1st UAlberta MS Centre Research Symposium

June 24th 2016
Allard Family Lecture Hall (Katz 1080)
University of Alberta
# UAlberta MS Centre Research Symposium

## June 24th 2016

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Welcome to the first UAlberta MS Centre Research Symposium!

The understanding of multiple sclerosis together with its causes and treatments remain substantial challenges to scientists and clinicians, in part due to the diverse genetic make-up and environmental influence on individuals with MS. For this reason, we decided to create the MS Centre at the University of Alberta by way of bringing together scientists, clinicians and people affected by MS to advance the understanding of the diagnosis and treatment of MS. Indeed, the University of Alberta has had a long and illustrious track record in the area of MS research and care dating back to the mid 1970’s when Dr. Ken Warren established the first MS clinic and laboratory at the University of Alberta. He was supported by other investigators and today the University of Alberta MS Centre boasts over 25 Faculty members, several of whom will be presenting at today’s Research Symposium.

We are delighted to feature an outstanding program filled with cross disciplinary speakers who investigate a broad range of subjects that are germane to the understanding of multiple sclerosis. Our goal is to provide a forum in which interdisciplinary discussions will promote cross-collaboration between our experts and their trainees in these areas, thereby accelerating progress in the field of MS research. A prime opportunity for a dialogue between students, faculty and people affected by MS will be the panel discussion at the end of the day that will involve individuals from different backgrounds. As mentioned above, the MS Centre was conceived in 2014 to foster research, education and innovative clinical care for multiple sclerosis through an interdisciplinary approach and through building a platform to service a catalyst for the exchange of ideas among scientists, clinicians and trainees at all stages of their careers. We encourage attendees of the Research Symposium to network with each other and our invited speakers in the hope that this free-flowing exchange of ideas will advance their research program and understanding of multiple sclerosis.

We would like to thank everyone who has worked diligently to organize this Research Symposium, particularly Grace Boldireff for her enthusiasm and dedication to its planning, and we are especially grateful for the sponsors for their assistance in supporting this event. We look forward to an exciting meeting that promises exciting scientific debate and enjoyable social interactions.

Our very best wishes,

Christopher Power, MD & Brad Kerr, PhD
Co-Directors of the UAlberta MS Centre
Dr. Bradley Kerr completed a BSc at McGill University and went on to do a PhD in the UK at King's College London. From London he moved to the California Institute of Technology for a post doc and then returned to McGill for a second post doc with Dr. Sam David. Dr. Kerr became a faculty member at UofA in 2007 and is now an Associate Professor in the Department of Anesthesiology and Pain medicine. Dr. Kerr is a co-director of the newly formed UAlberta MS Centre and Assistant Chair for Research in Anesthesiology and Pain Medicine.

Dr. Christopher Power is a Professor of Neurology at the University of Alberta, holds a Canada Research Chair in Neurologic Infection and Immunity and is a Fellow of the Canadian Academy of Health Sciences. He is an internationally recognized clinician-scientist, focused on the causes and treatments of neuroinflammatory diseases. Aside from leading the Laboratory for Neurologic Infection and Immunity, he is also an attending consultant at the University of Alberta Hospital is Co-Director of the University of Alberta MS Centre. He is the author of over 160 peer-reviewed publications and 20 book chapters with an $h$ index of 51.
~ Guest Speakers ~

Dr. Anne H. Cross is a Professor of Neurology and Section Head of Neuroimmunology at Washington University School of Medicine in St. Louis, Missouri, where she has been on faculty for over 20 years. She is a neurologist specialized in the care of patients with multiple sclerosis and similar disorders. Dr Cross was raised in Alabama, attended the University of Alabama School of Medicine. Following neurology residency at George Washington University, she did fellowship training in neuroimmunology at the National Institutes of Health and at Albert Einstein College of Medicine. She led one of the first studies of B cell depletion with rituximab in MS patients, and continues to work to understand the critical role of B cells and their products in the formation of MS lesions. Additional research interests include the use of novel imaging techniques in the CNS to better understand MS lesion development.

Dr. Shannon Dunn is a scientist at the Toronto General Research Institute and Women’s College Research Institute in 2009 and is an Assistant professor in the Department of Immunology at the University of Toronto. She held a Don Paty Career Development Award from the MS Society of Canada. She leads a research program that focuses on how various risk factors for multiple sclerosis (MS) development impact biology to modulate autoimmune risk in an animal model of MS called experimental autoimmune encephalomyelitis (EAE). Currently, she is exploring the role of female sex and the interaction of sex with obesity on EAE development. She is also exploring possible roles for peroxisome proliferator-activated receptors as molecular mediators of sex differences in autoimmune initiation and progression.

Dr. Anthony Feinstein received his medical degree in South Africa at the University of the Witwatersrand. Thereafter he completed his training in Psychiatry at the Royal Free Hospital in London, England, before training as a neuropsychiatrist at the Institute of Neurology, Queen Square in London. His Master of Philosophy and Ph.D. were obtained through the University of London, England. He is Professor of Psychiatry at the University of Toronto.

His neuropsychiatry research focuses on the search for cerebral correlates of behavioral disorders associated with multiple sclerosis, traumatic brain injury, and hysteria (Conversion Disorders). He is past Chair of the Medical Advisory Committee of the MS Society of Canada. In 2000-2001 he was awarded a Guggenheim Fellowship to study mental health issues in post-apartheid Namibia. He is a recipient of a 2012 Peabody Award.
Dr. Garry Wheeler is an Exercise Physiologist and Registered Psychologist in the Province of Alberta. He is currently President of the Alberta and Northwest Territories Division of the Multiple Society of Canada. He has published more than 47 articles in peer reviewed journals during his academic career and has over 80 conference abstracts and presentations in scientific conferences to his name. Garry specialized in two areas of publications: physiology and psychology. His main topics of study were the effects of chronic exercise stress on endocrine profiles and males and retirement in athletes with disability respectively. In addition he has also co-authored and edited a number of text books and chapters of books in the Adapted Physical Activity domain. For his work in the area of disability sports he was awarded the 2007 International Paralympic Committee Scientific Award in Seoul Korea for outstanding research and services to athletes with disability. Other career roles have included: member of the International Paralympic Sports Science Committee; publishing and writing in the health fitness and lifestyle area and Coaching Association of Canada. In the latter area he has been responsible for creating ethics modules for coaches of children in Canada.

Deb Vollrath graduated from the University of Alberta with a Bachelor of Science in Laboratory Medicine in 1996. Her keen interest in research aided in the discovery of the Bordetella Pertussis gene that is responsible for turning this organism’s virulence factors on and off. This discovery has created an opportunity to develop a more potent vaccine for whooping cough. Deb worked as a Laboratory Technologist for a few years prior to pursuing a rewarding career as a Pharmaceutical Sales Representative with both Pfizer and Janssen-Ortho. Deb was working as a Hospital Specialty Representative for Janseen-Ortho when she experienced her first debilitating MS relapse in 2009. Eventually, Deb had to stop working due to her MS symptoms. It was very hard for her to accept this “forced retirement” at such an early stage of a promising career. The silver lining was that by making her health her “full time job”, she was able to spend more time with her husband and two active children. Deb continues to battle MS on a daily basis. Her interest in research and development has her keenly interested in new and advancing drugs/treatments for MS.
Dr. Penny Smyth is a neurologist in the Department of Medicine at the University of Alberta. She went to the University of Alberta for medical school and neurology residency. Then, she moved to Vancouver and completed a 2 year clinical fellowship in multiple sclerosis, supporting her family as a stroke neurologist. For 4 years, she worked as an MS specialist, stroke neurologist and general neurologist in private practice at Vancouver General Hospital. In late 2008, she moved back to Edmonton to become more involved in medical education. Since being back at the University of Alberta, she practices mainly in the area of multiple sclerosis, also working as the associate program director for the Neurology residency program. She does research in medical professionalism and medical education, collaborating in clinical trials and epidemiologic studies as well as MRI research studies.

Dr. Annunziata (Ann) Marcoccia has a PhD in Clinical Psychology from the University of Windsor. She currently works in the psychology department at the University of Alberta Hospital where she provides psychological treatment services to patients and families who are affected by MS and other medical diagnoses.

Dr. Sunita Vohra is a clinician scientist with dual specialties in pediatrics and clinical pharmacology, her research program focuses on patient-centred evidence-based approaches to complementary medicine. She leads innovation in clinical research methods, including N-of-1 trials, active surveillance, and improved outcomes reporting. Her accomplishments have been recognized nationally with the Roger's Prize for Excellence in CAM research, induction into the Canadian Academy of Health Sciences, and internationally, as she has chaired the American Academy of Pediatrics Section on Integrative Medicine and is a Co-Convenor for the Cochrane Collaboration Adverse Effects Methods Group. A Centennial professor in the Faculty of Medicine and Dentistry, and the founding Director of the Integrative Health Institute at the University of Alberta, Dr. Vohra has won over $17M in research funding and published over 160 peer-reviewed manuscripts.
Dr. Alan Wilman, PhD is a Professor in the Dept. of Biomedical Engineering, Faculty of Medicine & Dentistry, University of Alberta. His research interest is new MRI methods development and applications to tracking MS disease progression.

Dr. Jens Walter is an Associate Professor and Campus Alberta Innovation Program Chair for Nutrition, Microbes, and Gastrointestinal Health at the University of Alberta. After receiving his doctoral degree from the University of Hohenheim in Germany, he performed postdoctoral research into genetic and metagenomic approaches to study gut microbial ecology at the University of Otago in New Zealand. His main research interests are the investigation of ecological and evolutionary processes that shape host–microbial symbioses in the vertebrate gut, and specifically the effect of diet on composition and function of the gut microbiome in the context of health.

Dr. Thomas Simmen’s completed his PhD at the University of Lausanne (Switzerland), where he characterized the mechanism that was later identified to sort between axonal and dendritic proteins in neurons. Subsequently, he completed postdoctoral studies at the San Raffaele Hospital in Milan/Italy and Portland/OR, where he studied proteins that determine ER-mitochondria calcium singling in a redox-specific manner. His laboratory focuses on the characterization of connections between the endoplasmic reticulum (ER) and mitochondria and their role in disease. Dr. Simmen’s is currently interested in understanding how mitochondria influence ER homeostasis and how ER stress triggers ER-mitochondria calcium exchange and eventually apoptosis.
Sabrina Apel is a third year neurology resident at the University of Alberta. Her professional interests include medical education, and the intersection of neurology and palliative care. She hopes to pursue a fellowship in Palliative Care following residency, and be on the forefront of helping to recognize Neuro-palliative care as a core component in neurodegenerative disease case. In her free time, she enjoys camping, hiking, and scuba diving.

Brienne McKenzie is a PhD candidate in Dr. Christopher Power’s lab at the University of Alberta. She completed her Masters in cancer biology with Dr. Peter Forsyth at U of C, before working as an International Visiting Scholar in Experimental Neuro-Oncology at the Moffitt Cancer Center in Tampa, Florida. Brienne joined the Power lab in 2013, and began her PhD program in Medical Microbiology and Immunology in 2014. She has ten peer-reviewed publications, and has received funding from the MS Society of Canada, Canadian Institutes of Health Research, and Alberta Innovates Health Solutions. Her doctoral studies investigate the role of inflammasome activation in the brain during multiple sclerosis and experimental autoimmune encephalomyelitis.
P01 – Sabrina Apel, University of Alberta
The what and when of palliative care for multiple sclerosis: a scoping review
Apel, S., Carroll, L., Johnston, W., Smyth, P.

**Background:** Multiple Sclerosis (MS) is viewed as a chronic, rather than terminal illness, however MS has been shown to be a life-shortening disease. While the time course of MS is highly variable, approximately 75% of patients will go on to have a progressive disease course. Care in progressive MS is not aimed at curative intent, but rather with the goal of alleviating suffering from symptoms, as well as providing patient and caregiver support, similar to palliative care offered for other terminal illnesses such as cancer. Thus, there is a defined need for palliative care in MS populations, however what defines palliative intervention in MS is unclear, as is the timing for this intervention. We have undertaken a scoping review aimed at identifying the literature describing what palliative care is for MS and when it is initiated.

**Methods:** Six electronic databases were searched using keywords related to Multiple Sclerosis, palliative care, and terminal or end-of-life care.

**Results:** The search yielded 556 possibly relevant papers once duplicates were removed. Of these, 406 were excluded by consensus from two independent expert reviewers due to lack of relevance or specific mention of palliative care in MS, while 150 were found to meet inclusion criteria.

**Conclusions:** Our search revealed several broad categories defining areas of palliative care for MS patients, including issues related to quality of life, psychosocial and spiritual support, caregiver relief, symptom burden and management, advanced care planning, and discussion around euthanasia and/or physician-assisted suicide. The literature was mainly comprised of expert opinion, with little evidence-based components to MS palliative care. There was little consensus or mention of the optimal timing of palliative care intervention in MS. We conclude that despite the need for palliative care strategies in MS treatment, there is minimal evidence for palliative care involvement in MS populations.

P02 – Curtis Benson, University of Alberta
Altered mitochondrial protein expression within the dorsal spinal cord during early EAE

Curtis Benson1, Saad Yousuf1, Kasia Zubkow, Gustavo Tenorio, and Bradley J. Kerr*1, 2
1Neuroscience and mental health institute, 2Neurochemical Research Unit, Department of Psychiatry, and 3Department of Anesthesiology and Pain Medicine, University of Alberta, Edmonton, AB, Canada.

In addition to impaired motor function, multiple sclerosis (MS) is also associated with a high incidence of neuropathic pain. Previously our lab has shown that the animal model experimental autoimmune encephalomyelitis (EAE) displays neuropathic pain behaviours before and at the onset of clinical signs. We have recently observed that an exercise regimen in mice with EAE can diminish pain hypersensitivity in the model. We have found that EAE mice exhibiting reduced pain behaviours also have decreased signs of oxidative stress in the spinal cord. To further characterize this, we have now examined various mitochondrial proteins within the dorsal spinal cord at the onset of clinical signs in EAE. We find an increase in the levels of both the outer mitochondrial membrane protein Tom20 and the complex IV protein (COX IV) within the dorsal horn of the spinal cord compared to control mice. In the superficial dorsal horn, Tom20 can be found in GFAP positive cells and neurons. However, neurally expressed Tom20 appears to be driving the additional increase in this region. Using western blot analysis, we find that the elevated amount of dorsal horn Tom20 is associated with significant changes in the levels of several mitochondrial associated proteins. The expression of Drp1, a protein involved in mitochondrial fission, is increased at the onset of EAE disease. In addition, the phosphorylation of Drp1 at ser616 and ser637 is reduced indicating that the protein is significantly dysregulated. The levels of the fusion protein, MFN2 and the mitochondrial biogenesis regulator, PGC-1α are both significantly decreased within the dorsal spinal cord of EAE animals. These changes suggest that EAE leads to a severe disruption in mitochondrial function within the dorsal spinal cord.

P03 – Stephanie Black, University of Calgary
P04 – Roobina Boghozian, University of Alberta

The neurosteroid DHEA shows altered expression and signaling in multiple sclerosis.

Roobina Boghozian1, 4, Brienne McKenzie1, Farshid Noorbakhsh4, Ninad Mehta1, William Branton3, Glen B. Baker2, Christopher Power1, 2, 3

Background: Multiple sclerosis (MS) is an inflammatory demyelinating disease of the central nervous system (CNS) influenced by immunologic, genetic, and environmental factors. Previous studies have reported neurosteroids as protective and anti-inflammatory mediators in neurological disorders. We investigated the expression and actions of dehydroepiandrosterone (DHEA) in MS and associated experimental models.

Methods: Gene expression was quantified by real-time RT-PCR and western blotting in human and mouse CNS tissues and cells. Neurosteroid levels in tissue were quantified by liquid chromatography-mass spectrometry (LCMS). ELISA for IL-1β and IL-6 was performed using supernatants from cultured, activated and DHEA-treated human macrophages and microglia. C57BL/6 mice (10–12 weeks old) were immunized with MOG35-55 peptide emulsified with Complete Freund’s Adjuvant (CFA) to induce experimental autoimmune encephalomyelitis (EAE). At disease onset, EAE animals received daily intraperitoneal DHEA sulfate (DHEA-S) (10 mg/kg). Immunostaining was performed on human and mouse CNS tissues.

Results: MS and EAE CNS tissues displayed reduced DHEA levels, accompanied by diminished CYP17A1 expression compared to controls. CYP17A1-encoded transcript and protein were highly expressed in cultured human astrocytes and neurons. DHEA-S treatment suppressed the release of IL-1β from activated microphages and microglia but did not affect overall leukocyte viability. DHEA-S treated animals with EAE showed reduced inflammatory transcripts and protein levels in association with less demyelination in spinal cord, relative to untreated EAE animals. Neurobehavioral scores deficits in EAE were reduced in DHEA-S treated animals compared with untreated animals with EAE.

Conclusion: The enzyme responsible for DHEA synthesis, CYP17A1, is suppressed during inflammatory demyelination in MS and EAE. Provision of DHEA-S as therapy resulted in less severe inflammation and demyelination with associated improvement in neurobehavioral performance. These results revealed the impact of DHEA-S as a potential therapy for MS and other neuroinflammatory disorders.

P05 – Rachael Chan, University of Calgary

Sex Differences and Dose-Response Relationships in Experimental Autoimmune Encephalomyelitis

Rachael Chan, Stella Babatunde, Dennis Bettenson, Jessica Franken, Vaibhav Singh, Brian Ficiur, V.Wee Yong, Gerlinde A.S. Metz

Multiple sclerosis (MS) is an inflammatory, demyelinating disease of the CNS, with a high prevalence in Alberta. A common method of studying the pathology of MS is the use of animal models of experimental autoimmune encephalomyelitis (EAE). The present experiments investigate (1) a dose-response relationship between the EAE-inducing reagent myelin oligodendrocyte glycoprotein (MOG) and the severity of the resulting clinical symptoms in female C57BL mice; (2) compare the clinical severity of EAE among female and male mice.

Experiment 1: Females. Nine female mice were separated into three groups, each subcutaneously injected with 50 µl, 100 µl, or 150 µl of 1 mg/mL MOG solution to induce EAE. The behavioural test battery included open field exploration task, a skilled walking test, elevated plus maze, and Von Frey hair test. Experiment 2: Males. Ten male mice were induced with 100 microliters based on the latter experiment’s lack of a dose-response relationship. The behavioural test battery included open field exploratory activity, light/dark box anxiety, and plantar nociception test. These longitudinal studies recorded baseline and regular post-induction measurements of motor function, anxiety-like symptoms, and mechanical or heat nociception sensitivity. Over the course of eight weeks post-induction, all female animals showed EAE progression. The severity of the symptoms, however, was dose-independent. Conversely, only two of the ten males showed EAE progression after induction. In addition, some animals experienced a second attack and resembled multiphasic EAE models, as opposed to a single attack in typical monophasic models of EAE. Motivation, anxiety, and motility were best observed utilizing open field exploration, elevated plus maze, and ladder rung test, respectively. Nociceptive results can be equally attained by either the Von Frey hair and the plantar nociception tests. These results, and the test battery we have developed, set the stage for our future experiments.
P08 – Ahmed Elkhady, University of Alberta
Localized analysis of iron accumulation and demyelination in the deep grey matter in Multiple Sclerosis

INTRODUCTION Iron accumulation and demyelination are common features in deep gray matter (DGM) of Multiple sclerosis (MS) patients, as shown by histochemical analysis of MS formalin-fixed brain samples. Magnetic Resonance Imaging (MRI) evaluation of DGM iron levels has been demonstrated as a sensitive marker for MS disease progression using phase imaging, Quantitative Susceptibility Mapping (QSM), R2 and R2* mapping. While the focus of previous MRI studies has been interpretation of DGM iron accumulation in MS by analyzing increases in R2* and/or Q5, MRI assessment of postmortem MS brains has demonstrated that delineation of iron accumulation from demyelination in DGM can be estimated. This can be achieved with the combined use of Q5 and R2* mapping, where a uni-directional increase indicates iron accumulation and a bi-directional change (with R2* decreasing) is suggestive of demyelination.

PURPOSE The purpose of this study was to (1) create a reliable and automated framework for localized analysis of iron accumulation of the DGM, and (2) apply this framework to comparatively evaluate MS phenotypes relative to healthy controls (HC).

METHODS Sixteen patients diagnosed with clinically isolated syndrome (CIS), 94 Clinically Definite MS (CDMS) patients (41 relapsing-remitting (RR), 40 secondary-progressive (SP), 13 primary-progressive (PP)), and 75 age-matched HC subjects underwent multi-echo gradient echo acquisitions, from which Q5 and R2* maps were computed.

RESULTS Preliminary results identified DGM clusters that were significantly different in MS compared to HC, which demonstrated that iron accumulation progressively increases throughout the different stages of MS. Changes in MS DGM R2* and Q5 was found to be mostly explained by iron accumulation, whereas demyelination was found to be insignificant in DGM. In conclusion, we have developed an automated pipeline for analysis of iron accumulation and demyelination in the MS brain. We have also demonstrated the progressive iron accumulation that occurs throughout the disease course.
P09 – Emma Frieser, University of Alberta
Investigating the Effects of N2-Ac-PLZ in EAE
Emma Frieser, Dr. Bradley Kerr

Chronic pain is a highly prevalent symptom in Multiple Sclerosis (MS) that affects over half of patients at some stage of the disease. MS affects females at a higher rate than males and neuropathic pain is also reported at a much higher level in females. The underlying causes of neuropathic pain remain to be elucidated and it is unclear why females are more greatly affected. Previous work in our lab has demonstrated that the non-selective and irreversible monoamine oxidase inhibitor (MAOI), phenelzine (PLZ) and its derivative N2-Ac-PLZ, can decrease formalin evoked pain behaviours but interestingly, N2-Ac-PLZ was only effective in males. Given these sex-specific actions in the formalin model, we wanted to determine whether the effects of N2-Ac-PLZ would have similar sex-specific effects in the EAE model. Male and female mice were induced with 50ug of MOG35-55 and treated with 39.82 mg/kg N2 every other day. Von Frey hair thresholds and Hot Plate were used to assess mechanical and thermal hypersensitivity, Open Field and Elevated Plus Maze were used to measure anxiety, and Rota-Rod was used to assess motor behaviour. Surprisingly, N2-Ac-PLZ appeared to reduce hypersensitivity in the Von Frey assay in the onset of EAE in females, as well as producing a possible analgesic effect in the female controls. N2-Ac-PLZ also produced a small delay to the onset of disease in females, and decreased their clinical scores. The findings imply that, unlike the formalin model, N2-Ac-PLZ may have a positive effect on outcomes in female mice with EAE.

P10 – Yohannes Hale, University of Alberta

Establishing a model for the study of alternate autoimmunity post anti-CD52 treatment

Yohannes Haile and Colin C. Anderson
Alberta Diabetes Institute, Department of Surgery, University of Alberta, Edmonton, Canada

Introduction: Alemtuzumab is an anti-CD52 humanized antibody that treats multiple sclerosis (MS) by depleting lymphocytes leading to a temporary state of lymphopenia with a subsequent repopulation. Alemtuzumab is a promising therapy, however, more than 40% of alemtuzumab treated MS patients develop alternate autoimmune events with a major prevalence in thyroid autoimmunity. Hence, this study aims to develop an animal model of alternate autoimmunity post anti-CD52 treatment, and investigate the underlying mechanisms that led to the generation of this adverse effect.

Methods: We induced experimental autoimmune encephalomyelitis (EAE) in 8-12 week old Rag2p-GFP and Rag2p- GFPxPd-1 KO B6 mice that express GFP in newly generated T and B cells. Mice were treated with 10 mg/Kg/mouse anti-mouse CD52 (anti-muCD52) antibody on 5 consecutive days when they started to show symptoms of the disease. The baseline data, depletion and repopulation of lymphocytes were monitored using flow cytometry.

Results: Anti-muCD52 treatment reversed the severe symptoms of EAE mice to a clinical score grade 0 at the end of the 5-day treatment course. Interestingly, PD-1 KO EAE mice demonstrated a persistently high clinical score of 3; nevertheless, anti-muCD52 was just as effective in reversing this severe disease. Similarly, weight loss in EAE mice in both groups also followed the same pattern. This was associated with the fact that anti-muCD52 significantly depleted B, T and to a lesser extent natural killer cells within one week of the anti-muCD52 treatment in all EAE induced and non-EAE healthy control mice. However, the state of lymphopenia was reversed within about 4-6 weeks post mouse anti-muCD52 treatment. While T cells were not yet 100% back to the baseline, B cells recovered more rapidly, returning near baseline. In both wild type and PD-1 KO mice, the proportions of CD4+, CD8+ and memory cells post anti-muCD52 treatment returned nearly back to the baseline levels. While the percentage of Foxp3 expressing regulatory T cells increased, the absolute number remained low. On the other hand, recent thymic emigrants (RTE) cells appeared more resistant to anti-muCD52 mediated depletion.

Conclusion: The data show for the first time that anti-muCD52 efficacy is not dependent on the PD-1 signaling pathway of tolerance. The existence of low levels of Tregs and resistance of RTE to anti-muCD52 could together contribute to the development of autoimmune events.
~ Abstracts ~

P11 – Samuel Jensen, University of Calgary

Activation of oligodendroglial PGC1α by exercise accelerates remyelination

Treatment modalities that promote the functional regeneration of myelin, termed remyelination, remain in their infancy despite considerable investigation. In multiple sclerosis, physical exercise and participation in activities of daily living are associated with reduced disability with little mechanistic rationale. Considering environment-derived stimuli and activity are known to stimulate myelination in the healthy central nervous system, we investigated the capability of physical exercise to promote remyelination during pathology. We show that immediate therapeutic access to a running wheel enhances oligodendrocyte generation following a lysolecithin-induced demyelinating insult. We observe a 39% and 30% increase in oligodendrocytes at 7 and 14 days post lesion (dpi), respectively. At 7 dpi, 50% of all oligodendrocytes are PDGF+ progenitors and label for the proliferation marker Ki67. At 14 dpi, these newly formed progenitors functionally differentiate into CC1+ mature oligodendrocytes, with progenitors making up <20% of all oligodendrocytes. Moreover, exercise increases the capacity for individual oligodendrocytes to form myelin segments resulting in a 2.7 fold increase in the number of myelinated axons at 14 dpi and accelerates the thickening of myelin. We identify PGC-1α as an exercise-induced factor in oligodendrocyte progenitors, mature oligodendrocytes, and select astrocytes. Further, RNA sequencing of micro-dissected lesions reveals increases in key metabolic genes as a result of exercise. We suggest this is a PGC-1α-driven effect, as it is known to coordinate the transcription of metabolic genes in other contexts, and that this increased metabolic capacity in oligodendrocytes enhances remyelination. We are currently investigating this link further. Overall, this study demonstrates that physical exercise is an efficacious means to enhance remyelination and describes a novel PGC-1α-dependant mechanism through which oligodendrocytes respond to physical activity and myelination is coupled to metabolism.

P12 – John Kmech, University of Alberta

Deep Gray Matter Changes in Multiple Sclerosis

Jonn Kmech1, Esther Fujiwara1, Dana Cobzas2, Hongfu Sun3, Peter Seres3, Gregg Blevins4,5, Alan H. Wilman3

1 Department of Psychiatry, 2 Department of Computer Science, 3 Department of Biomedical Engineering, 4 Department of Medicine, Division of Neurology 5 Northern Alberta MS Clinic

Background: Deep grey matter (DGM) iron accumulation is increasingly recognized in association with Multiple Sclerosis (MS) and can be measured in-vivo with MRI. Cognitive implications of this pathology are not understood well, especially vis-à-vis DGM atrophy.

Objectives: To investigate relationships between cognition and DGM iron in MS using two MRI-based iron measures.

Methods: 40 MS patients (EDSS = 5.25; relapsing: N = 16; progressive: N= 24) and 27 controls were imaged at 4.7T using transverse relaxation rate (R2*) and quantitative susceptibility mapping (QSM). R2* and QSM values and volumes of caudate, putamen, globus pallidus, and thalamus were determined by multi-atlas segmentation. Cognition was assessed with the Brief Repeatable Battery of Neuropsychological Tests. Relationships between cognition and DGM iron were examined by hierarchical regressions.

Results: Compared to controls, patients showed reduced memory and processing speed, smaller putamen, globus pallidus, and thalamic volumes, and increased QSM indicative of iron accumulations in putamen and globus pallidus. Thalamus and putamen volume predicted cognition in patients. Controlling for atrophy, QSM values in the globus pallidus also predicted cognition.

Conclusions: QSM was more sensitive compared to R2* in detecting DGM iron accumulation. DGM atrophy and DGM iron have negative and separable relationships to cognition in MS.
P13 – Wei Liu, University of Calgary

Randomized, Controlled Pilot Trial of Domperidone in Relapsing-Remitting Multiple Sclerosis
Wei-Qiao Liu1,3, Simon Zhornitsky1, Jamie Greenfield1, Rand Pasha1, Graziela Cerchiaro Farah1, Yunyan Zhang1,2, G. Bruce Pike1,2, V. Wee Yong1, and Luanne Metz1
1Department of Clinical Neurosciences, University of Calgary
2Department of Radiology, University of Calgary
3Division of Neurology, Department of Medicine, University of British Columbia

OBJECTIVES:
We are conducting a phase 2, single-centre, randomized, controlled trial comparing domperidone add-on therapy vs no add-on therapy in (relapsing remitting multiple sclerosis) RRMS patients taking disease modifying therapy (DMT) and who have breakthrough lesions identified on MRI monitoring. Our primary objectives are: (1) To demonstrate that we can recruit to this trial; and (2) to obtain estimates of the magnitude and variability of lesion repair over 32 weeks.

BACKGROUND:
Prolactin enhances remyelination in animal models but moving to clinical trials is challenged by the lack of a marketed prolactin and lack of a clinical model to evaluate lesion repair, other than possibly optic neuritis. While prolactin can be produced we have chosen to use domperidone, a safe, inexpensive drug marketed to enhance gastric motility because it also raises serum prolactin levels sufficiently to be used off-label to improve lactation.

METHODS:
Consenting RRMS patients aged 18-60, taking an approved DMT, and shown to have breakthrough gadolinium enhancing lesions on DMT monitoring MRI, will be randomized (2:1) to domperidone add-on treatment 10 mg three times daily or no add-on treatment. After screening and baseline visits, follow-up will be at 6, 16, and 32 weeks. MRI scans will be obtained at baseline, 16 and 32 weeks. In addition to routine MRI sequences, we will evaluate and compare three MRI measures [texture analysis, diffusion tensor imaging (DTI), and magnetization transfer imaging (MTI)] for their ability to measure repair within acute enhancing lesions. We aim to enrol 24 patients over 36 months.

RESULTS: Recruitment is ongoing. Between November 2015 and April 2016 we screened 25 patients; 12 more are scheduled for screening. One patient has been randomized to domperidone and has tolerated treatment without any adverse effects. As expected, the majority of screen failures are due to the absence of enhancing lesions on MRI.

CONCLUSIONS:
We intend to determine if this trial model is appropriate for studying lesion repair in MS and determine if any of these imaging methods are sensitive enough to measure repair within enhancing lesions. We will present the study design and updated recruitment data. Recruitment was slow in the first few months but many more patients are currently being scheduled for screening. We anticipate that the results from this trial will inform the design of future phase 2 trials of therapies to promote lesion repair in MS.

P16 – Brienne McKenzie, University of Alberta

Central nervous system inflammasome activation during multiple sclerosis and experimental autoimmune encephalomyelitis
Brienne McKenzie, Manmeet Mamik, Roobina Boghozian, William Branton, Leina Saito, Christopher Power

Background: Inflammasomes are multi-protein signaling platforms that mediate maturation of pro-inflammatory cytokines (IL-1β, IL-18) by caspase-1/4. Initially identified in peripheral myeloid cells, inflammasome activation has recently been observed in microglia, the resident macrophages of the central nervous system (CNS), as well as in astrocytes and neurons. The goal of our study is to assess the contributions of CNS inflammasome activation to MS pathogenesis. Methods: Using qRT-PCR and IHC, individual inflammasome genes were analyzed in cerebral white matter autopsy samples from MS and non-MS patients, and in the CNS from MOG/CFA-induced experimental autoimmune encephalomyelitis (MOG-EAE) mice. To modulate inflammasome activation, MOG-EAE mice were treated with the caspase-1/4 inhibitor, VX-765 or vehicle. Efficacy of VX-765 was validated ex vivo in human microglia stimulated with inflammasome-activating stimuli (e.g. ATP, LPS). Results: Inflammasome-associated genes (e.g. caspase-1/4, IL-1β, NLRP3, NLRP1) showed higher transcript levels in white matter of MS (n=15) versus non-MS patients (n=12) (p<0.05). IL-1β, NLRP3 and caspase-1 p10 immunoreactivity was detected within MS lesions. In MOG-EAE, inflammasome-associated genes were induced in the CNS sequentially over the course of disease (p<0.05). Increased IL-1β and caspase-1 immunoreactivity was evident in spinal cord lesions at peak disease. Inhibition of the inflammasome with VX-765 suppressed EAE severity (p<0.05). Conclusion: Multiple inflammasome genes were activated in CNS myeloid cells during both MS and EAE, and inflammasome modulation by a caspase-1/4 inhibitor was protective in vivo. Understanding the pathogenic contributions of CNS inflammasomes may unveil novel therapeutic targets in MS.
**Abstracts**

**P15 – Georgina MacIntyre, University of Alberta**

TAGC: Simona Veniamin 1, Rachel Brown 1, Georgina Macintyre 1, Andrew L. Mason 2.

**Compositional diversity of the bile microbiome in PBC and PSC patients**

**Introduction:**

The Applied Genomics Core (TAGC) provides services related to nucleic acid extraction and analysis, including sample quality control, PCR, qPCR, ddPCR and Sanger and Next Generation Sequencing (NGS). We assist with the development of NGS workflows from submitted DNA and RNA samples through library generation to NGS data generation for analysis on custom-built bioinformatics pipelines for microbial profiling or RNAseq.

**Shotgun metagenomics at TAGC:** DNA samples undergo integrity checks (e.g., Qiaxcel) prior to library generation. Libraries are evaluated using the Agilent Bioanalyzer for size-distribution and Qubit for concentration estimates, prior to sequencing (Illumina MiSeq). The resultant fastq files can be analyzed by the end-user or using our bioinformatics pipeline for microbial profiling.

**Project highlight:** Microbial profiling of bile samples obtained from patients with Primary Biliary Cirrhosis (PBC) or Primary Sclerosing Cholangitis (PSC).

**Methods:** Study panels comprised 7 PSC, 8 PBC, and 10 non-PBC non-PSC controls. Gallbladder bile was collected from livers removed from patients. Extracted DNA was used to create libraries for NGS (Illumina). Paired-end reads were trimmed, filtered, and aligned to a bacterial database. Reads (>97% identity) were assigned to a bacterial family (BLASTn). Naive bayes/decision-tree hybrid classification (10-fold cross validation) was applied to identify bacterial families that distinguish PBC, PSC and control bile samples.

**Results and Conclusion:** Staphylococcaceae and Streptococcaceae were reduced and Vibrionaceae, Rhodocyclaceae, and Oxalobacteraceae were increased in bile from patients with PBC compared to both PSC and controls. Bile constituents may influence the microbial profiles of patients with PBC and PSC.

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**P14 – Tim Luo, University of Calgary**

**A Comparison of MRI Methods for Assessing Myelin Property in Mouse Brain**

**Tim Luo, Shurshrita Sharma, Mark Polivchuk, Peng Zhai, Yunyan Zhang**

**Introduction:**

Changes in myelin property are key features of many neurological diseases including MS. Several methods have demonstrated the potential to assess myelin content in vivo using MRI, including magnetic transfer imaging (MTR) and multi-echo T2 MRI (myelin water fraction, MWF). The aim was to compare the utility of these methods with our customized approach, MRI texture analysis, in assessing myelin integrity using mouse brains.

**Method:**

We acquired MRIs from 7 healthy mice with a 9.4T scanner. Whole brain MTR and single-slice MWF images were obtained using published protocols. Texture analysis was based on T2-weighted MRI. Measurements focused on the corpus callosum (CC), a large white matter structure. Outcomes were evaluated both by anatomy (genu, body and splenium) and hemispheres (right, center, and left).

**Results:**

No significant difference between hemispheres of the CC was suggested by any method. While MTR showed a difference between the body and splenium, there were no differences between anatomical structures of the CC in either MWF or texture analysis. Between animals, texture heterogeneity remained consistent with the least variability when compared to MWF and MTR.

**Conclusion:**

All three methods confirmed the uniformity of myelin structure in the CC between hemispheres. Texture analysis showed the best consistency between animals. This may be due to the high quality of T2-weighted MRIs from which texture was derived. However, no methods detected expected differences between anatomical regions, even though it has been indicated that the genu has thinner axons than the body and splenium. Further investigation is warranted.
P17 – Katherine Mifflin, University of Alberta

Sex differences in pain and disease related outcomes in a mouse model of MS

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Multiple Sclerosis (MS) is an inflammatory autoimmune and neurodegenerative disease. Although the primary symptoms of MS include the loss of sensory and motor function, many patients experience secondary symptoms such as chronic neuropathic pain. Recent work from our laboratory has explored voluntary exercise as a possible treatment for chronic pain in the MS mouse model experimental autoimmune encephalomyelitis (EAE). Initial studies were carried out in female mice. We found that daily voluntary wheel running reduced pain at disease onset and diminished dorsal horn microglial activation and T-cell infiltration. Voluntary wheel running also reduced oxidative stress in the spinal cord of female mice with EAE. Although exercise could delay the onset of clinical signs, there was no significant difference in the severity or progression of clinical signs once the disease was established.

In the present study we explored whether daily voluntary wheel running would also be effective at reducing nociceptive behaviour in male mice with EAE. Male mice were given access to a running wheel for 1 hour a day for 40 days. Surprisingly, voluntary wheel running did not reduce mechanical allodynia (tested with von Frey hairs) in males with the disease. We did find however, that unlike in females, daily wheel running significantly improved clinical scores in male mice with EAE. Direct comparison of inflammation, axonal injury and oxidative stress in male and female mice with EAE revealed significant differences in the amount of T-cell infiltration, microglia and astrocyte reactivity, demyelination and axon integrity in males and females with EAE. Male mice with EAE given daily access to running wheels had significantly less ongoing oxidative stress compared to all other groups. Taken together our results indicate that the inflammatory response generated in EAE is distinct between the sexes and its modulation by daily exercise can have sex specific.

P18 – Maryan Nakhaei-Nejad, University of Alberta

In-depth Immunophenotyping of Newly Diagnosed Relapsing Multiple Sclerosis (RRMS) Patients and Patients treated with Dimethyl Fumarate.

Authors: Maryam Nakhaei-Nejad, Gregg Blevins, Aaron Hirschfeld, and Fabrizio Giuliani

University of Alberta, Edmonton, Canada

Multiple sclerosis (MS) is an autoimmune disease of central nervous system. Inflammation and demyelination are two key features of MS. T cells seem to be an important player in MS pathogenesis. However, there is limited information about the subpopulation dynamics in MS patients. Our aim is to assess the changes in peripheral immune responses of MS patients with and without treatments.

We designed two multi-color panels (14 and 17 surface markers) to analyse cell surface expression of immune cells in whole blood. Panels were designed based on antigen density and consideration of spillover characteristics of selected fluorochrome conjugates. Stained samples and controls were run on a BD LSRFortessa™ SORP. We have consistently identified 41 various leukocyte subpopulations within each sample including B cells, monocytes, CD4+ and CD8+, NK cells and also dendritic cells. To date, we have processed 8 healthy controls (HC), 23 relapsing remitting patients (14 treated with dimethyl fumarate (DMF), and 5 newly diagnosed patients (NT). In agreement with previous reports, all MS patients had lower PBMC frequency than HC (41% vs. 30%; p value 0.00161). T cell % was significantly different in NT comparing to DMF group (70% vs. 50%; p value 0.000688). Within the CD3+ population, activated T cell % (CD122+) was also lower in DMF treatment (13.5% NT versus 8.7% DMF; p value 0.0437). We observed lower CD8% in NT group vs. HC (20% vs 35%; P value 0.0109).

While B cell % was not different in DMF treatment compared to NT or HC, the B-cell subtype CD27+IgD- was 16% in HC, 6.5% in DMF (P value 0.0128) and 9.7% in NT (p value=NS versus HC or DMF).

Our data will shed new light on the mechanisms of inflammatory events in MS and also help to identify and predict patients’ response to treatment.
P19 – Liam Potter, University of Alberta
Altered excitatory-inhibitory balance within somatosensory cortex is associated with enhanced plasticity and pain sensitivity in a mouse model of multiple sclerosis
Liam E Potter; John Wesley Paylor, MSc; Jee Su Suh; Gustavo Tenorio, MSc; Jayalakshmi Caliaperumal, PhD; Fred Colbourne, PhD; Glen Baker, PhD; Ian Winship, PhD; Bradley Kerr, PhD.

Chronic neuropathic pain is a common symptom of multiple sclerosis (MS). MOG35-55-induced experimental autoimmune encephalomyelitis (EAE) has been used as an animal model to investigate the mechanisms of pain in MS. Previous studies have implicated sensitization of spinal nociceptive networks in the pathogenesis of pain in EAE. However, the involvement of supraspinal sites of nociceptive integration, such as the primary somatosensory cortex (S1), has not been investigated. We used in vivo flavoprotein autofluorescence imaging (FAI) of S1 to assess cortical responses to a vibrotactile stimulus of the limbs in early EAE, when pain behaviors first become prominent. Mice with early EAE exhibited significantly intensified and expanded FAI responses in S1 compared to controls. Immunohistochemical analysis revealed increased vGlut1 expression and disrupted parvalbumin+ (PV+) GABAergic interneuron connectivity within S1 of EAE mice. Morphological analysis of pyramidal neurons in layers 2/3 and layer 4 of S1 also revealed increased dendritic spine density along the distal dendrites. Cortical microglia were found to be significantly elevated early in the disease. Furthermore, perineuronal nets (PNNs), components of the extracellular matrix that surround and support PV+ interneurons, were significantly reduced in S1. Treatment with the monoamine oxidase inhibitor phenelzine (PLZ), which restores CNS levels of GABA and the monoamine neurotransmitters (5-HT, NA), was found to normalize mechanical thresholds in EAE. PLZ also normalized S1 FAI responses, neuronal morphologies, and cortical microglia levels, and attenuated the increase in vGlut1 reactivity in S1 - but did not significantly attenuate the loss of PNNs. PLZ did not affect basal mechanical thresholds or FAI responses in control animals. These findings implicate a shift in the balance of excitation and inhibition within the CNS in EAE, leading to large-scale functional and structural plasticity in S1.

P20– Khalil Rawji, University of Calgary
Factors that impair remyelination in the aging central nervous system
Khalil S. Rawji, David Tang, Janson Kappen, Michael B. Keough, and V. Wee Yong

Studies using animal models of remyelination demonstrate diminished repair with aging, posing a significant challenge to therapy in white matter diseases. This decreased remyelination is thought to be due to impairment in oligodendrocyte differentiation, and is associated with a delayed inflammatory response. Macrophages/microglia are components of this inflammatory response and are essential to successful remyelination, as these innate immune cells release growth factors and clear inhibitory molecules such as myelin debris. In this study, we have used a focal lyssolecithin demyelination model in young adult (~2 months) and aging (~9-12 months) mice to determine factors that may impair the remyelination observed with aging. We have confirmed that remyelination is decreased with aging and have observed an associated delay in macrophage/microglia recruitment. Impaired clearance of inhibitory myelin debris and chondroitin sulphate proteoglycans are also evident, suggesting an impaired phagocytic response. Using ex-vivo live imaging of CX3CR1-GFP/Thy1-YFP mice with multi-photon microscopy, we have characterized the dynamic activity of aging macrophages/microglia in the young and aging lesions in real time. Using this technique, we have observed a decrease in the number of macrophages/microglia at the lesion site in aging compared to young mice, and aging macrophages/microglia display slower motility. In addition, there are significantly less phagocytic macrophages/microglia in the aging lesion. Pharmacologically activating aging macrophages/microglia to migrate to the lesion and clear inhibitory molecules may promote a more conducive extracellular environment for remyelination in the aging CNS.
~ Abstracts ~

P21 – Leina Saito, University of Alberta

Innate immune responses in human oligodendrocytes depends on maturation.
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Background: Innate immune responses in the central nervous system are widely assumed to be mediated by microglia and astrocytes. Oligodendrocytes are responsible for myelination although their maturation status is an important determinant of remyelination competence. Herein we investigated innate immune responses in undifferentiated and differentiated human progenitor-derived oligodendrocytes (PDOs).

Methods: Differentiated and undifferentiated PDOs were exposed to TNF-α at different concentrations for 24 hr and host responses were analysed subsequently by sqRT-PCR. RNA extracted from normal appearing white matter from MS (n=6) and non-MS (n=6) patients was subjected deep sequencing (RNAseq).

Results: Both differentiated and undifferentiated PDOs exhibited robust induction of Type 1 interferon-associated genes including IRF3 and IRF7 in a TNF-α concentration dependent manner although the gene induction was greater (4 fold) in differentiated PDOs. IFNB was induced in differentiated PDOs while IFNA and IFNL were not detected in PDOs. CASP1 was induced by TNF-α exposure in differentiated PDOs together with NLRP3 induction, while IL-1B was suppressed by TNF-α in undifferentiated PDOs. Deep sequencing of human brain showed that Mx1 expression was highly induced in MS white matter (30 fold) compared to non-MS brain.

Conclusions: Depending on the maturation stage, PDOs display differing innate immune response capacities that might influence cellular survival and potential for remyelination.

P22 – Erin Stephenson, University of Calgary

Chondroitin sulfate proteoglycans in experimental autoimmune encephalomyelitis
Erin Stephenson, James A. Rogers, Michael B. Keough, Manoj Mishra, V. Wee Yong
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Multiple sclerosis (MS) is a chronic inflammatory and degenerative disease of the central nervous system, presenting with profound destruction of myelin and axons that form focal areas of demyelination. MS also presents with changes in the extracellular matrix, a network of molecules involved in maintaining brain architecture. The extracellular matrix molecules observed at the border of active demyelinating lesions in MS are the chondroitin sulfate proteoglycans (CSPGs) (Sobel and Ahmed J Neuropathol Exp Neurol 2001). CSPGs may have the ability to promote activation of immune cell subsets, migration of immune cells into the CNS, and killing of neurons. Thus, we hypothesize that interfering with CSPG production in MS and in an animal model of MS, experimental autoimmune encephalomyelitis (EAE), will improve outcomes. The purpose of this work was to investigate the ability of CSPGs to influence the immune system, and characterize changes in CSPGs over the course of EAE and MS. We also tested whether a CSPG-targeting glucosamine derivative can affect inflammation, neuropathology, and clinical disease scores in EAE. Current results show that CSPGs promoted proliferation of T cells in culture in an activation-dependent manner. At peak severity of EAE in mice, we observed an upregulation of transcripts and protein of the CSPG member versican. Other CSPG members were not significantly altered. Versican was also upregulated in MS tissue, particularly in the vicinity of inflammatory perivascular cuffs. EAE mice treated at onset or peak with a CSPG-lowering glucosamine derivative had reduced EAE severity and reduction of inflammatory transcripts. Thus, CSPGs, such as versican, may promote inflammatory disease in MS and EAE. These results emphasize that reducing CSPGs may prove useful in pathologies such as MS, where the immune system plays a role in central nervous system damage.
Abstracts

P23 – Robert Stobbe, University of Alberta

Exploring Sodium MRI Contrast Beyond Concentration in Multiple Sclerosis Lesions

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Purpose - Standard magnetic resonance imaging (MRI) acquires signal from the hydrogen (1H) atoms in water. However, with proper hardware and methods, images can also be obtained from sodium (23Na) in tissue. Sodium MRI has shown concentration increases in MS lesions and normal appearing white matter (NAWM) [1–5]. In this study we investigate sodium MRI sequences that provide contrast beyond 23Na concentration for the first time in MS. The physics dependencies of these techniques may provide novel micro-structural and metabolic information.

Methods - Images were acquired from 30 MS volunteers (50 ± 10 yr; 19F/11M; 11-RRMS / 10-SPMS / 9-PPMS) on a 4.7T MRI using density-weighted (NaDW) [6], relaxation (NaPACMAN) [7], and fluid-suppressed (NaSIRFLA) [8] 23Na sequences. Sodium image intensity was measured in ~400 lesions outlined on FLAIR images and comparable NAWM. A paired t-test was used to test for statistical difference in lesion contrast between sequence types. Linear regression of 23Na sequence signal with lesion volume was also performed.

Results - Compared to non-lesion brain, the average MS lesion signal was increased by 18% for NaDW and 33% for NaPACMAN, but decreased by 6% for NaSIRFLA – statistically different contrast in each case (p < 1x10^-10). For each sequence 23Na sequence signal correlated with individual lesion volume (p < 1x10^-10).

Discussion and Conclusions - We show that different types of sodium sequences (i.e. NaDW, NaPACMAN, and NaSIRFLA) produce significantly different MS lesion contrast. Note that NaDW increase is consistent with that measured in previous papers [2,4,5]. Sodium concentration (NaDW) increase likely reflects increased extracellular fluid volume fraction and/or altered cellular Na+ metabolism. However, increased NaPACMAN contrast is expected to correlate with loss of macromolecular density and structure [7], while reduced signal on NaSIRFLA points to an increased ‘fluid-like’ nature [8]. Future work will correlate 23Na MRI signal with MS types and disability scores.

P24 – Wulin Teo, University of Calgary

RNA Imaging of myelin in the CNS

W Teo

The maintenance of cellular structure, metabolism and trophic factor transport in axon of neuron is determined by the cell body, however the efficiency of such activities might be slow in long axon. How could a cell body metabolically sustain a long axon comprising more than 100-fold its mass? Recently, long axon in the PNS are supported by the local neighboring Schwann cell. Schwann cells myelinated axon intimately and delivered Ribosomal RNA package into axon for local protein translation. We wonder such cellular activity can be observed in the long axon of the CNS. We performed ex vivo imaging of adult cervical dorsal column of mouse for this study. First, we used ethidium bromide to label myelin, then we performed immunostaining using L4 antibody to detect ribosomal protein in the myelin. We found myelin membrane vesicle injected into the axon after stimulation with glutamate or high potassium concentration in artificial cerebrospinal fluid. We concluded that long axons in the CNS share the similar mechanism with the PNS. Long axons not just dependent on their cell body for cellular maintenance but their neighbor cells oligodendrocytes provide RNA to axon.
P25 – Kevin Thorburn, University of Alberta

Using the EAE animal model to understand MS-related trigeminal neuralgia.

Kevin C. Thorburn, John W. Paylor, Christine A. Webber, Ian R. Winship, and Bradley J Kerr

Trigeminal neuropathic pain is a well-recognized complication of the demyelinating disease multiple sclerosis (MS). The mechanisms underlying MS-related trigeminal neuropathic pain are poorly understood. This can be attributed, at least in part, to the lack of an animal model that exhibits trigeminal pathology similar to that described in MS. Experimental autoimmune encephalomyelitis (EAE) is an animal model that is commonly used to study the pathophysiology of MS. We show here that mice with EAE exhibit increased sensitivity to air puffs applied to the whisker pad. The increased sensitivity to air puff stimulation is accompanied by T cell infiltration and glial activation at several points along the trigeminal primary afferent pathway. We also observe demyelination of the intra- and extra-pontine aspects of the trigeminal sensory root and the spinal trigeminal tract. Interestingly, this pattern of trigeminal demyelination is similar to what has been described in people with MS-related trigeminal neuropathic pain. This is the first study to show orofacial sensory disturbances and trigeminal demyelination in EAE. Collectively, our data suggest that EAE may be a useful model for understanding MS-related trigeminal neuropathic pain conditions such as trigeminal neuralgia.

P26 – Saad Yousef, University of Alberta

Investigating changes in chloride cotransporters in mediating pain in EAE: Implication for neuropathic pain in Multiple Sclerosis

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More than half of Multiple Sclerosis (MS) patients complain of neuropathic pain during their disease course. It primarily arises as a result of hyperexcitability of spinal dorsal horn neurons. Various proteins are important for regulating the balance between excitation and inhibition in the CNS. Notably, the Na⁺-K⁺-Cl⁻ co-transporter 1 (NKCC1) and the K⁺-Cl⁻ co-transporter 2 (KCC2) are crucial for determining the strength and polarity of GABA, the principle inhibitory neurotransmitter in the CNS. Phosphorylation of these proteins (pNKCC1, pKCC2) enhances their transporter activity multifold. NKCC1 on presynaptic terminals in the dorsal horn regulates primary afferent depolarization whereas KCC2 is crucial for mediating disinhibition on projection neurons. An impairment in cytoskeletal proteins may prevent NKCC1 transport to terminals, reducing presynaptic inhibition. To mimic the T-cell mediated inflammation seen in MS, our experiments used the animal model of experimental autoimmune encephalomyelitis (EAE). We aim to identify changes in the expression of chloride co-transporters and cytoskeletal proteins in mice with EAE that may relate to pain hypersensitivity in the disease. PCR analysis of dorsal spinal cord shows a marked decrease in NKCC1 and KCC2 mRNA expression at disease onset and peak. A similar protein expression for NKCC1, KCC2, and their phosphorylated counterparts is found in the dorsal spinal cord. In the DRG, NKCC1 protein levels are elevated at EAE onset although mRNA levels remain relatively constant. An accumulation of NKCC1 in the DRG while a reduction in the dorsal horn may be a result of impaired anterograde transport in DRG neurons. Axon-specific microtubule associated protein, tau, was found to be consistently downregulated after the onset of symptoms. Furthermore, kinesin levels are also decreased at EAE onset only to return back to control levels. These results imply an important role of the cytoskeleton in mediating levels of chloride co-transporters and the resulting hypernociception.
Thank you!

There is only one cure for Multiple Sclerosis: Research