336 Blood flow
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Exhibit/Poster Hall Poster Session
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Program Number: 3029 **Poster Board Number:** A0024
Presentation Time: 11:00 AM–12:45 PM
Alteration of ocular blood pressure and flow leads to ophthalmic vascular and neuroretinal dysfunction
Shu-Huai Tsai[1], Wankun Xie[1, 2], Min Zhao[1, 2], Robert H. Rosa[1, 2], Travis W. Hein[2], Lih Kuo[1, 2]. [1]Department of Medical Physiology, Baylor Scott & White Health, Texas A&M University Health Science Center, Temple, TX; [2]Department of Ophthalmology, Baylor Scott & White Health, Texas A&M University Health Science Center, Temple, TX.
Purpose: Although systemic hypertension is a major risk factor for retinopathy, its impact on the ocular circulation and retinal function remains unclear. Transverse aortic coarctation (TAC) induces differential blood pressure/flow distribution to the left vs. right eye. Herein, utilizing the TAC model, we investigated the impact of ocular blood pressure/flow alteration on ophthalmic vasomotor regulation and retinal function.
Methods: TAC was performed in mice by banding the aorta between the brachiocephalic artery and the left common carotid artery (CCA). Sham-operated mice served as controls. Blood velocity in the central retinal artery (CRA) was measured by Doppler ultrasound. Ophthalmic arteries were isolated, cannulated, and pressurized for functional study. Retinal function and morphology were examined using electroretinography and optical coherence tomography, respectively.
Results: TAC for one month elicited left ventricular hypertrophy and dysfunction. Mean arterial pressure was increased by 50% and decreased by 30% in the right and left CCAs, respectively. Mean blood velocity was gradually increased by 40% and decreased by 20% in the right and left CRAs, respectively, at 4 weeks after aortic banding. Fundus imaging revealed no changes in the retinal vessel diameters in both eyes. The responses of right (hypertensive) ophthalmic arteries to the endothelium-dependent, nitric oxide-mediated vasodilators acetylcholine and adenosine were significantly reduced and the vasoconstriction to endothelin-1 was increased in TAC animals. In contrast, the responses of left ophthalmic (hypotensive) arteries to the above agonists were unchanged. Decreased retinal oscillatory potentials and increased implicit times in the scotopic electroretinogram were found in both right and left eyes. No significant changes in retinal morphology were observed in both eyes.
Conclusions: The elevation of blood pressure and flow to the ocular circulation for 4 weeks causes ophthalmic vascular dysfunction, which is associated with impaired retinal function without altering retinal morphology. The ophthalmic vasomotor function is insensitive to hypotensive insult; however, the insufficient blood flow might cause retinal injury possibly due to ischemia from exhausted vasodilator reserve. Our results provide further insight on the development of retinopathy in the early phase of ocular blood pressure and flow alterations.
Commercial Relationships: Shu-Huai Tsai, None; **Wankun Xie**, None; **Min Zhao**, None; **Robert H. Rosa**, None; **Travis W. Hein**, None; **Lih Kuo**, None
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Program Number: 3030 **Poster Board Number:** A0025
Presentation Time: 11:00 AM–12:45 PM
Thrombin causes biphasic regulation of vascular tone in porcine retinal arteries
Kengo Takahashi, Taiji Nagaoka, Tsuneaki Omae, Shinji Ono, Takayuki Kamiya, Akira Tanner, Akitoshi Yoshida. Ophthalmology, AsahikawaMedicalUniversity, Asahikawa, Japan.
Purpose: Thrombin, a serine protease, plays an important role in hemostasis by converting fibrinogen to fibrin. In the vasculature, thrombin had been reported to cause vasodilation, whereas a previous study reported that thrombin causes vasoconstriction in bovine retinal arteries. However, the effect of thrombin on the retinal microcirculation remains unclear. We examined the effects of thrombin on the retinal microvasculature and investigated the signaling mechanisms.
Methods: Porcine retinal arterioles were isolated, cannulated, and pressurized (55 cm H_2O) without flow for this in vitro study. Videomicroscopic techniques were used to record the changes in diameter in the retinal arterioles in response to thrombin at concentrations of 0.001 mU/ml to 20 mU/ml.
Results: Thrombin induced a concentration-dependent vascular response, i.e., initially vasoconstriction at a low concentration (<5 mU/mL) and then vasodilation at a high concentration (>5 mU/mL). Pretreatment with a protease-activated receptor (PAR)-1 inhibitor but not a PAR-2 or PAR-4 inhibitor significantly (p<0.01) suppressed thrombin-induced constriction. Endothelial denudation suppressed vasodilation caused by a high concentration of thrombin.
Conclusions: The current findings suggested that the low dose of thrombin causes vasoconstriction via a PAR-1 on smooth muscle, and the high dose of thrombin causes endothelium-dependent vasodilation in the retinal arterioles.
Commercial Relationships: Kengo Takahashi; **Taiji Nagaoka**, None; **Tsuneaki Omae**, None; **Shinji Ono**, None; **Takayuki Kamiya**, None; **Akira Tanner**, None; **Akitoshi Yoshida**, None

Program Number: 3031 **Poster Board Number:** A0026
Presentation Time: 11:00 AM–12:45 PM
Optical Coherence Tomography Angiography in preperimetric and glaucomatous eyes
Teresa Rolle, Laura Dallorto, Marco Tavassoli, Raffaele Nuzzi. Department of Surgical Sciences, University of Torino, Eye Clinic, Torino, Italy.
Purpose: The aim of the study was to evaluate optic nerve head (ONH), peripapillary and macular vasculature with Optical coherence tomography angiography (OCT-A) in preperimetric (PPG), primary open-angle glaucoma (POAG) and normal eyes, and to assess associations among the vasculature, structural and functional damage.
Methods: This was an observational, cross-sectional study. Preperimetric, glaucomatous and healthy subjects underwent a comprehensive ocular examination included RNFL, GCC thicknesses and whole image(wiVD), inside disc (idVD), peripapillary(ppVD) and macular (mVD) vessel densities imaging with RTVue-XR Avanti. Vessel densities were collected at two different levels: nerve head(NH) and radial peripapillary capillary (RPC) for the optic disc scan, superficial and deep plexus for the macular scan. Analysis of variance (ANOVA), post-hoc test and linear regression analysis were used.
Results: 14 healthy, 33 PPG and 26 POAG eyes were included. wiVD and idVD in POAG (51.31±4.95%, 47.30±6.59%) and PPG eyes (55.12±4.66%, 51.39±5.42%) were significantly decreased compared to healthy eyes (59.47±3.07, 56.01±5.15%, p<0.001, p=0.004 for wiVD, p<0.001, p=0.01 for idVD). ppVD demonstrated

a significant lower rate in POAG than healthy eyes in both NH and RCP segments (greatest reduction of 24.60% in inferotemporal region in RCP segment). PPG showed significant ppVD reduction of 7-11% in all sections of NH segment except for temporal sector and in RCP segment reduction of 8.84% and 8.29% in inferotemporal and superonasal sectors respectively. The mVDs were significantly lower in eyes with POAG vs normal eyes. Vessel density reduction was found in temporal, superior and inferior segments of parafoveal region of PPG eyes in superficial plexus. Regression analysis showed that the ONH vessel densities strongly associated with the RNFL thicknesses ($r=0.81$, $p<0.001$ for ppVD inferotemporal-RNFLinferior) and mVD of the deep plexus correlated with macular inner retinal layer thickness ($r=0.61$, $p<0.001$).
Conclusions: Vessel densities in POAG and PPG eyes are significantly lower than healthy eyes both in the optic nerve head and macular regions. A diminished microvascular network is associated with RNFL and macular inner retinal layer thicknesses reduction. OCT-A may provide further information about pathophysiology of the onset and progression of glaucomatous disease.
Commercial Relationships: Teresa Rolle, None; **Laura Dallorto**, None; **Marco Tavassoli**, None; **Raffaele Nuzzi**, None

Program Number: 3032 **Poster Board Number:** A0027
Presentation Time: 11:00 AM–12:45 PM
Ripasudil (K-115) elicits dilation of isolated porcine retinal arterioles
Takayuki Kamiya, Taiji Nagaoka, Motofumi Kawai, Tsuneaki Omae, Seigo Nakabayashi, Shinji Ono, Kengo Takahashi, Akira Tanner, Akitoshi Yoshida. Asahikawa Medical University, Asahikawa, Japan.
Purpose: Ripasudil (K-115), a Rho kinase inhibitor, has an intraocular pressure-lowering effect. In our previous study, we found that administration of intravitreal ripasudil increased the retinal blood flow in cats. However, the mechanism underlying the effect of ripasudil on the retinal microcirculation is unknown. Therefore, we investigated the direct effect of ripasudil on the retinal arterioles in pigs and the mechanism underlying the ripasudil-mediated effect.
Methods: The porcine retinal arterioles were isolated, cannulated, and pressurized at 55 cm H_2O without flow for the in vitro study. Diameter changes were recorded using videomicroscopic techniques.
Results: Ripasudil elicited dilation of the isolated retinal arterioles in a dose-dependent (100 pM-30 μM) manner within 3 minutes. Pretreatment with N^G-nitro-L- arginine methyl ester and denudation did not inhibit the effect of ripasudil on the retinal arterioles.
Conclusions: Ripasudil elicits endothelium-independent vasodilation of the retinal arterioles.
Commercial Relationships: Takayuki Kamiya; **Taiji Nagaoka**, None; **Motofumi Kawai**, None; **Tsuneaki Omae**, None; **Seigo Nakabayashi**, None; **Shinji Ono**, None; **Kengo Takahashi**, None; **Akira Tanner**, None; **Akitoshi Yoshida**, TOMEY (P), Topcon (P), Nidek (P)

Program Number: 3033 **Poster Board Number:** A0028
Presentation Time: 11:00 AM–12:45 PM
Retinal morphology and vessel oxygen saturation in normal and diabetic patients
Adam M. Dubis[1, 2], Luke Nicholson[2], Roxanne Crosby-Nwaobi[2], Phil Hykin[2], Sobha Sivaprasad[2]. [1]Visual Neuroscience, UCL-Institute of Ophthalmology, London, United Kingdom; [2]Research and Development, NIHR Biomedical Resource Centre at Moorfields Eye Hospital NHS Trust, London, United Kingdom.
Purpose: Sufficient capillary flow and dispersion of oxygenated blood is required for the maintenance of all tissue, with disruptions leading to disease. There is a gap in our understanding of how diabetic eye disease affects retinal oxygenation. Current analysis schemes are time consuming and do not provide comprehensive analysis of retinal oxygenation. There is a need to develop better analysis tools for understanding how oxygen saturation changes between normal and diabetic state. This knowledge is essentially for better treatment planning and prediction of therapeutic success.
Methods: Fourteen controls and twenty-one subjects with diabetes but without areas of non-perfusion were recruited for this prospective study. Subjects were imaged using SDOCT and Oxymap T1 retinal oximeter. Normal subjects were imaged twice, roughly 30 minutes apart to assess repeatability. For oxygen saturation measurements, image size was 35deg and centered at the optic nerve. Vessel size, location and saturation were exported using instrument software and analysed using custom Matlab software. Offline analysis separated arteriole and venule derived signals and assessed global oxygenation saturation, saturation by distance from the optic nerve, saturation by vessel diameter.
Results: Agreement between multiple measures of global saturation was +0.33% (-9.5 to 10.1% 95% confidence interval) for arteriole and -0.9% (-12.3 to 10.5% 95% confidence interval). There was no difference in global arteriole oxygen saturation, 91.6% in normal and 89.5% in diabetics, however there was a significant difference in venule saturation (63.7% verse 55.6%, $p = 0.0219$). The difference between arteriole and venule oxygen saturation was seen in the larger vessels; 100-120um (36.3 vs 42.1%; $p = 0.045$) and 120-140um (37.5 vs 45.2%; $p = 0.032$). The difference was due to venule saturation being higher in the diabetics than normal controls in larger vessels.
Conclusions: These data suggest that even without areas of non-perfusion, there is a fundamental change in retinal oxygen dispersion. The new tools developed here provide a more comprehensive analysis. The new data suggests that the lack of difference in small verse large vessel is probably an artefact of the greater pooling of blood in large vessels near the optic nerve. To further understand these changes, instrumentation to understand blood flow and oxygen dispersion in the capillaries is needed.
Commercial Relationships: Adam M. Dubis; **Luke Nicholson**, None; **Roxanne Crosby-Nwaobi**, None; **Phil Hykin**, Novartis (C), Bayer (C), Bayer (R), Novartis (F), Allergan (F), Alergan (C), Allergan (R), Bayer (F); **Sobha Sivaprasad**, Novartis (C), Bayer (C), Bayer (R), Novartis (F), Allergan (F), Allergan (R), Bayer (F), Allergan (C)
Support: The work was supported by grants from the National Institute for Health Research Biomedical Research Centre at Moorfields Eye Hospital National Health Service Foundation Trust, UCL Institute of Ophthalmology.

Program Number: 3034 **Poster Board Number:** A0029
Presentation Time: 11:00 AM–12:45 PM
Immunohistochemical characterization of neurotransmitters in the episcleral circulation in rats
Anja-Maria Ladek, Andrea Trost, Christian Runge, Falk Schroedl, Clemens Strohmaier, Herbert A. Reitsamer. Ophthalmology, Paracelsus Medical University, SALK, Müllner Hauptstrasse 48, 5020 Salzburg, Austria, Salzburg, Austria.
Purpose: Episcleral venous pressure (EVP) is an important parameter of steady-state intraocular pressure and recent evidence suggests neuronal influence on EVP. Yet little is known about the innervation and more specifically, the neurotransmitters involved. The present study aims at identifying possible neurotransmitter candidates for the episcleral circulation in rats.
Methods: Four previously untreated, enucleated, immersion fixated rat eyes, taken from three animals were cut into serial sections using a cryostat, followed by standard immunohistochemistry. Additionally

to the neurotransmitter under investigation, each slide was stained with antibodies against smooth muscle actin and neurofilament (200kDa).
Results: Throughout the slides neurofilament positive cells could be identified in close proximity to the vascular endothelium. These cells showed synapse like structures and stained positive for synaptophysine. Furthermore, they stained positively for ChAT, Galanin, CGRP, Substance P and PGP 9.5. Staining with antibodies against nNOS, Tyrosine Hydroxylase, Serotonin (5-HT) Transporter and Alarine yielded negative results.
Conclusions: These findings indicate that there is neuronal input to the episcleral circulation. However no assumption can be made whether the positively stained structures are of functional significance for the regulation of the episcleral venous pressure and thereby intraocular pressure.
Commercial Relationships: Anja-Maria Ladek, None; **Andrea Trost**, None; **Christian Runge**, None; **Falk Schroedl**, None; **Clemens Strohmaier**, None; **Herbert A. Reitsamer**, None

Program Number: 3035 **Poster Board Number:** A0030
Presentation Time: 11:00 AM–12:45 PM
Effect of anti-VEGF on retinal blood flow in diabetic mouse model
Praveen K. Balne[1], Neha Khandelwal[2], Sai Bo Bo Tun[1], Veluchamy A. Barathi[1], Rupesh V. Agrawal[2]. [1]Translational Pre Clinical Model Platform, Singapore Eye Research Institute, Singapore, Singapore; [2]Tan Tock Seng Hospital, Singapore, Singapore.
Purpose: To study the effect of anti-VEGF on retinal blood flow in Ins2 (Akita) diabetic retinopathy mouse model using laser speckle flowgraphy (LSFG) of optic nerve head (ONH) region.
Methods: The study was approved by the Institutional Animal Care and Use Committee of SingHealth, Singapore. All experiments were conducted according to the guidelines of the Association for Research in Vision and Ophthalmology (ARVO) for the use of animals in research. Fourteen eyes of eight Akita mice were included in the study. All the mice were anesthetized using the combination of ketamine hydrochloride (50 mg/kg) and xylazine hydrochloride (0.5 mg/kg). Intravitreal anti-VEGF (Eylea 1 µg/eye) injection was given to each eye and the IOP and retinal blood flow were measured longitudinally at baseline (pre-injection), day 7, 14, 21 and 28 post injection. For retinal blood flow the relative blood flow velocity "mean blur rate (MBR)" was measured for different regions of the ONH and the changes over the time and its association with IOP was analyzed by t-test.
Results: The age of the mice was ranged from 12 to 19 weeks. The MBR at baseline was 6.55±2.5, 6.99±6.34, 6.34±2.15, 6.5±2.13 and 7.63±3.2, 7.74±2.37, 7.85±3.10, 7.80±2.49 at 28 days post injection in superior, nasal, inferior and temporal regions of the ONH respectively ($p>0.05$). At all time points there was elevation in retinal flow, which was not statistically significant. There was also no statistically significant association between IOP and MBR ($p>0.05$).
Conclusions: Intravitreal anti-VEGF (Eylea) injection does not lead to significant changes in retinal blood flow in Ins2 (Akita) diabetic retinopathy mouse model.
Commercial Relationships: Praveen K. Balne, None; **Neha Khandelwal**; **Sai Bo Bo Tun**, None; **Veluchamy A. Barathi**, None; **Rupesh V. Agrawal**, None
Support: NMRC/CNIG14NOV005;

Program Number: 3036 **Poster Board Number:** A0031
Presentation Time: 11:00 AM–12:45 PM
Ocular and cerebral hemo-fluid dynamics in microgravity: a mathematical model
Giovanna Guidoboni[1], Fabrizia Salerni[3], Alon Harris[4], Christophe Prud'homme[7], Marcela Szopos[5], Peter M. Pinsky[6], Rodolfo Repetto[2]. [1]LabEx IRMIA, Strasbourg, France; [2]DICCA, University of Genova, Genova, Italy; [3]Physics and Earth Sciences, University of Parma, Prma, Italy; [4]Ophthalmology, Indiana University School of Medicine, Indianapolis, IN; [5]Université de Strasbourg, CNRS, IRMA UMR 7501, Strasbourg, France; [6]Mechanical Engineering, Stanford University, Stanford, CA; [7]Université de Strasbourg, CNRS, IRMA UMR 7501, Strasbourg, France.
Purpose: Long-term exposure to microgravity is known to lead to visual impairment in many astronauts (VIIP syndrome). Upper body fluid shift, alterations in intraocular pressure (IOP), intracranial pressure (ICP), tissue biomechanics and blood flow are among the many factors hypothesized to contribute to VIIP. Due to the difficulty to single out each of these factors using in-vivo studies, we present a mathematical model to theoretically evaluate the interactions between fluid flows and pressures in the brain (blood, cerebrospinal and interstitial fluid) and eyes (blood and aqueous humor) and their possible mechanical implications in VIIP.
Methods: We propose a 0-dimensional (0d) mathematical model of fluid circulation in the eyes and brain, embedded into a 0d whole-body circulation model. The model includes retinal, choroidal and ciliary circulations in the eyes. As suggested in the literature, the effect of microgravity is accounted for by i) considering zero hydrostatic pressure, ii) imposing zero central venous pressure, iii) decreasing the blood/aqueous humor oncotic pressure difference ($\Delta\pi_p$) and iv) increasing the blood-brain barrier permeability. The model simulates microgravity-induced fluid redistribution in the upper body vasculature and variations in IOP and ICP, which are critical factors for VIIP.
Results: The model predicts that, in microgravity conditions, ICP and IOP increase (Fig1a) and ocular blood flow decreases markedly in the choroid and ciliary circulations (Fig1b). Blood flow in the retina is found to experience smaller variations, owing to a purely mechanical perfusion control mechanism mainly enacted by the intraocular venous segments.
Conclusions: The model suggests that i) the venous segments play a fundamental role in controlling pressures and fluxes in the ocular circulation, owing to the possibility of their collapse, and ii) the retinal circulation is less susceptible to microgravity-induced alterations than choroid and ciliary. These findings point towards further clinical assessment of ocular venous function in microgravity as a potential determinant factor for VIIP.

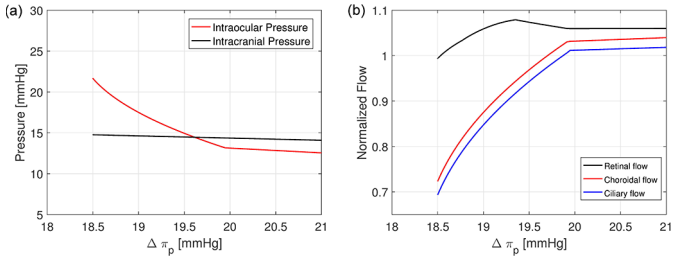

Fig 1: a) IOP and ICP and b) retinal, choroidal and ciliary blood flow vs blood/aqueous humor oncotic pressure difference $\Delta\pi_p$. Earth baseline values of IOP, ICP and $\Delta\pi_p$ are 15 mmHg, 11 mmHg and 25 mmHg, respectively. In space, $\Delta\pi_p$ is thought to decrease to ≈18.5 mmHg. Flows are normalized with the corresponding earth values.

Commercial Relationships: Giovanna Guidoboni, Indiana University Research and Technology Corporation (P); **Fabrizia Salerni**, None; **Alon Harris**, Ono Pharmaceuticals (C), ScienceBased Health (C), Oxymap (I), Nano Retina (C), AdOM (C), Biolight (C), Nano Retina (I), Isarna Therapeutics (C), Indiana University Research and Technology Corporation (P), AdOM (I), Stemnion (C); **Christophe Prud'homme**, None; **Marcela Szopos**, None; **Peter M. Pinsky**; **Rodolfo Repetto**, None
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Program Number: 3038 **Poster Board Number:** A0033
Presentation Time: 11:00 AM–12:45 PM
Flicker-Induced Retinal Vasodilation in Healthy Subjects
Leopold Schmetterer[2, 1], Mozhgan Sharifizad[1], Gerold Aschinger[3], Doreen Schmidl[1], Gerhard Garhofer[1], Rene M. Werkmeister[3]. [1]Clinical Pharmacology, Medical University of Vienna, Vienna, Austria; [2]Singapore Eye Research Institute, Singapore, Singapore; [3]CMPBME, Medical University of Vienna, Vienna, Austria.
Purpose: The aim of the present study was to analyze factors associated with retinal arterial and venous responses to stimulation with diffuse luminance flicker in healthy subjects.
Methods: We analyzed results obtained in 374 healthy subjects who had previously participated in clinical studies. A total of 153 subjects underwent a protocol in which flicker stimulation was delivered at a frequency of 8 Hz (protocol 1). In this protocol measurement and stimulation light are separated based on the wavelength. Another 221 subjects underwent a protocol in which diffuse luminance flicker was delivered at 12.5 Hz (protocol 2). Modulation depth in this protocol was almost 100%. We investigated whether sex, systemic blood pressure, baseline vessel size, blood plasma concentration of fasting glucose and hematocrit, and serum concentration of cholesterol, triglycerides, creatinine and C-reactive protein were associated with the retinal arterial and venous in response to flicker stimulation.
Results: Flicker responses in arteries and veins were more pronounced in protocol 2 as compared to protocol 1 ($P < 0.001$, each). In both protocols the vascular response to stimulation with diffuse luminance flicker was larger in smaller vessels (P between 0.001 and 0.016). In protocol 2 the retinal arterial flicker response was negatively associated with cholesterol serum levels ($P = 0.033$). In protocol 1, only a tendency toward this effect was observed ($P = 0.056$).
Conclusions: The present analysis indicates that retinal arterial and venous responses to stimulation with diffuse luminance flicker depend on the way the stimulation is delivered through the fundus camera. The protocol providing a modulation depth of almost 100% resulted in a more pronounced vascular response. In addition, the flicker response varied with vessel size: the smaller the vessel width, the larger the flicker response. Finally, our data indicate that, even within the normal range, higher cholesterol serum levels are associated with lower hyperemic flicker responses.
Commercial Relationships: Leopold Schmetterer, None; **Mozhgan Sharifizad**, None; **Gerold Aschinger**, None; **Doreen Schmidl**, None; **Gerhard Garhofer**, None; **Rene M. Werkmeister**, None

Program Number: 3039 **Poster Board Number:** A0034
Presentation Time: 11:00 AM–12:45 PM
Effect of Neuronal Nitric Oxide Synthase Deletion on Choroidal Blood Flow and Retinal Morphology and Function
Chunyan Li[1], Catherine Taylor[3], Natalie Guley[3], Nobel Del Mar[1], Malinda E. Fitzgerald[1, 2], Anton Reiner[1]. [1]Anatomy & Neurobiology, Univ of Tennessee Hlth Sci Ctr, Memphis, TN; [2]Biology, Christian Brothers University, Memphis, TN; [3]Univ of Tennessee Hlth Sci Ctr, Memphis, TN.
Purpose: Basal and adaptive vasodilatory control of choroidal blood flow (ChBF) is mediated by parasympathetic innervation from the pterygopalatine ganglion (PPG), which uses nitric oxide (NO) as a vasodilator, as well as vasoactive intestinal polypeptide (VIP). We examined the impact of hemizygous or homozygous deletion of the enzyme responsible for NO synthesis by the PPG, namely neuronal nitric oxide synthase (nNOS), on ChBF and on retinal structure and function.
Methods: The following assessments were made in male and female WT, nNOS+/-, and nNOS-/- mice from 90-500 days of age: 1) ChBF measured by transscleral Laser Doppler Flowmetry while monitoring arterial blood pressure (ABP); 2) visual acuity as measured using OptoMotry; and 3) retinal thickness as measured in plastic-embedded sections using StereoInvestigator.
Results: Basal ChBF was ~60% of WT in both nNOS+/- and nNOS-/- mice at 3 months of age, but baroregulation (i.e. stable basal ChBF during ABP fluctuation) appeared to be intact. Visual acuity, thickness of retinal layers and photoreceptor abundance did not differ among WT, nNOS+/- and nNOS-/- mice at 3 months of age. By contrast, visual acuity was only 60% of age-matched WT in nNOS+/- mice at 200-500 days of age. Surprisingly, visual acuity in nNOS-/- mice at 200-500 days of age was only reduced to ~85% of WT. A significant age-related decline in photoreceptor abundance and retinal ganglion cell layer thickness was seen over the 200-500 day range in nNOS+/- mice, but not in nNOS-/- mice. Total retinal and RPE thickness were reduced in both nNOS+/- and nNOS-/- mice compared to WT at 200 to 500 days of age.
Conclusions: Our studies indicate that basal ChBF is diminished in nNOS+/- and nNOS-/- mice, but baroregulation seems intact. Age-related decline in visual acuity was observed in both nNOS+/- and nNOS-/- mice, but surprisingly it was more severe in nNOS+/- mice. Moreover, although retinal thinning was seen in both nNOS+/- and nNOS-/- mice, acceleration of age-related ganglion cell and photoreceptor loss beyond one year of age was only seen in nNOS+/- mice. The basis of the greater age-related decline in nNOS+/- than nNOS-/- mice is uncertain, but may stem from diminished NO-mediated free radical injury in the nNOS-/- mice. In any case, our studies suggest the important role of NO-mediated parasympathetic regulation of ChBF in the long-term maintenance of retinal health.
Commercial Relationships: Chunyan Li, None; **Catherine Taylor**, None; **Natalie Guley**, None; **Nobel Del Mar**, None; **Malinda E. Fitzgerald**, None; **Anton Reiner**, None
Support: The Methodist Hospitals Professor of Neuroscience, RO1EY05298 (AR), Research to Prevent Blindness (MECF), Le Bonheur Fund 64-1015 and 64-1002 (CT), NSF DUE 9850780 (MECF), and The University of Tennessee Neuroscience Institute (CL).

Program Number: 3040 **Poster Board Number:** A0035
Presentation Time: 11:00 AM–12:45 PM
Retinal circulation changes with aging effect in normotensive healthy subjects
Tomofumi Tani[1], Young-Seok Song[1], Kenji Sogawa[1], Taknari Wada[1], Akihiro Ishibazawa[1], Tsuneaki Omae[1], Eiichi Sato[1], Shunsuke Nakamura[2], Jun Sakai[2], Masahiro Akiba[2], Takafumi Yoshioka[1], Akitoshi Yoshida[1]. [1]Ophthalmology, Asahikawa Medical University, Asahikawa, Japan; [2]Topcon corp, Tokyo, Japan.
Purpose: To report the effects of aging on the retinal microcirculation using a newly developed Doppler optical coherence tomography (OCT) auto-flowmeter (Auto-DOCT) in healthy male subjects.
Methods: We measured the retinal arterial blood flow in 33 healthy male volunteers (age; 21 to 68). The auto-DOCT system is based on spectral-domain OCT with fully automated measurement functionality. We measured blood velocity (V) and calculated the retinal pulsatility ratio (PR) in the retinal temporal arteries. PR is expressed as PSV/EDV, where PSV is the peak velocity during systole and EDV is the end diastolic velocity. Typically, an increased PR is an indicator of increased vascular resistance to flow and/or decreased vascular compliance, distal to the measurement site. Pearson's correlation analysis was used to determine whether PR was correlated with the aging.
Results: The PR showed strong correlation with age (r=0.62, p<0.0001), suggesting that the increased blood flow pulsatility in the retinal arteries with healthy male subjects is due to normal aging.
Conclusions: The current study showed that the auto-DOCT enables estimation of vessel aging from the measurements of retinal arterial blood flow in normotensive healthy humans.
Commercial Relationships: Tomofumi Tani; **Young-Seok Song**, None; **Kenji Sogawa**, None; **Taknari Wada**, None; **Akihiro Ishibazawa**, None; **Tsuneaki Omae**, None; **Eiichi Sato**, None; **Shunsuke Nakamura**, Topcon corp (E); **Jun Sakai**, Topcon corp (E); **Masahiro Akiba**, Topcon corp (E); **Takafumi Yoshioka**, None; **Akitoshi Yoshida**, Topcoat corp (P)

Program Number: 3041 **Poster Board Number:** A0036
Presentation Time: 11:00 AM–12:45 PM
Retinal Blood Flow in Glaucomatous Eyes with Single Hemifield Damage
takafumi yoshioka[1], Takayuki Kamiya[1], Motofumi Kawai[1], Tomofumi Tani[1], Seigo Nakabayashi[1], Masahiro Akiba[2], Akitoshi Yoshida[1]. [1]Asahikawa Medical University, Asahikawa Medical University, Asahikawa, Japan; [2]Topcon Corp, Tokyo, Japan.
Purpose: To examine the hypotheses that in glaucomatous eyes with single hemifield damage, retinal blood flow (RBF) is significantly reduced in retinal hemisphere corresponding abnormal visual hemifield; and that there are significant associations between MD value in abnormal hemifield, RBF, and structural measurements in the corresponding hemisphere.
Methods: Six eyes of 19 glaucoma patients with visual field loss confined to a single hemifield. Glaucomatous eyes underwent Doppler Optical Coherence Tomography Auto-Flowmeter (auto-DOCT) and standard automated perimetry. Auto-DOCT with a segmental scanning was used to measure RBF. RBF was derived from the recorded doppler frequency shift and the measured angle between the beam and the vessel. RBF in temporal artery and retinal nerve fiber layer (RNFL) values were calculated. Statistical analyses were performed.
Results: The RBF was reduced in abnormal hemisphere compared to the opposite hemisphere (6.86±2.33 vs 3.99±2.03μL/min, p<0.0001). The RNFL was thinner in the corresponding abnormal hemisphere compared with the opposite hemisphere (78.1±21.1, 104.3±15.5μm, p=0.0001). The RBF was correlated with RNFL (r=0.500, p=0.029) in the normal hemisphere, but was not correlated with RNFL (r=0.072, p=0.761) and MD value (r=0.215, p=0.3779) in the abnormal hemisphere.
Conclusions: In glaucomatous eyes with single-hemifield damage, the RBF is significantly reduced in the hemisphere associated with the abnormal hemifield. Reduced RBF is associated with thinner RNFL in the corresponding abnormal hemisphere.
Commercial Relationships: takafumi yoshioka; **Takayuki Kamiya**, None; **Motofumi Kawai**, None; **Tomofumi Tani**, None; **Seigo Nakabayashi**, None; **Masahiro Akiba**, Topcon Corp (E); **Akitoshi Yoshida**, Topcon Corp (P)

Program Number: 3042 **Poster Board Number:** A0037
Presentation Time: 11:00 AM–12:45 PM
***In ovo* chick chorioallantoic membrane (CAM) model to assess vascular reactivity**
catherine chia, Elizabeth Winters, Huyen Nguyen, Diego Tapias, Jeanette hyer, Jay M. Stewart, Ricardo Lamy. Ophthalmology, University of California San Francisco, San Francisco, CA.
Purpose: The chicken embryo chorioallantoic membrane (CAM) has been extensively used in many models for studying angiogenesis and surgical procedures. The purpose of this study is to develop a rapid and low cost *in vivo* model using CAM to assess vascular reactivity of eye drops and other topical drugs and drug candidates.
Methods: The *in ovo* CAM was prepared for experimentation without its inner shell membrane 48h before the application of the experimental drugs. A thin sterile silicone ring was applied on top of the CAM to demarcate the vascular area to be tested. Eye drops and topical drugs were applied inside the silicone ring and a series of pictures were taken at 0, 2, 5, 10, 15 and 20 minutes using a cellphone camera adapted to a microscope. The diameter of the vessels was assessed at each time point and compared using the publicly available ImageJ software.
Results: Changes in the vascular diameter were clearly observed and easily documented for image post processing and quantification. Vasodilation with a maximum increase of 106% on the vessel diameter was observed after 5 minutes of application of topical propranolol 6mg/ml solution. Phenylephrine 2.5% eye drops promoted the maximum observed vasoconstriction, with total closure of some vessels after 15 minutes.
Conclusions: This *in ovo* CAM model is a low cost, easily accessible, quantifiable, *in vivo* model to screen and study vascular reactivity of eye drops and other topical drugs.
Commercial Relationships: catherine chia, None; **Elizabeth Winters**, None; **Huyen Nguyen**, None; **Diego Tapias**, None; **Jeanette hyer**, None; **Jay M. Stewart**, None; **Ricardo Lamy**, None
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Human Retinal Vascular Reactivity to Flickering Light and Cold Water Immersion Measured by Laser Speckle Flowgraphy

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Purpose: We performed a prospective analysis of retinal blood flow, ocular perfusion pressure and changes in vascular resistance in response to stimuli in humans. We sought to understand the dynamics of vascular reactivity in response to two external stimuli: flickering light and hand immersion in ice water.
Methods: 12 normal subjects (6 male and 6 female, mean age=31; range 24-44) underwent laser speckle flowgraphy (LSFG-NAVI; Softcare LTD, Fukuoka, Japan) in the left eye following mydriasis with tropicamide 0.5%. Baseline flow measurements over 4 heartbeats were recorded in a retinal region of interest of approximately 15 x 10 degrees, including the optic nerve and fovea. Following baseline blood flow measurements, a 10Hz flickering light stimulus with 50% duty cycle and intensity of 14,600 cd/m2 was given for 2 minutes while recording flow and blood pressure at 30, 60, 90, and 120 seconds during and after the stimulus. The same sequence of measurements was also made during and after immersing the left hand into ice water.
Results: A light-evoked increase in retinal blood flow occurred at onset of light flicker and peaked at 90 seconds (mean=20%±14 increase; minimum 4%, maximum 44%). A parallel reduction in vascular resistance occurred (mean=14%±10; minimum 0%, maximum 29%). Cold-water hand immersion caused an increase in retinal blood flow peaking at 90 seconds (mean=15%±14; minimum 18% decrease, maximum 44% increase) with a concomitant rise in perfusion pressure. Vascular resistance increased (mean=18%±11, minimum 17% decrease, maximum 35% increase).
Conclusions: A flickering light caused an increase in blood flow and decrease in vascular resistance, representing a metabolic stress test. Cold-water caused an increase in retinal blood flow, but an increase in vascular resistance, associated with a rise in mean arterial blood pressure and ocular perfusion pressure. Cold-water immersion represents a sympathomimetic stimulus with corresponding autoregulation of retinal blood flow, resulting in vasoconstriction. These two stress tests may be useful for characterizing vascular reactivity in healthy and diseased states.

Commercial Relationships: Randy Kardon, None; **Cole Starkey**, None; **Jan M. Full**, None; **Anna Ketcham**, None; **Brian Tong**, None; **Gary Pierce**, None; **Seth Holwerda**, None; **Jess Fiedorowicz**, None
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