ABSTRACT

The complement system consists of three pathways (classical, lectin and alternative) and is an essential player in innate and adaptive immunity. Excessive activation of the alternative pathway (AP) occurs in various inflammatory conditions, resulting from, or contributing to, disease. Properdin, an oligomeric glycoprotein, acts as an essential positive regulator of the AP of complement by stabilizing enzymatic convertases. Identical properdin monomers form head-to-tail associations of oligomers in a reported 20:54:26 ratio (most often described as an approximate 1:2:1 ratio) of tetramers (P_4), trimers (P_3), and dimers (P_2), in blood, under normal physiological conditions. Oligomeric size is proportional to properdin function with P_4 being more active, followed by P_3, and P_2. The ratio of properdin oligomers in disease, which may affect function independent of concentration, becomes essential, yet remains unknown. This first objective of this study was to develop a novel ELISA-based technique to evaluate the function of properdin in biological samples with a readout that correlated with the oligomeric distribution of properdin in the sample. The second objective applied this technique to screen patient-derived biospecimens from microenvironments (i.e., ascites, synovial fluid) and circulation that represented a diverse range of inflammatory diseases known to involve complement to determine the relevance of properdin function. After identifying that properdin function was decreased in rheumatoid arthritis synovial fluid, size exclusion chromatography was used to determine that the lower function was due to a significantly different P_4:P_3:P_2 ratio in synovial fluid properdin (15:51:34) as compared to healthy serum (22:51:28). This phenomenon likely reflected both preferential consumption of P_4 and a newly described regulatory mechanism by which neutrophils, abundant in the joint space, release properdin as mainly P_2 to control the rate of complement activation in cellular microenvironments. The third objective characterized overall AP function and markers of AP activation/regulation in rheumatoid arthritis biospecimens to address the significance of properdin oligomer distribution to overall complement activity. AP activity and complement proteins levels were lower in synovial fluid compared to serum, representing a distinct complement profile in the sequestered joint space. Properdin oligomer distribution was strongly correlated to overall AP activity. The fourth objective related these findings to clinical variables and identified a strong correlation between the oligomer distribution in a rheumatoid arthritis serum sample and the disease duration of the patient. Overall, this study contributes to a fundamental understanding of properdin regulation in rheumatoid arthritis. Further studies are aimed at determining mechanisms that affect properdin oligomerization and expanding this work into additional disease cohorts.

DISSERTATION COMMITTEE

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DISSERTATION PRESENTATION

Sara R. Moore

April 19, 2022

Implications for Properdin, a Complement Regulatory Protein, in Rheumatoid Arthritis

Ph.D. in Biomedical Sciences

PUBLICATIONS

Moore SR*, Menon SS*, Galwankar N, Khuder SA, Pangburn MK, Ferreira VP. *Both authors contributed equally. "A Novel Assay that Characterizes Properdin Function Shows Neutrophil-Derived Properdin has a Distinct Oligomeric Distribution." Front Immunol. 2022. (*Under review*)

Moore SR, "*Protein key to understanding disease, immune response*. Toledo Blade. May 3, 2021.

Moore SR*, Menon SS*, Cortes C, Ferreira VP. *Both authors contributed equally. "Hijacking Factor H for Complement Immune Evasion." Front Immunol. 2021 Feb 25; 12:602277.

Bain W, Li H, van der Geest R, **Moore SR**, Olonisakin TF, Ahn B, Papke E, Moghbeli K, DeSensi R, Rapport S, Saul M, Hulver M, Xiong Z, Mallampalli RK, Ray P, Morris A, Ma L, Doi Y, Zhang Y, Kitsios GD, Kulkarni HS, McVerry BJ, Ferreira VP, Nouraie M, Lee JS. "Increased Alternative Complement Pathway Function and Improved Survival during Critical Illness". Am J Respir Crit Care Med. 2020 Jul 15;202(2):230-240.

AWARDS

Earl H. Freimer Award for Outstanding M.D./Ph.D. Student in Medical Microbiology & Immunology, 2022

Oral Presentation Award at the 28th International Complement Workshop, 2022.

University of Toledo Graduate Student Association Research Award, 2021.

Third place poster presentation at UT Graduate Research Forum, 2021.

University of Toledo Retirees Association Health Science Campus Scholarship, 2020.

Satellites of University of Toledo Auxiliary Biomedical Science Program Scholarship, 2020.

Joint Biology Consortium Research Microgrant (NIH funded), 2020.

FUTURE PLANS

Sara will return to medical school to complete her M.D. degree by May 2024.

PUBLISHED ABSTRACTS

Moore SR, Nigrovic PA, Sparks JA, Lee JS, Bain W, Khuder, SA, Ferreira, VP. "Implications for properdin, a complement regulatory protein, in disease." 28th International Complement Workshop, Dec. 2021, Molecular Immunology, 141:116-235.

Menon S, Galwankar N, **Moore SR**, Ferreira, VP. Assessment of function of neutrophil-derived properdin in a novel functional assay. J Immunol. 2020. 204(1 Supplement): p. 152.19. Conference: American Association of Immunologists, Immunology 2020.

Bain W, Li H, van der Geest R, **Moore SR**, Olonisakin TF, Ahn B, Moghbeli K, DeSensi R, Rapport S, Saul M, Hulver M, Xiong Z, Mallampalli RK, Ray P, Morris AM, Doi Y, Zhang Y, Kitsios G, McVerry BJ, Ferreira VP, Nouraie S, Lee J. Increased Alternative Complement Pathway Function Improves Survival During Critical Illness. Conference: American Thoracic Society 2020.