University of Pittsburgh

Kenneth P. Dietrich School of Arts and Sciences
Department of Chemistry

603 Chevron Science Center
219 Parkman Avenue
Pittsburgh, Pennsylvania 15260
412.624.8520 (phone)
sweber@pitt.edu
www.chem.pitt.edu/person/steve-weber

Stephen G. Weber
Professor of Chemistry
Professor of Clinical Translational Science

October 26, 2017

Joshua S. Wesalo
1314S Chevron Science Center
219 Parkman Avenue
Pittsburgh, PA 15260

Dear Josh,

I am happy to serve as a collaborator on your project "Spatially controlled Nox2 inhibition for the targeted treatment of reperfusion injury in myocardial infarction." You have made a strong case for H_2O_2-responsive Nox2 inhibitors based on an extensive evaluation of the clinical and basic science literature. I believe that this approach has significant potential for treating the still unsolved problem of myocardial ischemia-reperfusion injury.

My group will collaborate with you on your proposed cell culture experiments. We have developed techniques to model ischemia-reperfusion in tissue culture by inducing changes in oxygen tension that occur nearly instantaneously. The technique has grown out of my longstanding work devoted to understanding redox biology in living tissue subjected to ischemic injury (see, for example, Wallin et al. 1999, Li et al. 1999, Wallin et al. 2000, Mitala et al. 2008, Jaquins-Gerstl et al. 2011, Yin et al. 2015, and Yin et al. 2017). Unlike previous approaches, we developed an online oxygenator with microfluidic channels containing a gas-permeable membrane. Applying a vacuum allows for rapid removal of perfusion solution from the tissue chamber. Our system has shortened the time required for tissue oxygen tension to equilibrate from over 12 minutes in the standard setup to below 30 seconds for our system. Consequently, this approach models ischemia-reperfusion much more realistically than previous approaches, as it changes oxygen tension nearly instantaneously. As you know, there are various processes that occur at different times following the onset of ischemia. Our approach will permit you to reliably establish the onset of "ischemia" in a culture and thus to rigorously establish the time course of peroxide production. So far, my group has demonstrated this in our model of ischemic stroke, and this collaboration is a great opportunity to expand the scope of our technique to myocardial ischemia-reperfusion. I am excited to collaborate with you and apply our innovative and highly effective superfusion setup to the evaluation of your new approach for treating myocardial reperfusion injury. Additionally, my group has an IACUC-approved protocol and routinely uses Sprague-Dawley rats to prepare tissue culture specimens for our stroke research using this system. As you proceed, you can obtain cardiac myocytes from the same animals concurrently.

My office is a mere 30 second elevator ride from yours, and my door is always open. I am pleased to field any questions you may have. Please note that our group has also extensive experience monitoring cellular redox status using H_2O_2- and GSH-responsive readouts, and we will be happy to answer questions that come up as you use these approaches to complete your proposed experiments. I am also happy to help and provide general advice during your graduate training. I am particularly attuned to the unique training requirements you face as an MSTP trainee, having mentored one myself (Amir Faraji, who is currently a PGY-5 neurosurgery resident at the University of Pittsburgh and whom you have shadowed). Best wishes as you continue your experimental work, and feel free to stop by any time!

Sincerely,

Stephen G. Weber, PhD
Professor of Chemistry
Professor of Clinical Translational Science