Keynote Speaker

Dr. Peter Dolman

Controversies and Research in Endonasal Dacryocystorhinostomy

Evaluation and Management of Thyroid Orbitopathy

Monday, April 3

2017
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Welcoming Remarks

Today’s event marks the 28th Annual Research Day for the Department of Ophthalmology & Visual Sciences at Dalhousie University. The program reflects how research has grown and been fostered within the Department.

We are honored to welcome our Keynote Speaker and External Guest Judge, Dr. Peter Dolman, Clinical Professor at the University of British Columbia. Dr. Dolman’s talks are entitled: Controversies and Research in Endonasal Dacryocystorhinostomy and Evaluation and Management of Thyroid Orbitopathy.

Please enjoy today’s presentations.

Research Committee
Department of Ophthalmology and Visual Sciences

Acknowledgements

The Department of Ophthalmology and Visual Sciences would like to thank the following individuals for serving as judges for the 28th Annual Research Day:

**Dr. Peter Dolman**  
Department of Ophthalmology and Visual Sciences  
University of British Columbia  
Vancouver, BC

**Dr. Jayme Vianna**  
Department of Ophthalmology and Visual Sciences  
Dalhousie University  
Halifax, NS

**Dr. Curtis Archibald**  
Department of Ophthalmology and Visual Sciences  
Dalhousie University  
Halifax, NS
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@dalophtho
Dr. Peter Dolman, M.D.

Peter Dolman is a clinical professor at the University of British Columbia (UBC) in Vancouver, Canada and is on active staff at five hospitals. He is the director of ophthalmology fellowship programs at UBC, division head of oculoplastics and orbit, and a past president of the BC Society of Eye Physicians and the Canadian Society of Oculoplastics and Reconstructive Surgeons. He is the past president of the International Thyroid Eye Disease Society and a member of the Orbit Society, an international consortium of orbit experts. He has supervised over 46 international oculoplastics fellows, has delivered over 230 invited lectures, and published 20 chapters and over 75 journal articles. He recently co-edited a textbook on diseases of the orbit and ocular adnexae which was published in January this year. He has volunteered as a surgeon or lecturer in over 25 developing nations and has received several departmental research and teaching awards, the ASOPRS research award (2007) and the Queen Elizabeth Gold Medal for community service.
THE UTILITY OF A FZD4-/ ZEBRAFISH MODEL IN THE DEVELOPMENT OF TREATMENTS FOR FAMILIAL EXUDATIVE VITREORETINOPATHY

Gjerde, H.1; Prykhozhij, S.2; Ngo, M.3; McMaster, C.3; Berman, J.2; Robitaille, J.1

1Department of Ophthalmology and Visual Sciences, Dalhousie University, Halifax; 2Department of Pediatrics, Dalhousie University, Halifax; 3Department of Pharmacology, Dalhousie University, Halifax

Purpose: Familial exudative vitreoretinopathy (FEVR) is a rare, incurable genetic disorder characterized by the failure of peripheral retinal vascularization during development. Visual impairment occurs in approximately 40% of eyes (30% of patients), typically during early childhood. Approximately 50% of FEVR cases are due to a gene mutation disrupting the Norrin-FZD4 signaling pathway. We developed a fzd4-/- zebrafish model at Dalhousie University in the Berman Zebrafish Laboratory using TALEN technology, with the goal of using this model to efficiently screen compounds that ameliorate this vascular defect. A disorganized retinal vascular phenotype was only observed in fzd4-/- adult zebrafish but not in larvae, necessitating the laborious process of generating a flat mount of the retina for imaging. Thus, as an initial step, we needed to develop a novel approach to image adult retinas in situ to enable the throughput necessary for drug screening.

Methods: Fluorescence-based microscopy requires the removal of pigment to image the retinal vasculature. Pigmentation of the retinal pigment epithelium in zebrafish embryos initiates at 1 day post-fertilization (dpf); however, this can be inhibited for the first 5 dpf by applying phenylthiourea (PTU). To block ocular pigmentation later in development, we are introducing tyrosinase (tyr) mutations using CRISPR/Cas9 technology by injecting tyr-targeted single-guide RNAs (sgRNAs) into zygotic zebrafish with either Cas9 mRNA or protein at the one-cell stage. Retinal vasculature of the resultant mosaic tyr mutant- wildtype (WT) and fzd4-/- zebrafish will be imaged using a Zeiss Lightsheet Z.1 microscope, and analyzed at 14, 21, 28, and 35 dpf (10 fish/group/time point) using Arivis and ImageJ software. Vascular progression, vessel caliber, and total retinal vascular volume will be compared between time points and analyzed using descriptive statistics.

Results: We are currently optimizing CRISPR-based approaches to enhance survival in both WT and fzd4-/- zebrafish. Confocal microscopy reveals normal retinal vessel development in both the tyr mutant-WT and tyr mutant-fzd4-/- fish at 7 dpf.

Conclusion: We are developing unique retinal pigment-mutant zebrafish that will facilitate comparing and evaluating therapeutic responses and vascular phenotypes between WT and fzd4-/- mutants. These fish will provide a valuable pre-clinical animal model for identifying new, prospective therapies for FEVR.
TRANSVERSE VENOUS SINUS STENOSIS ON MAGNETIC RESONANCE IMAGING IN PATIENTS WITH IDIOPATHIC INTRACRANIAL HYPERTENSION – A PILOT STUDY

Chan, W.1,2, Mishra, A.1,2, Green, L.2,3, Purdy, A.2,3, Maxner, C.1,2,3, Shankar, J.2,4

1Department of Ophthalmology and Visual Sciences, Nova Scotia Health Authority, Halifax; 2Dalhousie University, Halifax; 3Division of Neurology, Department of Medicine, Nova Scotia Health Authority, Halifax; 4Division of Neuroradiology, Department of Diagnostic Imaging, Nova Scotia Health Authority, Halifax.

Purpose: Idiopathic intracranial hypertension (IIH) is a condition in which increased pressure within the head compresses the brain and can result in irreversible vision loss and chronic severe headaches. IIH mainly affects overweight women of child-bearing age, many of whom are asymptomatic until they present with vision loss. Ocular examination often reveals optic disc swelling. Years after initial presentation, visual acuity continues to be affected in 20% of IIH patients, even following treatment, while 50% of patients have a visual field abnormality at an average of 18 weeks after instituting treatment.

There is currently no known cause for IIH; however, it has been noted that 90% of patients with IIH have transverse venous sinus stenosis (TVSS) seen on magnetic resonance imaging (MRI) and venography (MRV) of the brain. Whether the venous stenosis has a causal relationship to IIH or is an effect of the increased intracranial pressures is controversial. This study aims to examine the feasibility of prospectively observing transverse venous sinus stenosis in patients with IIH from diagnosis, through treatment, and after treatment.

Methods: Patients diagnosed with IIH according to the Modified Dandy Criteria with evidence of TVSS on their MRI/MRV are recruited to the study. As part of standard of care, all IIH patients receive a MRI/MRV as well as a lumbar puncture as part of their diagnosis for IIH. To follow TVSS over time, participants undergo additional MRV immediately following lumbar puncture, 3-6 months after diagnosis when there has been resolution of IIH symptoms, and one year after diagnosis. Ophthalmological data, such as visual acuity, colour vision testing, documentation of papilledema with disc photographs, optic coherence tomography OCT of the optic discs and maculae, and Goldmann visual fields, are also collected at these follow-up time points. Feasibility data including patient recruitment barriers and logistical issues will also be recorded for protocol optimization.

Results: Since the beginning of patient recruitment in September 2016, 11 suspected IIH patients were screened by either ophthalmology or neurology. Of these 11 patients, 2 were successfully enrolled, 7 did not meet study eligibility criteria, one declined participation, and one withdrew from the study. We have optimized the logistical aspects of our research protocol and have bolstered recruitment with frequent communication with the departments of ophthalmology, neurology, and radiology.

Conclusions: With optimized recruitment and study pathway, we continue to enrol IIH patients in our ongoing prospective study.
THE TEMPORAL RAPHE OF THE RETINAL NERVE FIBRE LAYER AND GANGLION CELL LAYER IN GLAUCOMA PATIENTS WITH A HORIZONTAL HEMI-FIELD VISUAL FIELD DEFECT

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1 Ophthalmology and Visual Sciences, Dalhousie University, Halifax, NS, Canada.

Purpose: To determine whether the angle between the fovea and the temporal raphe (temporal raphe angle, TRA) respected the angle of a clear nasal step visual field (VF) defect.

Methods: Open-angle glaucoma patients followed in a prospective study who had all 5 locations immediately above or below the horizontal midline with a pattern standard deviation value of P < 0.5% in at least the last 3 consecutive VF (24-2, Humphrey Field Analyzer) were recruited. Two optical coherence tomography macular scans were performed; a 30°x15° comprising 398 B-scans aligned to horizontal image frame, and a 30°x25° comprising 61 B-scans aligned to the fovea to Bruch’s membrane opening (FoBMO) angle. A customized VF with 7 stimuli separated by 1° vertically at each of 5 horizontal eccentricities (6° apart) nasal to the fovea was performed. The TRA of the RNFL reflectance was measured. The ganglion cell layer angle (GCLA) was measured with the best fit line of the highest difference in GCL thickness and the FoBMO axis centred on fovea. The VF defect angle was measured with a best fit line with the highest difference in sensitivity at each horizontal eccentricity and the fovea.

Results: We included 12 (7 right and 5 left) eyes of 9 patients with median age of 67 (range: 63 to 74) years. There were 9 patients with an inferior hemi-field VF defect. The FoBMO angle was always negative with a median of -7.45° (-9.9° to -1.4°) while the median TRA, relative to horizontal, was +2.98° (-1.76° to +11.59°). The GCLA did not correspond to the FoBMO axis (median difference, +2.24° (range, 0.09° to 6.31°). The correlation between the TRA and VF angles (rho= 0.62, p= 0.04) was higher than that between the TRA and FoBMO angle (rho = -0.17, p = 0.59).

Conclusions: Glaucomatous hemifield scotomas do not respect the horizontal midline. Instead, the VF angle correlated with the orientation of the RNFL temporal raphe which itself is variable among patients and does not respect the horizontal midline. Additionally, the highest vertical asymmetry of GCL thickness did not follow the FoBMO axis demonstrating an additional aspect of inter-individual anatomical variation.
DALHOUSIE UNDERGRADUATE OPHTHALMOLOGY EDUCATION ASSESSMENT: FOCUS GROUPS AND TARGETED NEEDS ASSESSMENT

Best, W. 1,2, Belliveau, D. 1,2

1Department of Ophthalmology and Visual Sciences, Nova Scotia Health Authority, Halifax 2Dalhousie University, Halifax

Purpose: The past several decades have seen steady erosion of ophthalmology content in the mainstream medical school curriculum resulting in graduates who are underprepared to evaluate and manage ophthalmic complaints. At Dalhousie, the ophthalmology curriculum is constantly evolving to meet this perceived deficit. However, a comprehensive assessment has not been undertaken to evaluate the overall strengths of weaknesses of the curriculum. The purpose of this study is to gather the opinions of current medical students at Dalhousie regarding their undergraduate ophthalmology education in order to better understand their needs and help guide future curriculum changes.

Method: Three individual focus groups consisting of current Dalhousie medical students in years two, three and four were conducted. Two individuals reviewed the transcripts of these sessions using content analysis to identify major and minor themes. Results were subsequently incorporated into a targeted needs assessment to be distributed to the entire medical student population to gather a more comprehensive overview.

Results: Content analysis revealed major themes in currently employed curriculum, acquisition of ophthalmic knowledge and skills, as well as preferences for curriculum delivery. These themes were incorporated into a targeted needs assessment.

Conclusion: The current curriculum at Dalhousie is well received while still leaving room for improvement. Further exploration will be achieved with a targeted needs assessment.
PHOTOSWITCH PROTEINS AS A CHEMICAL VISUAL PROSTHESIS IN A MODEL OF ACQUIRED RETINAL DEGENERATION

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Purpose: DENAQ, BENAQ, and AAQ are a family of novel membrane photoswitch protein which target retinal ganglion cells to provide light-induced depolarization. In this way, they show promise as a chemical visual prosthesis to recreate visual function where photoreceptors are damaged beyond repair but the inner retina is still intact. In this study, we aim to determine the efficacy of these photoswitch proteins in restoring photosensitivity and vision in a rat model of acquired retinal degeneration.

Methods: Retinal damage is induced by the light induced retinopathy (LIR) model, by exposing Brown-Norway rats to 4-6 hours of intense fluorescent light, which produces progressive photoreceptor degeneration over the weeks that follow. At two weeks post-light exposure, one rat will be sacrificed and its retina used for histology to assess the light-induced retinal degeneration. The rest of the rats will undergo an intravitreal injection of the photoswitch protein in vehicle in one eye. For the other eye, half of the rats will receive a sham injection with vehicle only, and the other half will receive no injection. After one day of recovery post-injection, the rats will undergo electroretinogram (ERG) recording in each eye individually, followed by recording of visual evoked potentials (VEP). Following VEP, the rats will be sacrificed and the retinas of both eyes extracted and used for multi-electrode array (MEA) analysis. The isolated retinas used in the MEA will allow comparison of treatments with different concentrations of DENAQ, BENAQ, and AAQ tested following initial assessment of individual ganglion cell responses to light stimulation prior to drug application.

Results: Currently four rats have undergone the light exposure and will act as a pilot for the remainder of the studies to come. Initial results with MEA demonstrate that the technique is viable in rat retina and provides a reliable platform for testing visual restoration provided by these photoswitch chemicals. Further data demonstrating light damage to retina as well as the effect of the photoswitch chemicals is pending.

Conclusion: DENAQ is a promising new chemical photoswitch which may grant some vision back to severely damage retina. We demonstrate preliminary results that the LIR produces a phenotype of acquired retinal degeneration and that MEA testing is a reliable means with which to measure retinal activity in experimental rats.
Purpose: Retinal ganglion cell (RGC) loss leads to irreversible blindness and is often associated with elevated intraocular pressure (IOP). Studies of experimentally elevated IOP have identified axonal transport (AT) disruption as a contributing factor to RGC death. However, the capacity of AT to recover following a transient disruption remains unclear. This research examined AT function over time following a transient elevation of IOP causing ischemia.

Methods: Retrograde and anterograde AT in Brown Norway rats was monitored by injections of cholera-toxin β-subunit (CTB) Alexa488 conjugate into the superior colliculus or vitreous, respectively. IOP was unilaterally elevated to 120 mmHg by cannulation of the anterior chamber and infusion of saline, and the fellow eye was used as a control. IOP was maintained for 90 minutes, an injury known to cause progressive RGC loss, or 30 minutes, causing no RGC loss. Rats were sacrificed after 3, 6 and 24 hours for anterograde analysis, and 3, 7 and 14 days for retrograde analysis. AT in the optic nerve (ON) was analyzed by measuring average CTB fluorescence along the length of the nerve from longitudinal sections. Immunohistochemistry for Brn3a and phosphorylated neurofilament was used to assess RGC loss and axonal integrity, respectively.

Results: Thirty minutes of elevated IOP had no significant effect on anterograde AT, while retrograde AT of CTB was mildly reduced in experimental ONs (Δ fluorescence (mean arbitrary units (AU) ± SEM) = -9.16 ± 16.41 (-12%), N=11, p<0.05), with no significant differences between the time-points examined. Ninety minutes of elevated IOP caused a significant and progressive disruption of anterograde AT of CTB in experimental ONs (Δ fluorescence (mean AU ± SEM) per time-point: 3h = -28.09 ± 15.60 (-43%), 6h = -31.28 ± 8.49 (-44%), 24h = -42.37 ± 13.50 (-49%); N=8/ group, all p<0.001). Retrograde AT of CTB was also significantly disrupted following 90 minutes of elevated (Δ fluorescence (mean AU ± SEM) = -18.13 ± 7.22 (-27%), N=12, p<0.01), again with no significant difference between time-points. Significant loss of Brn3a+ nuclei and aberrant phosphorylated neurofilament labelling in RGC somas was observed following the 90-minute insult, whereas this pathology was absent following the 30-minute insult.

Conclusions: These findings suggest that, rather than exhibiting reversible AT dysfunction following a transient elevation of IOP causing ischemia, RGCs may succumb to high IOP-induced stress in a cell-by-cell manner that is acute and permanent, the degree of which depends on the duration of IOP elevation.
NON-RETINAL NERVE FIBRE LAYERS WITHIN THE OPTIC NERVE HEAD NEURORETINAL RIM

Torres, L.¹; Vianna, J.¹; Jarrar, F.¹; Butty, Z.¹; Sharpe, G.¹; Hutchison, D.¹; Nicolela, M.¹; Chauhan, B.¹

¹Department of Ophthalmology and Visual Sciences, Dalhousie University, Halifax, Nova Scotia, Canada.

Purpose: The presence of retinal layers other than retinal nerve fibre layer (RNFL) within the optic nerve head (ONH) minimum rim width (MRW) was previously described in monkeys (Fortune B et al., IOVS 2016). We aimed to determine the occurrence and relevance of these layers within the MRW, herein referred to as “protruded layers” (PL), in normal human subjects.

Methods: 24 radial ONH B-scans, centred on the Bruch’s membrane opening and aligned to the fovea, were acquired from 30 healthy subjects with optical coherence tomography (Spectralis). One randomly selected eye per subject was analyzed. In each of the 2 positions for each B-scan, the MRW was examined for the presence of PL, which was manually measured when detected. The proportion of PL comprising the MRW was calculated. To evaluate the reproducibility of PL detection and measurement, 2 observers (Observers 1 and 2) analyzed a subsample of 240 scans from 15 subjects.

Results: The median age of the subjects was 69 years (interquartile range [IQR]: 63.2 – 76.7 years). Of the 240 B-scans evaluated for reproducibility, Observer 1 detected PL in 36 (15%) positions and Observer 2 in 38 (16%) positions. The observers agreed about the presence of PL in 31 of 43 positions (72%) and about the PL’s absence in 188 of 208 positions (90%). The median PL thickness difference among the observers was 4.0 µm (IQR: -3.0 µm – 16.5 µm). Of all the 720 B-scans analyzed, PL was detected in 199 (14%) of all 1440 positions. Each eye had a median of 8 positions out of 48 (17%) with PL (IQR: 3.5 [7%] – 14.5 [30%]). 23 (77%) of the subjects had at least one position with PL. The temporal half of the ONH contained 183 (91%) of all 199 positions with PL. When present, the median PL thickness was 65 µm (IQR: 48 µm – 86.5 µm), making up a median of 28% (IQR: 18% - 38%) of the MRW.

Conclusion: The frequent presence of PL in the ONH radial scans, mainly in the temporal quadrant, and the variable proportion of PL that constitutes the neuroretinal rim, may impact diagnostic accuracy in single examinations. However, we expect the presence of PL to have minimal impact in detecting change over serial examinations.
CANNABINOID 2 RECEPTOR (CB2R), A NOVEL THERAPEUTIC TARGET IN THE TREATMENT OF OCULAR PAIN AND INFLAMMATION

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Purpose: Damage to corneal tissues resulting from chemical burn, surgeries, trauma, neurological diseases, infections etc., produce ocular pain as well as inflammation. Existing pharmacotherapies for ocular pain and inflammation including topical corticosteroids, tricyclic antidepressants, benzodiazepines, GABAergic drugs and opioids are frequently failed to provide adequate pain relief and are also associated with severe side-effects (corneal thinning and decreased wound healing). Activation of the endocannabinoid system (ECS), which includes cannabinoid 1 (CB1R) and cannabinoid 2 (CB2R) receptors, is analgesic and anti-inflammatory, respectively. However, CB1R activation, in spite of their analgesic effects, has potential to produce behavioral psychoactive side-effects. Therefore, the purpose of this research is to investigate the antinociceptive and anti-inflammatory effects of CB2R activation using non-psychoactive phytocannabinoid, CBD, and its derivative-a CB2R agonist, HU308, in a mouse model of corneal hyperalgesia and inflammation.

Methods: Experimental corneal hyperalgesia and inflammation were generated using chemical cauterization of the cornea in wildtype (WT) and CB2R knockout (CB2R−/−) mice. Cauterized eyes were treated with topical cannabinoids (1-5% w/v) in the presence/absence of the CB1R antagonist AM281 (2.5mg/kg ip) at: 30, 60 and 120 min (M) post-injury. The ocular pain response was quantified from 30 seconds video recordings of the behavioral pain responses (blink response, squints and eye wipes) recorded at 6 hours post-injury using capsaicin-stimulation. Eyes were enucleated at 12 hours post-injury and corneal inflammation was analyzed using neutrophil counts.

Results: Application of 5% CBD and 1.5% HU308 reduced ocular pain in WT mice compared to vehicle-treated eyes (p < 0.01). The antinociceptive effects of CBD and HU308 were reduced in CB2R−/− mice (p < 0.05 and p > 0.05, respectively) whereas these effects were remained unaffected in mice treated with the CB1R antagonist AM281 (p < 0.0001). Neutrophil infiltration to cauterized cornea was increased 12 hours post injury, compared to non-cauterized eyes in WT mice (p < 0.0001). 5% CBD and 1.5% HU308 reduced neutrophil infiltration (p <0.001 & p <0.0001, respectively); these effects were reduced in CB2R−/− mice (p < 0.01 & p > 0.05, respectively).

Conclusion: The anti-nociceptive and anti-inflammatory actions of CBD and HU308 are independent of CB1R and mediated through CB2R activation. Cannabinoids activating CB2R could offer a novel therapy for ocular pain and inflammation.
COMPARISON OF OUTCOMES OF TRABECULECTOMY WITH SUBCONJUNCTIVAL INJECTION OF MITOMYCIN C VERSUS TOPICAL APPLICATION WITH CELLULOSE SPONGE

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Purpose: To present results of a retrospective case series of surgical outcomes in trabeculectomy comparing sponge application of Mitomycin C (MMC) with an intraoperative subconjunctival injection method of 10 or 20 micrograms of MMC.

Study design: Retrospective case series and chart review

Methods: Charts of consecutive patients from on surgeon’s practice (LS) that received trabeculectomy with mitomycin C by sponge application or subconjunctival injection at the Nova Scotia Eye Care Center between 2010 and 2016 at the Nova Scotia Eye Care Center satisfying inclusion and exclusion criteria were reviewed. Inclusion criteria were open or closed angle glaucoma and history of trabeculectomy or phaco-trabeculectomy. Exclusion criteria were age less than 40, less than 3 months follow up post surgery, history of previous intraocular surgery other than uncomplicated cataract surgery (phaco/PCIOL) and inflammatory or neovascular glaucoma. Primary outcome measures were (1) complete success - number of patient with IOP less than 21 mmHg of >=30% reduction from the pre-operative IOP without glaucoma medications, and (2) qualified success - number of patients who achieved the goals of complete success with addition of glaucoma medications. Secondary outcomes included number of glaucoma medications, intra- and post-operative complications, other post-operative procedures and visual acuity. Results from three groups ((1) MMC sponge, (2) 10 micrograms subconjunctival MMC, (3) 20 micrograms subconjunctival MMC) will be presented with a non-inferiority analysis.

Results and Conclusions: Preliminary results demonstrate cumulative success (defined as complete and qualified success) of 67.5% in the sponge application method and 80% in the 20 microgram injection method (N=62, p = 0.032) at 3 months post-operatively. Additional post-operative data, and the outcomes using 10 microgram injection will also be presented and discussed. Intra-operative and post-operative complications rates were similarly low across groups.
EXACERBATION OF GABAC RECEPTORS BY ANTIPELPTIC VIGABATRIN IN THE RETINA: AN EXPLORATORY STUDY

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1Dalhousie University, Halifax; 2Department of Ophthalmology and Visual Sciences, Nova Scotia Health Authority, Halifax

Purpose: Vigabatrin (VGB; Sabril®) is an irreversible GABA-Transaminase inhibitor used in the control of pharmacoresistant epilepsy and in the treatment of infantile spasms. While very potent at the cortical level, it is associated with retinal toxicity on long-term use. We hypothesized that the toxicity is due to the high concentration of GABAc receptors found in the retina, those receptors being tenfold more sensitive and having slower activation/inactivation kinetics as well as lower desensitization than other GABA receptors.

Methods: Guinea pigs are gavaged with VGB (2ml, 250mg/kg) daily for either 5-7 days (VGB-S) or 90 days (VGB-L), after which electroretinograms (ERGs) were recorded under ketamine/xylazine anesthesia. The retina was then isolated and put on a multielectrode array where the retinal ganglion cell spiking activity was recorded. GABAA antagonist Gabazine (200µM) and GABAc antagonist TPMPA (150 µM) were used to challenge the retina.

Results: Short-term treatment with VGB affected the ERG signal (n=15 control,13 VGB-s), increasing the amplitude of a- and b-waves, while selectively reducing the late Oscillatory Potentials (OPS). Low concentration of GABA in the vitreous of control retina produced a similar effect, while a high concentration made the b-wave disappear and the whole signal electronegative. The RGC responses were also affected by VGB, showing a decrease in the primary flash responses and an increase in the OFF component of the long-duration stimulus (n=5controls 330cells, 6 VGB-s 396cells). GABA at low concentration increased the ON and OFF responses in normal retinae and abolished the photic responses at high concentration (n=4 264 cells), somewhat in parallel with what was observed with the ERGs. The main effect of Gabazine was a reduction in the ERG oscillatory potentials and increased spike counts for RGCs in both groups while TPMPA had little effect on OPs but made the b-wave disappear and increased the toxicity of RGCs.

Conclusion: Much remains to be done to understand the mechanisms by which VGB is affecting the retinal function. But it can already be said that, like in human observations, there is a short-term effect that may be associated with the long-term development of a toxic effect.
CLINICAL UTILITY OF OCT ANGIOGRAPHY IN THE DETECTION OF SUBRETINAL NEOVASCULARIZATION IN AGE-RELATED MACULAR DEGENERATION

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1Dalhousie University, Halifax; 2Department of Ophthalmology and Visual Sciences, Nova Scotia Health Authority, Halifax

Purpose: To determine the sensitivity and specificity of OCT-A in the detection of subretinal neovascularization in patients with age related macular degeneration.

Methods: Retrospective review of 30 consecutive patients diagnosed with exudative ARMD, with subretinal neovascularization confirmed by intravenous fluorescein angiography (IVF), the current gold standard. OCT-A performed on the initial visit, was interpreted by an ophthalmologist blind to all other patient parameters including imaging. The sensitivity was determined by the proportion of true positives: proportion of patients with subretinal neovascularization detected by OCT-A out of the total number of patients with exudative AMD. OCT-A of 30 eyes of patients with AMD and no active subretinal neovascularization as determined by IVF were reviewed by a blind observer. The specificity was determined by the proportion of true negatives: proportion of patients with no subretinal neovascularization detected by OCT-A out of the total number of patients with no evidence of subretinal neovascularization of IVF.

Results: TBD

Conclusion: TBD
PILOT STUDY INTO THE UTILITY OF FLUORESCIN ANGIOGRAPHY-BASED COMPUTER ALGORITHM ASSESSMENT OF RETINAL VASCULAR HEALTH IN DIABETIC RETINOPATHY

Winter, A.1,2, Cruess, A.1,2, Murthy, S., Vema, K.3, Friedman, A.3

1Dept of Ophthalmology and Visual Sciences, Nova Scotia Health Authority, Halifax; 2Dalhousie University, Halifax; 3Department of Medical Neuroscience, Dalhousie University, Halifax, Nova Scotia, Canada

Purpose: Pilot study to assess and help validate a novel fluorescein angiography (FA) based computer algorithm to quantitatively assess retinal vessel permeability and blood flow in patients with diabetic retinopathy.

Methods: 100 patients with a clinical diagnosis of non-proliferative diabetic retinopathy (NPDR) undergoing clinically indicated FA will be assessed and divided into two groups: a. Standard FA for NPDR complicated by diabetic macular edema (DME) (n=50) and b. Ultra wide field OPTOS FA for assessment of peripheral diabetic retinopathy in patients without clinically significant DME (n=50). Image pre-processing includes removal of non-retinal and noisy images and registration to achieve spatial and temporal pixel-based analysis. Retinal vessel permeability and blood flow are assessed for each pixel by computing intensity kinetics normalized to arterial values. A linear curve is fitted and a slope value assigned, color-coded and displayed. Standard FA studies and the computed permeability and blood flow maps will be interpreted in a masked and randomized manner by three experienced ophthalmologists for statistical validation of diagnosis accuracy and efficacy. Intra and inter-observer variability will be measured using Pearson correlation coefficients. FA-based macular permeability maps will be correlated with OCT macular thickness maps.

Results: Preliminary results of test pilot patients will be presented.

Conclusions: Quantitative analysis of FA using a computer algorithm may represent a faster, objective, sensitive and more accurate methodology to assess retinal perfusion and vessel permeability compared to the current subjective physician diagnosis of FA-based retinal pathology. Computer algorithm-based FA analysis may provide an earlier and more reliable detection of diabetic retinopathy and more precise disease classification and staging allowing better follow-up of disease progression and response to treatment. Permeability maps may facilitate more targeted therapy limiting the overall dose of laser photocoagulation thereby reducing the degree of chorioretinal scarring and concomitant visual field loss.
CHARACTERIZING LONGITUDINAL IN VIVO CHANGES OF RGC DENDRITES AFTER RETINAL INJURY

Henderson, D.², Gobran, J.², Hooper, M.¹, Farrell, S.¹, Chauhan, B.²,¹

¹Ophthalmology and Visual Sciences, Nova Scotia Health Authority, Halifax, NS, Canada; ²Physiology and Biophysics, Dalhousie University, Halifax, NS, Canada

Purpose: Retinal ganglion cell (RGC) death is preceded by retraction of dendrites and an overall loss of dendritic branch complexity; however, longitudinal characterization of these changes is lacking. The aim of the present study was to examine structural changes of RGC dendrites, longitudinally, to characterize the effects of acute and chronic retinal injury on RGC dendritic arbours.

Methods: Animals (Thy-1 YFP, line H; Jackson Laboratories, ME) had either optic nerve transection (ONT; n=9) or magnetic microbead injection (MMI; n=2) in one eye. MMI caused sustained elevations in intraocular pressure (IOP) compared to the fellow, uninjected, eye for the duration of the experiment. Confocal scanning laser ophthalmoscopy (CSLO; Spectralis, Heidelberg Engineering) was used to perform longitudinal in vivo imaging of YFP+ RGCs through the progression of retinal injury. Sholl analysis was conducted on in vivo CSLO images to quantify changes in dendritic complexity and arbour size of YFP+ RGCs over the time course of both injuries.

Results: ONT causes a rapid and severe loss of RGCs over three weeks. Early time points were examined to characterize RGC dendritic arbours prior to cell death. Dendritic arbour radii retracted (mean (SD): 179 (33) vs. 160 (49) μm; baseline vs. Day 11, n=20 cells) and the peak number of branch intersections was decreased (22 (5) vs. 10 (4)) after ONT. Also, 4 of the 20 RGCs followed died prior to day 11. Ocular MMI causes sustained IOP elevation, representing a chronic retinal injury over several weeks. Like ONT, RGC arbour radii were reduced (173 (18) vs. 165 (22) μm; baseline vs. Day 28, n=6 cells) and peak number of intersections was smaller (26 (4) vs. 19 (4)) after prolonged IOP elevation. The area under the curve (AUC) of RGC Sholl profiles, a measure of dendritic complexity, decreased following both retinal injuries; however, to a lesser degree in the MMI group (64% vs. 26%, 11 days post-ONT vs. 28 days post-MMI).

Conclusion: The present study demonstrates that longitudinal in vivo imaging and Sholl analysis can be used to track the progression of RGC injury prior to cell death. Also, the MMI model, caused a prolonged elevation of IOP, resulted in a slower time course of dendritic retraction and loss of RGC arbour complexity compared to ONT. These results suggest that chronic, but moderate IOP elevation is a good model to study cellular changes in RGCs preceding cell death.
INVESTIGATING NOVEL SELECTIVE CANNABINOID 2 RECEPTOR AGONISTS AS POTENTIAL THERAPEUTIC DRUGS FOR THE TREATMENT OF OCULAR INFLAMMATION

Porter, R.¹, Szczesniak, A.¹, Toguri, T.¹, Gebremeskel, S.², Johnston, B.², Lehmann, C.¹², Grether, U.⁴, Ullmer, C.⁴, Kelly, M.¹³

Departments of Pharmacology¹, Microbiology & Immunology², Ophthalmology & Visual Sciences³, Dalhousie University, Halifax, NS, Canada. Roche Innovation Center Basel⁴, F. Hoffman-La Roche Ltd., 4070 Basel, Switzerland

Purpose: Uveitis is a heterogeneous group of ocular inflammatory diseases. Mainstay drugs used to treat uveitis, such as steroids, have many adverse effects. Therefore, development of new non-steroidal drug targets is desirable. Activation of the cannabinoid 2 receptor (CB₂R) can decrease ocular inflammation. Therefore, drugs selectively targeting CB₂R could represent novel therapeutics for uveitis. The objective of this study was to examine the anti-inflammatory actions of RO6871304 and RO6871085, highly potent and selective CB₂R agonists originating from two chemically diverse series, and RO6851228, a novel CB₂R inverse agonist, in a model of experimental endotoxin-induced uveitis (EIU).

Methods: EIU was induced in mice by intravitreal injection of lipopolysaccharide (LPS). Real-time intravital microscopy was used to visualize and quantify leukocyte-endothelial interactions in the iridial microvasculature as a measure of inflammation. An in vitro Boyden chamber bioassay was used to determine whether the novel CB₂R agonists modulated neutrophil migration. To further examine the immune cell subtype targeted by these novel CB₂R selective agonists, neutrophils were depleted prior to induction of EIU. Leukocytes were adoptively transferred 5 hr post EIU.

Results: Topical treatment with the CB₂R agonists, RO6871304 (Ki hCB₂R 8 nM; Ki hCB1R / Ki hCB₂R >1250) and RO6871085 (Ki hCB₂R 25 nM; Ki hCB1R / Ki hCB₂R 122) (1.5% w/v), significantly decreased LPS-induced leukocyte-endothelial adhesion compared to vehicle. Conversely, treatment with the CB₂R inverse agonist, RO6851228 – a close analogue of RO6871085 (Ki hCB₂R 26 nM; Ki hCB1R / Ki hCB₂R 250) - increased LPS-induced leukocyte-endothelial adhesion. Consistent with in vivo inhibition of leukocyte adhesion, RO6871304 (0.01 μm) significantly decreased neutrophil migration in vitro compared to vehicle. Topical treatment with RO6871304 in neutrophil-depleted mice significantly decreased the LPS-induced adhesion of adoptively-transferred leukocytes compared to vehicle.

Conclusion: Treatment with the novel CB₂R agonists, RO6871304 and RO6871085, was associated with decreased inflammation in EIU and reduced neutrophil migration in vitro (RO6871304). Conversely, the CB₂R inverse agonist, RO6851228, increased inflammation. Adoptive leukocyte transfer experiments suggest that, in addition to neutrophils, CB₂R agonists may also exert their anti-inflammatory actions through resident immune cells, such as microglia or macrophages. Taken together, these data demonstrate an anti-inflammatory role for CB₂R in the eye and suggest that drugs targeting the endocannabinoid system maybe a therapeutic target for ocular inflammation.
THE EFFECT OF INTRAVITREAL BEVACIZUMAB IN EXPERIMENTAL MODELS OF OCULAR INFLAMMATION

Seamone, M.,1 Toguri, T.,2 Lafreniere, D.,2 Gupta, R.,1 Cruess, A.,1 Kelly, M.2

1Department of Ophthalmology & Visual Sciences, Dalhousie University

Purpose: VEGF-A contributes to ocular inflammation by increasing vascular permeability and by upregulating cellular adhesion molecules. Levels of VEGF-A are increased in Exogenous Endophthalmitis in humans. Experimental Endotoxin-Induced Uveitis (EIU) and Peptidoglycan Induced Uveitis (PIU) mimic gram-negative and gram-positive ocular infection respectively. The purpose of this manuscript is to determine if intravitreal (IVT) Bevacizumab decreases inflammation in EIU and PIU.

Materials and Methods: Six groups of BALB/c mice (n= 8-10 per group) were studied: wild-type (WT) control (saline, IVT), wild-type vehicle (VEH, α – trehalose dehydrate, polysorbate 20, sodium phosphate) WT EIU (LPS, 250 ng, IVT), WT EIU + Bevacizumab (BEV, 2.5 µg, 25 µg and 45 µg IVT), WT PIU (2 µg PGN) and WT PIU + BEV (2.5 µg, 25 µg and 45 µg). Intravitreal injection of vehicle did not result in a significant change in leukocyte-endothelial adhesion (p>0.05). Intravitreal injection of Bevacizumab did not decrease leukocyte-endothelial interactions. However, administration of intravitreal Bevacizumab (2.5 µg) in EIU resulted in a significant decrease in leukocyte adhesion to microvasculature at 6 hours (p<0.05)

Results: A significant increase in leukocyte adhesion to iris microvasculature was observed 6 hours following induction of EIU and PIU (p<0.001). Intravitreal injection of vehicle did not result in a significant change in leukocyte-endothelial adhesion (p>0.05). In PIU, intravitreal injection of Bevacizumab did not decrease leukocyte-endothelial interactions. However, administration of intravitreal Bevacizumab (2.5 µg) in EIU resulted in a significant decrease in leukocyte adhesion to microvasculature at 6 hours (p<0.05)

Conclusions: Bevacizumab significantly decreased leukocyte adherence to iris microvasculature in BALB/c mice in EIU but not PIU, i.e. the anti-inflammatory effects of Bevacizumab were observed only in the setting of simulated gram-negative inflammation. These results are of significance as Gram-negative endophthalmitis is associated with severe ocular inflammation and worse visual outcomes than Gram-positive endophthalmitis. These results provide evidence for VEGF-A blockade as adjunct therapy to intravitreal antibiotics (and vitrectomy) in exogenous endophthalmitis.
DR. R. EVATT AND RITA MATHERS TRAINEE SCHOLARSHIPS

The Department of Ophthalmology and Visual Sciences is pleased to announce the Dr. R. Evatt and Rita Mathers Trainee Scholarships in Ophthalmology and Visual Sciences.

Five separate funding opportunities:

- Dr. R. Evatt and Rita Mathers Research Fellowship in Ophthalmology and Visual Sciences
- Dr. R. Evatt and Rita Mathers Graduate Scholarship: PhD, Vision Sciences, Faculty of Medicine
- Dr. R. Evatt and Rita Mathers Graduate Scholarship: MSc, Vision Sciences, Faculty of Medicine
- Dr. R. Evatt and Rita Mathers Graduate Scholarship: MSc, Clinical Vision Science, Faculty of Health Professions
- Dr. R. Evatt and Rita Mathers Graduate Scholarship: Concurrent MSc and Ophthalmology Residency Program (Tuition Recovery Scholarship)

Applications must be received by May 1, 2017

For more information please visit medicine.dal.ca/ophthalmology
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<tr>
<th>Year</th>
<th>Recipients</th>
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<tr>
<td>1989/90</td>
<td>Dr. Denis Falvey; Dr. Andrew Orr</td>
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<tr>
<td>1990/91</td>
<td>Dr. Robin Cottle; Dr. Inge DeBecker; Dr. Robert Scott</td>
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<tr>
<td>1991/92</td>
<td>Dr. Simon Lam; Dr. Robert Scott; Dr. Rajender Mohandas</td>
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<td>1992/93</td>
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<td>1995/96</td>
<td>Dr. Robert Scott; Dr. Michael Altaweel</td>
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<td>Dr. Lesya Shuba; Dr. Lica Chui</td>
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<td>Dr. Jeff Steeves; Dr. Lesya Shuba</td>
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<td>Dr. Alex Tan; Dr. Darren Behn</td>
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<td>Dr. Curtis Archibald; Dr. Stephanie Dotchin</td>
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<td>2008/09</td>
<td>Dr. Nigel Rawlings; Dr. Ken Roberts</td>
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<td>Dr. Jeremy Murphy; Dr. Xu Wang</td>
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<td>2009/10</td>
<td>Dr. Ken Roberts; Dr. Sohail Safi</td>
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<td>Dr. Luis Pérez de Sevilla Müeller; Dr. Stuart Trenholm</td>
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<td>2010/11</td>
<td>Dr. Lisa Heckler; Dr. Ken Roberts</td>
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<td>Meggie Beazley; Dr. Stuart Trenholm; Michael Betts</td>
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<td>2011/12</td>
<td>Dr. Anuradha Mishra; Dr. Lisa Heckler</td>
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<td>Dr. Tony Redmond; Sally Dickinson</td>
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NOTES
What have you learned through attending Research Day?

Has Research Day stimulated any new ideas for you?

Will you make any changes to your clinical approach based on new information presented today?
Are there any changes you would suggest to improve the organization of future Research Days?

This program is free of commercial bias:

- Strongly agree
- Somewhat agree
- Equivocal
- Somewhat disagree
- Strongly disagree

This program enhanced my knowledge

- Strongly agree
- Somewhat agree
- Equivocal
- Somewhat disagree
- Strongly disagree

Please indicate which CanMEDS roles you felt were addressed during this educational activity

- Expert
- Communicator
- Collaborator
- Manager
- Health Advocate
- Scholar
- Professional

Please return to Leah Wood, Department of Ophthalmology & Visual Sciences, 1276 South Park St., 2W Victoria, Room 2026A, Halifax, NS B3H 2Y9
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