2019
Cardiac Research, CiPA & the Impact on Drug Discovery - Ion Channel

Computational Modeling of Cardiac Ion Channels

To be biologically active, small molecule drugs must physically fit into their binding site(s) within their targets. However, in reaching their precise binding locations, drugs may interact with a variety of cellular components of various structures and functions. Recent seminal advances in computer software and hardware render in silico methods increasingly important. They can build detailed atomistic dynamical structural models to predict ligands' off-target interactions. In our lab, we focus on the potential blockade of cardiac ion channels by small molecule drugs, a critical event that can lead to acquired cardiac long QT syndrome (LQTS) and fatal cardiac arrhythmias. We build computational models for the human Ether-à-go-go-Related Gene (hERG) channel, the Nav1.5 sodium channel, the Cav1.2 calcium channel and the KCNQ1 and Kir2.1 potassium channels. The ultimate goal is to investigate their interactions with drugs *in silico* and predict the mode of binding of these drugs and their potential blockage capacity. This talk will provide an overview on the different ion channels models established in our lab and their success in predicting cardiotoxicity for various drugs.

Khaled Barakat
Assistant Professor, Pharmacy and Pharmaceutical Sciences
University of Alberta

July 25th, 2019
Cardiac Research, CiPA & the Impact on Drug Discovery - Ion Channel

Adult Human Ex-Vivo Models for Preclinical Cardiac Safety Assessment of Drugs

Reliable assessment of the cardiac safety of drugs is a critical requirement of pharmaceutical development. However, current approaches have significant limitations, whereby toxic drugs can still escape detection or, potential life-saving therapies are abandoned due to false positive signals. Therefore, AnaBios has established a novel, reliable and predictive, preclinical drug safety paradigm that uses ex-vivo human cardiac tissues and cardiomyocytes derived from donor hearts to provide a predictive assessment of drug-induced cardiotoxicity in human. Groundbreaking translational research demonstrating the potential of the adult human primary cardiomyocyte contractility-based model for the prediction of drug-induced inotropic and pro-arrhythmia risk at early stages in preclinical drug discovery will be presented at the 2019 Precision Medicine & Ion Channel Retreat Conference.

Najah Abi-Gerges
Vice President, Research & Development
AnaBios Corporation Inc.

July 25th, 2019
Cardiac Research, CiPA & the Impact on Drug Discovery - Ion Channel

Gating pore currents underlay a new cardiac channelopathy

Voltage gated sodium channels (Nav) are broadly expressed in the human body. They are responsible for the initiation and the propagation of electrical impulses in excitable cells. They underlie several physiological processes such as the cognitive, the sensitive, the motor and the cardiac functions. Nav1.5 channel encoded by SCN5A gene is the main Nav expressed in the heart. Its dysfunction is usually associated to the development of pure electrical disorders such as long QT syndrome, Brugada syndrome, Sick sinus syndrome, atrial fibrillation and cardiac conduction disorders. However, mutations of Nav1.5 have recently been linked to the development of an atypical clinical entity combining complex arrhythmias and structural heart diseases such as dilated cardiomyopathy (DCM). Several Nav1.5 mutations are linked to DCM phenotypes, but their pathogenic mechanism remains to be elusive. The omega pore might constitute a common biophysical defect for all Nav1.5 mutations located in the channel's voltage sensor domains. The creation of a gating pore may disrupt the ionic homeostasis of cardiomyocytes, this homeostatic imbalance then affects electrical signals, cell morphology and also the function of cardiac myocytes. We therefore suggest that Nav1.5-linked gating pore may underlay cardiac arrhythmias associated with structural heart diseases.

Mohamed Chahine
Professor
Laval University and CERVO Brain Research Centre

July 25th, 2019
Stem Cells in Drug Discovery I - Precision Medicine

Overexpression of mutated ion channels in human induced pluripotent stem cell-derived cardiomyocytes (hiPSC-CM) to model arrhythmogenic diseases

Cellular models developed to better predict proarrhythmic liability of drug candidates include commercially-available human stem cell-derived cardiomyocytes (hiPSC-CM) obtained from healthy subjects.
Cardiac safety assessment, however, should not be limited to preclinical models using "healthy" cellular systems. It is relevant to test drugs in systems recapitulating various cardiac conditions found in the general population. Similarly, modeling diseases in hiPSC-CM can be used for the discovery of novel therapies. Developing an assay using hiPSC-CM to model mutation-specific arrhythmogenic diseases to screen drug candidates represents a real opportunity at a time where personalized medicine is the focus of many drug safety/ discovery programs. The objective of this study was to modify hiPSC-CM (Cor.4U™) by transfecting LQT type 2 (hERG G628S). G628S was generated by side-directed mutagenesis and tagged with the green fluorescent protein or Cherry to identify successfully transfected cells. Ventricular action potentials were recorded from both transfected and non- transfected cells using the perforated patch clamp method at physiological temperatures and at stimulation frequencies of 0.5 to 1.5Hz. The effects of dofetilide (10 nM), a specific hERG channel inhibitor on AP morphology were compared between control and transfected cells. In non-transfected cells, maximum diastolic potential (MDP)= -75±3 mV, AP amplitude (APA)= 117±2 mV, APD90= 332±36ms and APD50= 263±29 ms (n=5). Superfusion of dofetilide resulted in a depolarized RMP (-63±5 mV), shorter APA (98±8 mV), prolonged APD90 (392±60ms, 18% increase) and APD50 (284±33ms, 8% increase) (n=5). In G628S transfected cells, RMP and APA were unchanged whereas APD50 and APD90 were somewhat increased (281±23ms and 382±29 ms, respectively (n=8). G628S cells were more sensitive to dofetilide than control cells. In dofetilide, APD90 and APD50 were increased by 71% and 23% respectively. In 4 out of 8 cells, Early after depolarizations were recorded. These results suggest that overexpression of mutated ion channels in hiPSC-CM might be used as a model of cellular arrhythmogenic diseases to evaluate proarrhtyhmic liability or efficacy of test molecules.

Marc Pourrier
Vice President, Operations
IonsGate Preclinical Services Inc

July 25th, 2019
Stem Cells in Drug Discovery I - Precision Medicine

Rescue of In Vitro Drug-Induced Long QT Syndrome Type 2 Using a HERG channel Activator in Human Pluripotent Stem Cell-Derived Ventricular-Like Cardiomyocytes

An important step in drug discovery is the evaluation of cardiac toxicity. Candidate drugs must have minimal effects on the HERG current that is essential for cardiac repolarization. Drugs that block the HERG channel can prolong the QT interval leading to lethal ventricular arrhythmias. The HERG channel is prone to promiscuous interactions with drugs due to easy access to the channel pore. HERG channel activators can shorten repolarization in human pluripotent stem cell-derived cardiomyocytes (hPSC-CMs). One such activator, Ginsenoside, was recently shown to interact with the voltage-sensing domain of the HERG channel to stabilize the activated state. Ginsenoside may change the conformation of the open channel and potentially limit subsequent drug blockade. Human PSC lines were differentiated into cardiomyocytes using the STEMdiff™ Cardiomyocyte Differentiation Kit. Excitability of hPSC-CMs was assayed using microelectrode array between days 22 and 25 in the presence of 10 nM E-4031 with and without 10 µM Ginsenoside. Differentiation of the hPSC lines produced a population of beating cardiomyocytes with >80% cTnT-positive cells. A shift in gene expression to ventricular-like cardiomyocytes from day 15 to day 30 was observed (*Myl2/Myl7* = 14.2 at day 30). Electrophysiological experiments were performed between days 22 and 25 on hPSC-CMs. Application of 10 nM E-4031 for 10 minutes prolonged the field potential duration (FPD) and reduced the repolarization signal amplitude. The sequential addition of 10 µM Ginsenoside for 10 minutes shortened the FPD and increased the repolarization signal amplitude. Ginsenoside restored cardiomyocyte excitability similar to pre-treatment with E-4031. Ginsenoside rescued drug-induced LQTS2 in hPSC-CMs, suggesting that HERG activators which target the voltage-sensing domain may be used to offset cardiac toxicity of promising candidate drugs.

Vincenzo Macri
Senior Research Scientist
STEMCELL Technologies

July 25th, 2019
Stem Cells in Drug Discovery I - Precision Medicine

Development of stem cell based cardiac pacemaker cells for in vitro drug testing

A regular heart rhythm and the coordinate and reliable functioning of the cardiac pacemaker system are of fundamental importance for cardiovascular performance. To study cardiac pacemaker cells *in vitro* we programmed murine embryonic (ESCs) and human induced pluripotent stem cells (iPSCs) to differentiate into pacemaker cells. To this end plasmid based vector systems were used to express the transcription factor Tbx3 in the PSCs. Both types of stem cells differentiated into spontaneously contracting cells and tended to form cell aggregates. Cells and aggregates could be studied by live cell imaging for many hours. Contraction frequencies of differentiated cells ranged between 0.5 Hz and 2 Hz. Cell aggregates were obviously electrically coupled, since their contractile activity was synchronized. The contractile activity remained stable for up to six weeks in culture. Electrophysiological characterization of the differentiated pacemaker cells revealed action potentials with amplitudes of about 100 mV and short (10 ms) duration interrupted by about 1-s lasting hyperpolarization phases. Cells showed instable membrane potentials drifting into positive direction. Using the fluorescence Ca2+ indicator Fura-2 we could show rhythmic cytoplasmic increases of Ca2+ with similar frequencies as mentioned above and in agreement with the observed contractile activity. Whole cell recordings of membrane currents revealed the presence of large voltage-gated Na+ currents with amplitudes of >10 nA. Attempts to record voltage-gated Ca2+currents were less successful. However, small currents (< 0.5 nA) could be occasionally detected. In conclusion, the applied method of forward programming of iPSC to pacemaker like cells was successful and reproducible. However, action potentials were rather rapid and probably mainly Na+ driven. Further experiments will be required to test whether a combination of transcription factors has to be applied or whether the results are a matter of cellular differentiation and maturation.

Heinrich Brinkmeier
Professor& Chairman, Pathophysiology and Molecular Medicine
University Medicine Greifswald

July 25th, 2019
Stem Cells in Drug Discovery II - Precision Medicine

Overcoming Challenges in CNS Drug Discovery through Developing Translatable iPSC-derived Cell-Based Assays

Using direct reprogramming of iPSCs to generate defined human neural tissue, NeuCyte developed cell-based assays with complex neuronal structure and function readouts for versatile pre-clinical applications. Focusing on electrophysiological measurements, we demonstrate the capability of this approach to identify adverse neuroactive effects, evaluate compound efficacy, and serve phenotypic drug discovery.

Jonathan Davila
Chief Technology Officer & Co-Founder
NeuCyte

July 25th, 2019
Stem Cells in Drug Discovery II - Precision Medicine

Towards more physiological assays: iPSC-derived neurons tested on the 384 channel automated patch clamp platform Qube.

Human induced pluripotent stem cells (hiPSCs) can be differentiated into multiple cell types, including neurons and cardiomyocytes. This permits the establishment of novel, highly predictive human disease models in vitro. Ion channels represent a class of highly attractive therapeutic targets in the nervous and the cardiovascular system, rendering electrophysiological studies of hiPSCs important for their usage in drug discovery. However, such studies have traditionally been limited by the labor-intensive and low-throughput nature of the manual patch-clamp technique. In the present study, we used our automated, 384 channel patch clamp system Qube to develop robust assays for the study of hiPSC - derived neurons.
Three different hiPSC – based neuronal disease models were investigated. These hiPSC-derived neurons were generated from patients presenting either spinal muscular atrophy (SMA), amyotrophic lateral sclerosis (ALS) or severe epilepsy. Voltage - clamp was used to isolate and characterize individual endogenous currents and compare these to currents recorded from cells derived from healthy individuals. Both the SMA and ALS derived neurons exhibited larger voltage – gated sodium (Nav) currents relative to isogenic control neurons. We evaluated the assay with regard to stability and reproducibility and we tested a set of reference compounds, targeting various ion channels on all three disease models including compounds that reversed the increase in Nav current.
Our recordings demonstrate the feasibility of assaying hiPSC-derived neurons on the APC platforms Qube. Altogether, these results can facilitate evaluating the use of hiPSC for early drug development and in extension personal medicine.

Daniel R. Sauter
Laboratory Manager & Application Scientist
Sophion Bioscience

July 25th, 2019
Stem Cells in Drug Discovery II - Precision Medicine

Generation of an induced pluripotent stem cell (iPSC) line from a 16-year old male with autism spectrum disorder (ASD)

Autism spectrum disorder (ASD) refers to a broad range of conditions characterized by challenges with social interaction and communication and by restricted and repetitive behaviors. In this study, urine cells were collected from a 16-year old male with ASD. The urine cells were reprogrammed with the human SKOM transcription factors. The patient has a heterozygous C>T mutation of FCGR1B gene that was confirmed by sequencing analysis. The pluripotent was verified by gene expression and capacity of differentiation towards the three germ layers in vivo and in vitro. The iPSC line will be valuable for further understanding the pathogenesis of ASD and may be helpful for drugs development to treat ASD.

Zhiyuan Li
Principal Investigator
Guanzhou Institutes of Biomedicine and Health

July 25th, 2019
Stem Cells in Drug Discovery II - Precision Medicine

Small molecule drug discovery targeting innate regeneration and repair

In this presentation OxStem's concept of modulating endogenous cell differentiation with small organic molecules will be presented. Its therapeutic utility will be demonstrated in the context of two different disease areas, oncology and neurology, for which proof of concept has already been obtained. While OxStem's oncology programme is focussed on differentiation therapy for acute myeloid leukemia (AML), its neurology programme targets neurogenesis for Alzheimer's disease.

Georg Terstappen
Chief Scientific Officer
OxStem

July 26th, 2019
Keynote Presentation

Whole Genome Sequencing to Study the Role of HPV Integration in Oropharyngeal Squamous Cell Carcinoma

Human papillomavirus (HPV) plays an important role in the development of cervical cancers and a number of other anogenital cancers. HPV is also increasingly playing a role in the development of oropharyngeal squamous cell carcinoma (OPSCC) a cancer of the base of the tongue and the tonsils. In order to study how HPV is involved in OPSCC and to characterize the physical status of HPV in these cancers we have utilized whole genome sequencing strategies. We started with mate-pair sequencing (MP-Seq) on the Illumina platform and using MP-Seq we found that less than half HPV-positive OPSCCS had HPV integrated into the genome. We also found that HPV integration was independent of HPV copy number and that there was much greater complexity in the different roles that HPV could be playing in this cancer. We have now followed these studies up with high resolution whole genome sequencing (WGS) on the BGI sequencing platform. This sequencing revealed that 2/3 of HPV-positive OPSCCs have one (or more) HPV integrations into the genome, but still 1/3 of these cancers only have episomal copies of HPV present. WGS reveals that there are different HPV populations present in many HPV-positive OPSCCs and further challenges accepted models for how HPV is involved in cancer formation. Finally, WGS also has shown that some HPV integration events are associated with amplification of human sequences at the site of HPV integration. The role(s) that HPV plays in the formation of different OPSCCs may be different, and the same may be true of some of the HPV-positive anogenital cancers. The clinical impact of these results (most notably WGS on OPSCC genomes) will also be discussed.

David Smith
Professor, Department of Laboratory Medicine and Pathology
Mayo Clinic

July 26th, 2019
Keynote Presentation

Flux Assays for Ion Channels and Transporters (Novel Area for Cancer Drug Discovery)

Ion channel research has been a major proponent in drug discovery for cancer. Ion channels and transporters (ICTs) play a critical role in a wide variety of biological processes. They are involved in cancer cell proliferation, migration, invasion, and anti-tumor drug resistance. On the other hand, the medicinal value of natural products, such as: animal toxins and plant extracts, has received much attention in recent years and became a toxicological and pharmacological tool for identifying and studying voltage-gated ion channels. Therefore, the development of natural antitumor drugs targeting ion channels is expected to provide a new direction for cancer treatment. We developed Ion Channel Readers (ICR) to assist researchers in screening for new therapeutics with anticancer activity, identify tumor biomarkers, and perform cardiac drug assays.

Dong Liang
Chief Executive Officer & President
Aurora Biomed

July 26th, 2019
Ion Channels in Drug Discovery - Ion Channel

Voltage-Gated Sodium Channels Assemble and Gate as Dimers

Modification in Na+ current (I_{Na}) is known to contribute to both cardiac arrhythmias from acquired heart diseases and inherited cardiac arrhythmias. Since the original cloning of the genes encoding for voltage-gated sodium channels and the recording of its function by patch-clamping, the α-subunit of the sodium channel was thought to be a monomer. However, our studies of mutations found in SCN5A linked to different arrhythmic syndromes led us to question the traditional idea of the sodium channel forming a monomer. In fact, we and others have shown that several Brugada Syndrome (BrS) mutations display dominant-negative effects (DN-effect), which could only be attributed to interaction between α-subunits within multimeric complexes. Similarly, we have shown that the defects of several BrS or LQT3 SCN5A mutations could be rescued by different SCN5A polymorphisms expressed on a separate construct, again supporting the idea of an α-α subunit interaction. We demonstrated using different experimental approaches that sodium channels form functional dimers. We also identified the region modulating the dimerization and found that this physical dimerization results in coupled gating of the sodium channels and involves 14-3-3. We further demonstrated that the biophysical coupling is dynamically modulated. Understanding of the mechanisms involved in channel dimerization and functional biophysical coupling could open the door to new approaches and targets to treat and/or prevent sodium channelopathies and dysregulation of INa in heart failure.

Isabelle Deschenes
Professor of Medicine, Physiology & Biophysics and Biomedical Engineering
Case Western Reserve University

July 26th, 2019
Ion Channels in Drug Discovery - Ion Channel

Development of a novel cell-based assay system for high throughput screening of compounds acting on background two-pore domain K+ channels

Two-pore domain potassium (K_{2P}) channels are thought to be druggable targets. However, only a few agents specific for K_{2P} channels have been identified, presumably due to a lack of efficient screening system. To develop a new high throughput screening (HTS) system targeting these channels, we have established HEK293-based "test-cell" expressing a mutated Na+ channel (Nav1.5) with markedly slowed inactivation, as well as a K^+ channel (Kir2.1) that sets the membrane potential quite negative, close to K^+ equilibrium potential. We found in this system that Kir2.1 block by 100 μM Ba^{2+} application consistently elicited a large depolarization like a long-lasting action potential. This maneuver resulted in cell death, due to the sustained Na^+ influx. The Ba^{2+}-induced cell death was found to be mostly apoptosis, presumably due to Na+ accumulation and/or K^+ loss and subsequent depression of mitochondrial function. When either TWIK-related acid-sensitive K^+ channel (TASK)-1 or TASK-3 was expressed in the test-cells, Ba^{2+}-induced cell death was markedly weakened. Stronger activation of TASK-1 by extracellular acidification further decreased the cell death. In contrast, the presence of K^{2P} channel blockers enhanced cell death. IC_{50} values for TASK-1 and/or TASK-3 blockers acquired by measurements of relative cell viability were comparable to those obtained using patch-clamp recordings. Both blockers and openers of K_{2P} channels can be accurately assessed with high efficiency and throughput by this novel HTS system.

Yuji Imaizumi
Professor, Department of Molecular and Cellular Pharmacology
Nagoya City University

July 26th, 2019
Ion Channels in Drug Discovery - Ion Channel

A signalling pathway centred on hERG1 potassium channel identifies metastatic colorectal cancer patients with a positive response to Bevacizumab.

Tumour hypoxia and the ensuing neoangiogenesis are deeply involved in tumour progression of colorectal cancer (CRC). Their impact on anti-angiogenesis therapies response, in particular Bevacizumab, in metastatic mCRC is still challenging. We previously identified a signalling pathway, centred on the hERG1 potassium channel, linking the tumour microenvironment to hypoxia, which regulates VEGF-A secretion and drives angiogenesis and metastatic spread in CRC preclinical models. The whole pathway was analysed by immunohistochemistry on 80 surgical samples from mCRC patients treated in first line with Bevacizumab and chemotherapy. Data were analysed together with the clinicopathological characteristics of the patients, *KRAS* status, response to Bevacizumab and follow up. All the proteins were expressed in primary CRC samples, with several statistically significant associations (in example hERG1 and VEGF-A, β1-Integrin and HIF-2α, CA-IX and VEGF-A). hERG1, VEGF-A, the active form of HIF-2α (aHIF-2α) and the G3 histological grade showed a positive impact on Progression Free Survival (PFS univariate analysis). hERG1 and aHIF-2α maintained their positive impact on PFS at the multivariate analysis, and contributed to identifying patients with the best response to Bevacizumab. Notably hERG1 behaved as a protective factor for PFS independently on *KRAS* status. Since modulation of hERG channel function is related to CRC progression, motility and adhesion we also investigated hERG electrophysiological features in both HCT116 and Bevacizumab-treated cells. These data support the relevance of hERG1 centred network between the tumour microenvironment and intracellular hypoxia, and indicate that hERG1 and aHIF-2α might help to identify patients who would benefit from Bevacizumab treatment.

Alberto Montalbano
Postdoctoral Researcher, Department of Experimental and Clinical Medicine
University of Florence

July 26th, 2019
Ion Channels in Drug Discovery - Ion Channel

Electrophysiological Properties of Human Chondrocytes Can Yield Insights Into Design of Drug Targets and Discovery Platforms

In functional studies of mammalian articular joints, including humans, the chondrocyte has been a focus of attention based on its ability to synthesize and secrete substances that are essential for lubrication of the joint and extracellular matrix integrity. Application of electrophysiological methods to isolated single chondrocyte preparations has yielded important new physiological information concerning: i) the resting potential, E_m, of these cells, ii) their sensitivity to physiological mechanical perturbations, and iii) both paracrine and autocrine regulation of $[Ca^{2+}]_i$.

We have attempted to define the roles of a combination of K^+ channels that are expressed in chondrocytes for regulation of the E_m and related modulation of ligand-gated (e.g., ATP) conductances and alterations in Ca^{2+} fluxes and $[Ca^{2+}]_i$ levels. We find that in isolated human articular chondrocytes, E_m is governed mainly by contributions of 2-pore K^+ currents and delayed rectifier K^+ conductances. Under pathophysiological challenge or conditions that mimic key aspects of sterile inflammation, both the intermediate and the large conductance variants of Ca^{2+}-activated K^+ channels can significantly alter E_m.

The ability of even small changes in E_m to very strongly modulate $[Ca^{2+}]_i$ (and thus alter secretion), in nonexcitable cells such as chondrocytes can provide novel insights into progressive changes in essential function of these cells in chronic diseases such as osteoarthritis. Patterns of changes in ion channels in combination with surveys of transcript levels and proteomics can be used to guide the design of medium throughput assays for discovery of novel pharmacological agents for joint pain or osteoarthritis. However, details of the isolation and cell culture of these chondrocytes can, by themselves, significantly alter the functional phenotype. This information and the fact that in the articular joint the chondrocyte is functionally linked to its pericellular matrix (forming the chondron) must be taken into consideration in platform design and in interpretation of results.

Wayne Giles
Professor
University of Calgary

July 26th, 2019

Structural and Functional Aspects of Ion Channels and Ion Transporters - Ion Channel

Hunting the Molecular Target Sites for CFTR Potentiators

Major breakthroughs in solving the atomic structures of CFTR make structure-based drug design a real possibility. However, to date the binding sites for CFTR modulators remain unknown. In this study, we first used molecular docking algorism to identify potential binding sites for GLPG1837, a CFTR potentiator that shares a common mechanism of action with the FDA-approved drug VX770. Among the five top-scored binding modes, the two with highest score (sites I and IIN) are located at the interface between CFTR's two transmembrane domains. Using the patch-clamp technique to measure CFTR currents at different concentrations of GLPG1837, we observed little shift of the dose response relationship when residues in the other three lower-scored sites were substituted with alanine. However, glutamate, asparagine and alanine substitutions at position 924 in site I cause a graded right-ward shift of the dose-response curves. On the other hand, the apparent affinity for GLPG1837 is increased in N1138F, N1138Y, N1138L, S1141R and S1141K, mutations of residues located in site I. Among the five aromatic amino acids (F229, F236, Y304, F312 and F931) that compose site IIN, Y304A, F312A, and F931A decreased the affinity for GLPG1837 whereas F229A and F236A produced little change in EC50. These results support the idea that sites I and IIN are the target sites for GLPG1837. Furthermore, the current relaxation time constant upon removal of VX770 is shortened by D924N, Y304A, and F312A but prolonged by N1138L and S1141K, supporting the notion that site I and site IIN also contribute to VX-770 binding.

Tzyh-Chang Hwang
Professor, Department of Medical Pharmacology and Physiology
University of Missouri

July 26th, 2019
Structural and Functional Aspects of Ion Channels and Ion Transporters - Ion Channel

Structure-function, physiology and pharmacology of K-dependent Na-Ca exchangers of the SLC24 gene family.

The *SLC24* gene family encodes five distinct isoforms of K-dependent Na-Ca exchangers or NCKX1-5 proteins, belonging to the CaCA gene superfamily. NCKX proteins play important roles in a wide range of physiological processes in mammals including vision in retinal rod and cone photoreceptors, olfaction, enamel formation in teeth, human skin pigmentation, motor learning and memory, and melanocortin-4-receptor-dependent satiety. I will briefly describe the physiology of NCKX proteins, our work on the structure and function of NCKX proteins and I will describe the first submicromolar and isoform-specific inhibitor of NCKX proteins. Although no crystal structures have been obtained for eukaryotic NCKX proteins, our work on the topology and functionally important residues of the human NCKX2 isoform compare remarkably well with insights obtained from the crystal structure of a distantly related archaebacterial Na-Ca exchanger. To date, no inhibitors that are specific for NCKX proteins have been described hampering the assessment of physiological roles for NCKX proteins. Here, I will present the first submicromolar inhibitor specific to the NCKX4 protein as compared with the other four NCKX isoforms as well as members of the *SLC8* gene family of K-independent Na-Ca exchangers or NCX proteins.

Paul Schnetkamp
Professor, Associate Dean (Research Infrastructure)
University Of Calgary

July 26th, 2019
Structural and Functional Aspects of Ion Channels and Ion Transporters - Ion Channel

SSM-Based Functional Recordings of Cardiac Transporters and Pumps

Transporters are major pharmacological targets, however, their characterization by conventional electrophysiology is challenging, especially for low turnover transporters and proteins from intracellular compartments. We established new functional assay formats for several transporters and pumps relevant in cardiac safety pharmacology.

Plasma and mitochondrial membrane vesicles from CHO cells, human iPSC-derived cardiomyocytes (hiPSC-CMs) and cardiac tissue were purified and immobilized on a solid-supported membrane-based sensor system. Electrogenic transport was then triggered by the application of suitable cofactors or substrates, and measurements performed of the resultant charging of vesicles.

The Na^+/Ca2+-exchanger (NCX) is pivotal in cellular calcium homeostasis, however, has not been extensively investigated as a cardiac pharmacology target. We studied NCX overexpressed in CHO cells and hiPSC-CMs, recording high amplitude current responses and sensitivity to nickel, KB-R7943 and SEA0400. Further investigations on an impedance-based system showed significant beat rate increases of hiPSC-CMs on long term exposure to SEA0400.

NaK-ATPase plays basic cardiac functions, generating sodium and potassium gradients in cells, hence the membrane potential of cardiac myocytes. We investigated ion dependence and pharmacological properties of the NaK-ATPase pump.

Cardiac muscle tissues highly express mitochondrial pumps and transporters. Vesicles of inner mitochondrial membranes purified from pig heart tissue exhibited activity of the proton pumping complexes of the respiratory chain and the mitochondrial ATP/ADP carrier (ANT). This assay was scaled up to a high throughput format and showed dose-dependence of known substrates and inhibitors.

These findings demonstrate the power and reliability of these SSM-based transporter functional assays and their potential in cardiac safety pharmacology.

George Okeyo
Director
Nanion Technologies Inc.

July 26th, 2019
Key Methodologies in Precision Medicine & Ion Channels for Drug Discovery

High-resolution insights in ion channels involved in muscle excitation-contraction coupling

The contraction of both cardiac and skeletal muscle requires a cross-talk between two calcium-permeable channels: L-type voltage-gated calcium channels (CaV) at the plasma membrane, and Ryanodine Receptors (RyR) in the Sarcoplasmic Reticulum (SR). In cardiac muscle, the cross-talk is functional, with calcium ions providing direct feedback mechanisms to the other channel. In skeletal muscle, there is strong evidence for mechanical coupling between the CaV1.1 and RyR1 isoforms. Here I will present our structural investigation of individual domains and auxiliary proteins that modulate both channels. This includes STAC proteins, a new class of proteins that are critical for mechanical coupling, but that can also modulate channel inactivation. STAC proteins are the target for disease-associated mutations that cause severy myopathy and sensitivity to Malignant Hyperthermia. Calmodulin is a protein that can provide calcium-dependent feedback to both channel types, but which is also targeted by arrhythmia-associated proteins. I will show our crystallographic and functional insights into how these proteins engage CaVs, how they interfere with each other's function, and how disease mutations have both structural and functional consequences.

Filip Van Petegem
Professor, Biochemistry and Molecular Biology
University of British Columbia

July 26th, 2019
Key Methodologies in Precision Medicine & Ion Channels for Drug Discovery

Functional and Mutational Analysis with Rosetta and Cyrus Bench: KCNQ1 case study

Ion channels are one of the largest drug targets likely due to their role in regulation and signal transduction. KCNQ1 is a voltage-gated potassium channel associated with cardiac arrhythmias such as Long-QT syndrome. Several structures of the voltage sensing domain (VSD) or the pore domain have been resolved; however, there are no known structures of a resting VSD. In order to understand the mutational effects, the inactive VSD, closed pore state of KCNQ1 was modeled using Rosetta. RosettaCM enables the use of multiple templates in addition to fragment insertion during homology modeling. Knowledge from experimental studies of the charged residue pairing placement in the VSD was used as a filter during model selection. Finally, MolProbity, ProCheck, and PoreWalker were used to further assess the quality of the selected models.

Several methods exist for modeling mutation-induced stability changes from proteins of known structure or homology models; however, many of these methods perform poorly for membrane proteins. The amount of mutant thermostability data that is available for membrane proteins is sparse, making it nearly impossible to train an accurate predictor for the stabilizing effects of mutations. In a membrane protein benchmark, Rosetta outperformed most of the 10 methods assessed and was able to correctly classify nearly 70% of mutations as destabilizing. Combining these modeling efforts may help with identifying disease-associated mutations in the future. Cyrus Bench is capable of rapidly assessing the stabilizing effects of single point mutations using the newest Rosetta scoring function and thoroughly benchmarked protocols.

Amanda Duran
Protein Engineering Scientist
Cyrus Biotechnology

July 26th, 2019
Key Methodologies in Precision Medicine & Ion Channels for Drug Discovery

Importance of fluidic control and precision in serial activation of ionic channels

Extensive understanding of ion channel responses in serial applications of agonists and/or antagonist is dependent upon advanced abilities in solution control and precise fluidic manipulation. These capabilities have traditionally resided in the domain of slow but reliable single cell patch clamp techniques due to technologically limiting factors plaguing most automated patch clamp (APC) systems. Even with the implementation of microfluidics, precise timing in fluidic exchange while consistently applying continuous solution flow has been elusive to most high throughput APC systems, limiting the scope of most drug research in that domain.

By eliminating the need for external liquid handlers for in-assay solution exchange, IonFlux systems have always utilized a unique in-plate continuous perfusion and precise parallel application of compounds while clamping and recording from ensembles of cells in whole-cell or single cell patch clamp formation. Here we present case studies showing among others the effect of continuous flow on high throughput drug binding research in voltage gated channels and the importance of precision in fluidic exchange on stability of highly desensitizing ligand gated receptors.

Ali Yehia
Vice President, IonFlux Product
Fluxion Biosciences

Precision medicine targeting genetically defined CNS disorders using human iPS cell models

Q-State has developed cell-based models of genetically defined CNS diseases using patient-derived and gene-edited iPS cells. These cell models are coupled with optical methods that enable optical stimulation and recording of neuronal action potentials (APs) and sub-threshold voltage changes using genetically encoded proteins: the engineered channelrhodopsin CheRiff enables AP stimulation with blue light and the engineered voltage-sensitive fluorescent protein QuasAr enables high-speed electrical recordings with red light. These components have been combined into a robust, industrial scale platform for characterizing disease-related phenotypes as a means to develop small molecule and antisense oligonucleotide-based therapeutics. Applications of this approach to epileptic encephalopathies, Rett Syndrome and other monogenic CNS disorders will be discussed.

Owen McManus
Chief Technology Officer
Q-State