

325 ER stress and the unfolded protein response in ocular health and disease - Minisymposium

Tuesday, May 09, 2017 11:00 AM–12:45 PM

Ballroom 1 Minisymposium

Program #/Board # Range: 2940–2945

Organizing Section: Biochemistry/Molecular Biology

Contributing Section(s): Glaucoma, Low Vision, Physiology/
Pharmacology, Retina, Retinal Cell Biology

Program Number: 2940

Presentation Time: 11:05 AM–11:20 AM

Targeting the Unfolded Protein Response for the Prevention and Treatment of Diabetic Retinopathy

Sarah X. Zhang. Ophthalmology, University at Buffalo, Buffalo, NY.

Presentation Description: The endoplasmic reticulum (ER) is the primary cell machinery responsible for lipid and protein biosynthesis, protein folding and intracellular calcium storage. It also functions as a signaling hub when undesired changes disrupt the ER protein homeostasis. This condition, known as ER stress, activates signaling pathways of the unfolded protein response (UPR), through which the ER participates in many important cell processes and regulates cell fate. This presentation will discuss the role of the UPR in diabetes-related neurovascular injury of the retina and the potential of targeting UPR in the prevention and treatment of diabetic retinopathy.

Commercial Relationships: Sarah X. Zhang, None

Support: NIH/NEI grants EY019949 and EY025061, ADA research grant #7-11-BS-182, and an Unrestricted Grant to the Department of Ophthalmology, SUNY-Buffalo, from Research to Prevent Blindness.

Program Number: 2941

Presentation Time: 11:20 AM–11:35 AM

Targeting ER stress pathway for the treatment of glaucoma

Gulab Zode. The North Texas Eye Research Institute, Univ. of North Texas HSC, Fort Worth, TX.

Presentation Description: The pathological mechanisms leading to increased outflow resistance and intraocular pressure (IOP) elevation are poorly understood. We recently linked protein misfolding and endoplasmic reticulum (ER) stress to the development of glaucomatous trabecular meshwork (TM) damage and IOP elevation. TM cells activate protective unfolded protein response (UPR) pathway to eliminate abnormal protein accumulation. However chronic ER stress leads to induction of terminal UPR signals including ATF4 and CHOP that are known to induce cell death. Recently, we have shown significant increase in ATF4 and CHOP in human glaucomatous TM tissues. In the present study, we examined the role of ATF4/CHOP in the glaucomatous TM damage and IOP elevation. Adenoviral expression of Atf4 but not Chop significantly reduces outflow facility and elevates IOP in WT mice. Moreover, deletion of *Chop* protects from ER stress and IOP elevation in mouse models of glaucoma (*Tg-Myoc*^{Y437H} mice). CRISPR-Cas9 mediated ATF-4 or CHOP deficiency reduced myocilin accumulation in the TM cells and rescued glaucoma in *Tg-MYOC*^{Y437H} mice. Interestingly, expression of mutant myocilin leads to impaired autophagy and genetic knockdown of ATF4 and CHOP improved autophagic function of TM, reducing mutant myocilin accumulation and preventing ER stress in TM cells stably expressing mutant myocilin. Furthermore, improving autophagic flux by Tat-beclin 1 peptide reduced myocilin accumulation in TM cells and prevented IOP elevation in *Tg-MYOC*^{Y437H} mice. These studies provide a link between UPR and autophagy in myocilin glaucoma and further explore the possibility of treating glaucoma using Tat-beclin 1 peptide in degradation of abnormal protein accumulation.

Commercial Relationships: Gulab Zode, None

Support: NIH grant EY026177 and EY022077

Program Number: 2942

Presentation Time: 11:35 AM–11:50 AM

Involvement of ER stress in TULP1 induced retinal degeneration

Stephanie A. Hagstrom. Cole Eye Institute, Cleveland Clinic, Cleveland, OH.

Presentation Description: Inherited retinal disorders (IRDs) result in severe visual impairments in children and adults. A challenge in the field of retinal degenerations is identifying mechanisms of photoreceptor cell death related to specific genetic mutations. Mutations in the gene *TULP1* have been associated with two forms of IRDs, early-onset retinitis pigmentosa (RP) and Leber congenital amaurosis (LCA). *TULP1* is a cytoplasmic, membrane-associated protein shown to be involved in transportation of newly synthesized proteins destined for the outer segment compartment of photoreceptor cells; however, how mutant *TULP1* causes cell death is not understood. We have evidence that common missense mutations in *TULP1* express as misfolded protein products that accumulate within the endoplasmic reticulum (ER) causing prolonged ER stress. In an effort to maintain protein homeostasis, photoreceptor cells then activate the unfolded protein response (UPR) complex. Our results indicate that the two major apoptotic arms of the UPR pathway, PERK and IRE1, are activated. Additionally, we show that retinas expressing mutant *TULP1* significantly upregulate the expression of CHOP, a UPR signaling protein promoting apoptosis, and undergo photoreceptor cell death. Our data demonstrates that the ER-UPR, a known mechanism of apoptosis secondary to an overwhelming accumulation of misfolded protein, is involved in photoreceptor degeneration caused by missense mutations in *TULP1*. These observations suggest that modulating the UPR pathways might be a strategy for therapeutic intervention in *TULP1*-induced photoreceptor degeneration.

Commercial Relationships: Stephanie A. Hagstrom, None

Support: NIH Grant EY016072

Program Number: 2943

Presentation Time: 11:50 AM–12:05 PM

Achromatopsia mutations target sequential steps of ATF6 activation

Jonathan Lin. Pathology, UCSD, La Jolla, CA.

Presentation Description: Achromatopsia is an autosomal recessive disorder characterized by cone photoreceptor dysfunction. We recently identified Activating Transcription Factor 6 (ATF6) as a novel genetic cause of achromatopsia. ATF6 is a key regulator of the Unfolded Protein Response. In response to endoplasmic reticulum (ER) stress, ATF6 migrates from the ER to Golgi where it undergoes regulated intramembrane proteolysis to release a cytosolic domain containing a bZIP transcriptional activator. The cleaved ATF6 fragment migrates to the nucleus to transcriptionally upregulate protein folding enzymes and chaperones. ATF6 mutations in achromatopsia patients include missense, nonsense, splice site, and single-nucleotide deletion or duplication changes found across the entire gene. Here, we comprehensively tested the function of achromatopsia-associated ATF6 mutations and found that they group into three distinct molecular pathomechanisms. Class 1 ATF6 mutants show impaired ER to Golgi trafficking and diminished regulated intramembrane proteolysis and transcriptional activity. Class 2 ATF6 mutants bear the entire ATF6 cytosolic domain with fully intact transcriptional activity and constitutive induction of downstream target genes even in the absence of ER stress. Class 3 ATF6 mutants have complete loss of transcriptional activity due to absent or defective bZIP domains. Primary fibroblasts from patients with Class

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1 or Class 3 ATF6 mutations show increased cell death in response to ER stress. Our findings reveal that human ATF6 mutations interrupt distinct sequential steps of the ATF6 activation mechanism. We suggest that increased susceptibility to ER stress- induced damage during retinal development underlies the pathology of achromatopsia in patients with ATF6 mutations.

Commercial Relationships: Jonathan Lin, None

Program Number: 2944

Presentation Time: 12:05 PM–12:20 PM

ER Stress as a Gatekeeper of Cellular Senescence and Pathological Angiogenesis - for ER stress and the Unfolded Protein Response minisymposium

Przemyslaw Mike Sapiaha^{1,2}. ¹Ophthalmology, University of Montreal, Montreal, QC, Canada; ²Ophthalmology, Maisonneuve Rosemont Hospital, Montreal, QC, Canada.

Presentation Description: Proper retinal vascularization is vital for cellular function as it delivers oxygen, nutrients, hormones and immune cells and helps clear cellular debris and metabolic waste products. Conversely, pathological angiogenesis is the hallmark of blinding diseases such as diabetic retinopathy, retinopathy of prematurity and age related macular degeneration. Retinal angiogenesis occurs to satisfy energy requirements and cellular sensors of metabolic imbalance coordinate vessel growth. In this regard, classical pathways of the unfolded protein response activated under conditions of ER stress have recently been described to generate angiomodulatory or angiostatic signals. During retinal ischemia such as that occurring in retinopathies, pathways of ER stress are engaged and impact the vascular microenvironment. This occurs through modulation of the

retinal immune system or by prematurely triggering programs of cellular senescence in which cells adopt a senescence-associated secretory phenotype (SASP). This state of premature senescence is incompatible with vascular repair within the retina and contributes to disease progression. The role of ER stress in driving cellular senescence and its impact on vascular growth and ischemic retinopathies will be discussed.

Commercial Relationships: Przemyslaw Mike Sapiaha, SemaThera Inc (C), AmorChem Inc (F), SemaThera Inc (P)
Support: Wolfe Professorship, Canada Research Chairs, Canadian Institutes of Health Research (353770); Canadian Diabetes Association (OG-3-11-3329-PS)

Program Number: 2945

Presentation Time: 12:20 PM–12:35 PM

Targeting the unfolded protein response in retinal degenerative diseases

Marina Gorbatyuk. Optometry and Vision Sciences, UAB, Birmingham, AL.

Presentation Description: Persistent activation of the unfolded protein response (UPR) is known to contribute to the cellular mechanisms of retinal degeneration in various animal models including Retinitis Pigmentosa and Leber Congenital Amarois. Effects of modulations of the UPR markers on the progression of retinal degenerations will be discussed in the presentation. The key findings of the study revealed that diminishing the endoplasmic reticulum (ER) stress and the UPR marker activation may be feasible therapeutic approaches to retard retinal degeneration in mice.

Commercial Relationships: Marina Gorbatyuk, None