THE GRADUATE COLLEGE OF THE
UNIVERSITY OF OKLAHOMA HEALTH SCIENCES CENTER

ANNOUNCES THE FINAL EXAMINATION OF

Joseph L. Wilkerson

FOR THE DEFENSE OF THE DOCTOR OF PHILOSOPHY DEGREE
GRADUATE COLLEGE
Department of Cell Biology

January 8, 2018, 1 p.m.
Biomedical Research Center, Room 109

The role of sphingolipids in the maintenance of the neural retina and pathological neovascularization.

COMMITTEE IN CHARGE: Robert E. Anderson, MD, PhD, Nawajes A. Mandal, PhD, Eric W. Howard, PhD, Linda F. Thompson, PhD, James J. Tomasek, PhD, Dimitris Karamichos, PhD

ABSTRACT: Sphingolipids play many physiological roles in a cell. They are an essential part of the structure and rigidity of membranes. Cells tightly regulate sphingolipid species. Ceramides are the building block for many other sphingolipid species. However, an increase of ceramides within a cell activates apoptotic and pro-inflammatory pathways. To counter increases of ceramides cells must degrade it into sphingosine. Sphingosine is then quickly phosphorylated by one of the two sphingosine kinases to form sphingosine-1-phosphate (S1P). S1P serves as the ligand for 5 G-protein coupled receptors that, in general, initiate pro-survival, pro-migration, and anti-inflammatory pathways. S1P signaling is crucial during development. In the developing vasculature, S1P signals for adherens junctions to form endothelial-endothelial and pericyte-endothelial cell contacts. Without these contacts, vessels are unable to become flow competent and hemorrhage. S1P also plays a significant role in neural development. Migrating neuroblasts, destined for the olfactory bulb, follow an S1P gradient to navigate to where they will establish their neural connections. Both neurodegeneration and angiogenic pathologies are causes of vision loss. Because S1P is crucial for both neural and vascular development, it is an enticing target for research in the vision field. This study investigates the role S1P plays in two

pathologies that contribute to blindness: retinal degeneration and neovascular pathologies in the cornea. We used global knockout (KO) mice for sphingosine kinase-1 (*Sphk1*) to deplete extracellular S1P. In the neural retina, we show that S1P is necessary to maintain the outer limiting membrane (OLM). The OLM is composed of adherens junctions established by Müller glia with neighboring Müller glia and with photoreceptors. The loss of the OLM in the *Sphk1* KO mice is paired with a loss of retinal function, ascertained by electroretinography. Electron microscopy revealed that the retinal architecture collapses as the OLM is compromised. We go on to show in human cultured Müller glia that S1P can activate RAC1 leading to an increase in N-cadherin and rearrangement of the actin cytoskeleton. These findings indicate that S1P signaling in Müller glia may play a role in retinal degenerative disorders. We have also used *Sphk1* KO mice to study the effect of depleted plasmic S1P on neovascularization in the cornea. The cornea must remain transparent for adequate vision. A disruption of the corneal tissue by invading blood vessels leads to a loss of transparency and vision. In *Sphk1* KO mice, the cornea was protected from neovascular invasion after an injury. This corresponds to the loss of S1P seen in the blood plasma. These studies highlight the role of S1P and the SPHKs in retinal degeneration and neovascular pathologies of the cornea that result in vision loss.