Doctoral Dissertation Defense Announcement
"Borrelia burgdorferi peptidoglycan alters host immune responses"

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Committee in Charge:
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Chris Kristich, PhD
Anna Huppler, MD
Ann Rosenthal, MD

Date: Tuesday, February 27, 2024
Time: 11:00 AM (CST)
Defense Location: Bolger Auditorium
Zoom: https://mcw-edu.zoom.us/j/95172712965?pwd=blp1QVFFMnhld21qaEF3ME05L05Edz09
Meeting ID: 951 7271 2965 Passcode: Hu8eNnEM
Graduate Studies:
Intro to Biomedical Research
Biochemistry of the Cell
Techniques in Molecular & Cell Biology
Molecular & Cell Biology
Mechanisms of Cellular Signaling
Classical & Molecular Genetics
Cellular & Molecular Immunology
Ethics & Integrity in Science
Research Ethics Discussion Series
Statistics for Basic Sciences
Protein Chemistry Principles
Career Development in Biomedical Science
Mucosal Immunology
Immunological Tolerance
Classical Papers in Microbiology & Immunology
Immunology Journal Cub
Microbiology & Immunology Seminar Series
Reading and Research
Doctoral Dissertation
“*Borrelia burgdorferi* peptidoglycan alters host immune responses”

Fibroblast-like synoviocytes (FLS) promote wound healing and extracellular matrix (ECM) remodeling during resolution of inflammation and return to homeostasis. However, FLS are also associated with immune dysregulation and serve as a key cellular source of pro-inflammatory cytokines and chemokines in inflammatory joint diseases such as Lyme arthritis (LA). LA is the predominant late-stage manifestation of Lyme disease, caused by tick-borne spirochete *Borrelia burgdorferi*, which typically resolves with appropriate antibiotic therapy. However, in a subset of patients, LA can persist after antibiotic treatment, known as post-infectious (or antibiotic refractory) LA. Post-infectious LA is characterized by proliferative synovitis, dysregulated Th1/IFN$_\gamma$ responses and MHC-II-expressing FLS. Moreover, high levels of *B. burgdorferi* peptidoglycan (PG) have been found in LA patient synovia, yet the effects of PG on host immune responses are not well characterized. Thus, I hypothesize that PG acts as an adjuvant to promote pro-inflammatory effects of FLS and enhance CD4+ T cell activation in the joint.

To test this hypothesis, primary murine C57BL6/J (B6) FLS were isolated and cultured under disease conditions with pro-inflammatory molecules IFN$_\gamma$ and PG (or both simultaneously). To investigate FLS function as inducible antigen-presenting cells (APCs), MHC-II-activated FLS were co-cultured with CD4+ T cells and MHC-II-presented peptides were identified by mass spectrometry. The pro-inflammatory effects of FLS were further examined by co-culturing with NK cells and determining activation of innate inflammatory responses. Additionally, to investigate the effects of PG in other forms of inflammatory arthritis, we analyzed synovial samples derived from osteoarthritis (OA) patients for PG.

In summary, we demonstrate that murine FLS upregulate MHC-II in an IFN$_\gamma$-dependent manner, similar to what has been shown in FLS derived from Lap patients, and describe a mechanism for the transition to a pro-inflammatory FLS phenotype. We demonstrate that in the presence of IFN$_\gamma$, FLS exhibit impaired wound healing capacity and promote persistent inflammation by activating IFN$_\gamma$-producing NK cell responses in an IL-15-dependent manner. Moreover, IFN$_\gamma$-activated MHC-II+ FLS can act as APCs and are able to present Lyme autoantigens. However, only in the presence of PG do MHC-II+ FLS activate pro-inflammatory CD4+ T cell responses. Additionally, we show that presence of PG is associated with joint inflammation in OA. Together, these studies suggest that PG acts as an immune adjuvant enhancing the pro-inflammatory effects of MHC-II+ FLS and supports a role for PG in contributing to persistent inflammation in chronic inflammatory forms of arthritis.
Education and research
2018-present: Medical College of Wisconsin
Currently enrolled: PhD Biomedical Sciences, Department of Microbiology and Immunology
Doctoral dissertation: “Borrelia burgdorferi peptidoglycan alters host immune responses"
  - Doctoral coursework includes microbiology, biostatistics, bioethics, cell and molecular biology, genetics, cell signaling, biochemistry, and various advanced courses in immunology
  - Extensive oral communication and research presentation
  - Proficient in research techniques such as B. burgdorferi culture, primary cell culture, microscopy, mouse husbandry and organ harvest, in vivo murine infection models, lymphocyte isolation and proliferation assays, flow cytometry, multiplex cytokine immunoassay
  - Funding
    - Co-authored funding proposal for my primary research project, awarded by the Department of Defense (DOD) #FP00020687
    - Growth and Research in Immunology raining award in the amount of $10,000, awarded by the Center for Immunology at the Medical College of Wisconsin
  - Awards
    - AAI Trainee Poster Award, 2023
    - MCW Graduate Student Association Symposium Award, 2022
    - MCW Center for Immunology Travel Award, 2022
    - MCW Teaching excellence award, 2021
    - MCW Graduate School Conference Education Award, 2021-2023

2016-2018: University of Wisconsin-Parkside
M.S. Molecular Biology
  - Graduation date: May 2018, GPA: 3.6
  - Proficient in research techniques such as bacteria culture, primer design, growth assays, plasmid transformation, Miller assays, complementation assays, PCR, phage transduction, qPCR
  - Graduate coursework includes advanced molecular biology, advanced microbiology, and research seminar series

2013-2016: University of Wisconsin-Parkside
B.S. Molecular Biology and Bioinformatics
  - Graduation date: May 2016, GPA: 3.2
  - Thomson Research Fellowship Award, 2015
  - Trained in molecular biology techniques such as Northern Blot Assay, plasmid isolation and digestion, DNA sequencing, RNA isolation, protein purification, Bradford Assay, Perl and Python programming languages

Publications and Presentations
Meaghan N. Holub, Amanda Wahhab, Joseph R. Rouse, Rebecca Danner, Mecaila M. McClune, Jules M. Dressler, Klemen Sterle, Brandon L. Jutras, Adam I. Edelstein, and Robert B. Lochhead. “Peptidoglycan in osteoarthritis synovial tissue is associated with joint inflammation.” PREPRINT available at Research Square [https://doi.org/10.21203/rs.3.rs-2842385/v1]. Under review at Arthritis Research and Therapy.


2023: American Society for Microbiology
Poster presentation at the annual ASM: Microbe meeting in Houston, TX
- "Adjuvant effects of peptidoglycan on CD4+ T cell activation by MHC-II+ fibroblasts"

2023: American Association for Immunologists
Poster presentation at the annual AAI: Immunology meeting in Washington D.C.
- "Adjuvant effects of peptidoglycan on CD4+ T cell activation by MHC-II+ fibroblasts"

2022: International Conference on Lyme Borreliosis
Poster presentation at the 16th annual ICLB in Amsterdam, The Netherlands
- "Peptidoglycan acts as an immune adjuvant to enhance CD4+ T cell activation by MHC-II+ fibroblasts"

2022: MCW Center for Immunology
Abstract selected speaker at the 2022 Center for Immunology annual retreat in Milwaukee, WI
- "Peptidoglycan acts as an immune adjuvant to enhance CD4+ T cell activation by MHC-II+ fibroblasts"

2022: Gordon Research Conference
Abstract selected speaker at the 2022 GRC: Biology of Spirochetes in Ventura, California
- "Peptidoglycan acts as an immune adjuvant to enhance CD4+ T cell activation by MHC-II+ fibroblasts"

2019: University of Toledo
Poster presentation at the 2019 Midwest Microbial Pathogenesis Conference in Toledo, OH
- "Fibroblasts produce NK cell-activating factors to promote the arthritogenic IFNγ response in Lyme arthritis"
2017: University of Wisconsin-Madison
Poster presentation at the 2017 annual Molecular Genetics of Bacteria and Phages meeting in Madison, WI
  - "Characterizing novel regulatory effects of the global regulator Cra on the glucose-phosphate stress response in Escherichia coli."

Patents
Co-owner: Use of immunogenic T cell epitopes for Lyme disease vaccination and diagnosis (USPTO application 63/403,451)

Teaching and mentorship
2019-2023: Medical College of Wisconsin
Lab rotation mentor
  - Directly supervised training of 10 graduate student peers during their laboratory rotations
2021: Medical College of Wisconsin
Summer research program
  - Mentored medical students in conducting and presenting research projects, awarded the teaching excellence award
2021: Mount Mary University
Guest lecturer
  - Led classroom lecture on Lyme disease for microbiology students, focusing on basic research lab techniques
2018-2021: Medical College of Wisconsin
Student Health Initiative for Pipeline Programs (SHIPP) Graduate School Chair
  - Developed curriculum for students from underrepresented groups in health sciences
  - Provided mentorship and academic workshops for undergraduate students including lab tours and experimental techniques, how to read scientific publications, as well as interview skills

Leadership and service
2023: Medical College of Wisconsin
Clifton Strengths-based coaching program
  - Top 5 strengths: futuristic, maximizer, strategic, communication, activator
2022-2023: Medical College of Wisconsin
Invited and hosted guest speakers for the Department of Microbiology and Immunology seminar series
  - Nicole Baumgarth, DVM, PhD, Johns Hopkins Bloomberg School of Public Health
  - Elsio Wunder Jr, PhD, MS, DVM, Yale School of Public Health
2022-2023: Medical College of Wisconsin
Department Representative in the Graduate Student Association
  - Elected student government representative for the Department of Microbiology and Immunology
2021-2022: Medical College of Wisconsin
Enhancing Scholarly Culture Committee Chair
  - Organized and hosted science-based scholarly and social events known as the Science Social
  - Invited topic-expert guest speakers, gave public presentations, and directed group activities
2019-present: **Milwaukee LGBT Community Center**
Support Group Facilitator
- Lead support group meetings for male-identifying members of the LGBTQ+ community

2015-2016: **University of Wisconsin-Parkside**
Molecular Biology and Bioinformatics Club President
- Organized club events and hosted guest lecturers from academic and industry fields

**Professional Associations**
2022-present: American Society for Microbiology
2021-present: American Association of Immunologists

**Employment**
2015-2016: **University of Wisconsin-Parkside**
Laboratory Assistant
- Sanitation and sterilization of laboratory equipment and biohazard materials
- Instructional laboratory setup and microbiological culturing

2013-2015: **Oakwood Clinical Associates**
Clinical Assistant
- Customer service, clinical management of medical records and patient information
- Conducted employment interviews and participated in staff training
- Adherence to HIPAA and OSHA regulations

**Military Service**
**U. S. Army**
2009-2013: Active Duty Military
Deployments: Iraq, 2010; Afghanistan, 2012
Discharge: Honorable, 2017
Awards: Army Commendation Medal
Rank upon separation: E4, Specialist
Military Occupational Specialty: 68W Healthcare Specialist (Combat Medic)
- Leadership and tactical skills, emergency medicine and treatment techniques
- Specialty Training
  - Emergency Medical Technician certification, Fort Sam Houston, TX
  - Advanced Cardiovascular Life Support certification, Fort Sam Houston, TX
  - Basic Airborne Course paratrooper training, Fort Benning, GA