THE GRADUATE COLLEGE OF THE
UNIVERSITY OF OKLAHOMA HEALTH SCIENCES CENTER

ANNOUNCES THE FINAL EXAMINATION OF

Alanson W. Girton

FOR THE DEFENSE OF THE DOCTOR OF PHILOSOPHY DEGREE
GRADUATE COLLEGE

Department of Microbiology and Immunology

Thursday, May 3, 2018 | 1:00 pm
| Biomedical Research Center, Room 109 |

Innate Immune Responses to Bacillus anthracis Peptidoglycan Require Human Serum Opsonins and Occur Independently of TLR2

COMMITTEE IN CHARGE: K. Mark Coggeshall, PhD; Jimmy D. Ballard, PhD; Madeleine Cunningham, PhD, Eric Howard, PhD; Mark Lang, PhD

ABSTRACT: Late stage inhalational *B. anthracis* infection presents as fulminant bacteremia, which induces a sepsis-like pathophysiological state in both human patients and animal models. Our research has identified peptidoglycan (PGN) as one such PAMP that can induce activation of the innate immune system *in vitro* and is capable of recapitulating all aspects of live *B. anthracis* challenge in our non-human primate model.

Currently, our lab is focused on identifying the mechanisms by which the innate immune system recognizes PGN and subsequently drives the pathophysiology we observed *in vivo*. Previous research from our lab demonstrated that monocytes and neutrophils required serum factors, including naturally occurring anti-PGN IgG, to respond to PGN. However, we found no correlation between anti-PGN titer and PGN-specific activation of monocytes. Therefore, we hypothesized that other serum opsonins and/or cell surface receptors must be contributing to monocyte activation.

We first tested the ability of the surface Toll-like receptor 2 (TLR2), a suggested PGN pattern recognition receptor (PRR), to support macrophage activation. Using HEK 293 hTLR2-reporter cells and bone marrow-derived macrophages (BMDM) from either WT or TLR2-knockout mice, we discovered that neither *B. anthracis* PGN or PGN from a mutant strain of *S. aureus* devoid of lipoproteins was capable of activating these cells. From these data we concluded that TLR2 is not involved in PGN-mediated innate immune cell activation. We therefore analyzed the ability of another serum opsonin of carbohydrates, serum amyloid P to support monocyte responses to PGN. SAP supported a stronger cellular response to PGN than did IgG. Similar to IgG, we observed that inhibition of Syk kinase, Src-family kinases, phosphatidylinositol-3-kinases, and phagocytosis reduced the ability of the monocytes to respond to PGN stimulation, which indicates a possible involvement of Fcγ-receptors. However, unlike IgG, SAP appeared to have no role in PGN-induced complement activation. These data suggest convergent and divergent roles for IgG and SAP in supporting innate immune responses to PGN. Taken together, these studies provide insight into innate immune recognition of PGN.