

Blood Microbiome
Pre-Affinity Research Collaborative
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Blood Microbiome ARC Premise and Goal:

With advancement in microorganism genome detection in both basic and clinical sciences, it is a current NIH priority to quantify and catalogue the microorganisms present in the human body, since it is likely they play a role in human health and disease. Most expertise in the area of “microbiome” is focused on the gastrointestinal tract, the largest, most complex microbiome. However, evidence from small studies from various disciplines, including Endocrinology, Nutrition, Periodontology, Cardiology, Microbiology and Infectious Disease suggests that the relevance of the gut microbiome may depend on the interaction with the bloodstream. Moreover, the periodontal microbiome in certain diseases (e.g. obesity and type 2 diabetes) may be as, if not more, clinically relevant. Simply stated, it is possible, if not probable, that a blood microbiome exists and, in parallel with what is known of the gut microbiome, may be altered in some disease states with important immunologic consequence. Prior investigations in the area of bacterial presence in the bloodstream have not involved an interface for multiple disciplines to collectively define the clinical importance of chronic systemic exposure to bacteria. Such a group of investigators would ideally include clinical researchers who treat at-risk population (s), scientists with expertise in species-specific immunologic responses and microbiologists with expertise in microorganism DNA/RNA detection. This ARC intends to provide this interface within the Evans Biomedical Research Center.

Background: Chronic systemic inflammation is thought to directly contribute to the development of insulin resistance, and to the development of type 2 diabetes (T2D) in obese subjects, although the cause is unknown. A recent study detected elevated levels of circulating lipopolysaccharide (LPS) in obese T2D, and postulated that this “metabolic endotoxemia” activates the innate immune system, leading to the production of inflammatory cytokines, chemokines and acute phase reactants that cause inflammation. The source or sources of circulating LPS is not known, but a leading hypothesis is that bacteria and bacterial products enter the blood after translocation through a comprised gut epithelium. Alterations in the bacterial composition of the gut microbiome differs between obese (e.g., more from the Firmicutes phylum) and non-obese subjects and changes with weight loss (increased Bacteroidetes). Whether differences in the gut microbiome are reflected in an alteration in the blood compartment and the degree of systemic inflammation is not known. Another contributor to intermittent increases in blood levels of bacteria includes periodontal disease (PD). PD is an inflammatory condition that is common in the general population, is more common in obesity and T2D, is often underdiagnosed/asymptomatic, and has been associated with intermittent bacteremia. PD in subjects without obesity and/or T2D have higher LPS levels than healthy controls without PD. Companion studies using the B lymphocyte as a model for systemic immunomodulation have shown that circulating B cells are chronically and differentially “primed” (by way of Toll-Like Receptor 2 and 4 expression) to respond to bacterial products, specifically LPS, in uninfected diabetic patients vs. non diabetics, and PD patients vs. non-PD. We hypothesize that in obesity, LPS is accompanied by bacteria or their products in the blood and that a specific blood microbiome exists in obese and T2D patients. We also predict that the makeup of the blood microbiome will reflect organisms from the GI tract and oral cavity and will differ depending on the level of obesity and glucose intolerance. Furthermore, a specific microbiome ‘signature’ may correlate with systemic inflammation and glucose

intolerance. Finally, there may alternatively be a protective microbiome in individuals who are at lower risk of developing T2D despite other risk factors.

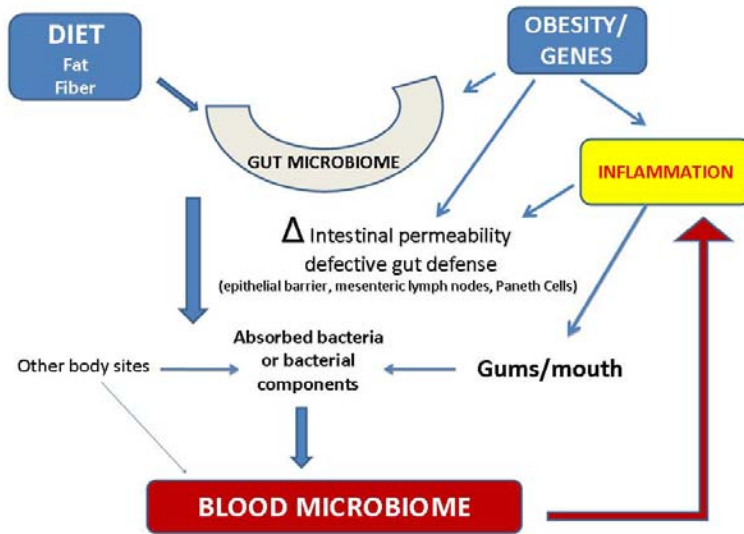


Figure 1: Type 2 diabetes is associated with increased circulating levels of LPS, which may result from enhanced movement (translocation) of bacteria from the gut lumen to the circulation. Enhanced movement of bacteria from the gut lumen occurs when there is a failure in the barrier function of the intestine and lymphatic system, or by active transport of bacteria by gut dendritic cells. Appearance of intact bacteria in the circulation could initiate and/or sustain the inflammatory process that links obesity, T2D and cardiovascular disease

Technology: We propose to investigate this hypothesis using technology developed by two extramural collaborators, Dr. Bruce Paster and Dr. David Ecker (see list of collaborators below).

Project 1: We will employ ongoing, IRB approved studies to analyze the blood microbiome in subjects with and without T2D, and with and without PD. We will study subjects in two conditions, both overnight-fasted and post-tooth cleaning by a dental hygienist testing the hypothesis that perturbations of mucosa in a way previously shown to induce bacteremia will exacerbate the level of 'metabolic endotoxemia' and shift the composition of the blood microbiome. Periodontal samples (saliva and/or plaque) will be obtained for bacterial sequencing as well. We will expect to find organisms native to the oral cavity in both the blood and mouth, but we also expect that the power of these two techniques will allow for detection of previously unidentified species of bacteria that may be associated with a blood microbiome. This will set the stage for future studies of the source (or sources) of the low level bacteremia that we expect to find in T2D. In addition to quantifying and identifying bacterial species present in the blood, we will perform complementary studies on blood will include flow cytometry analysis of B cell and monocyte TLR2 and TLR4 expression, measurement of adipocytokines and inflammatory cytokine expression using Luminex technology.

In summary, intermittent systemic exposure to bacteria in non-infectious chronic diseases could lead to persistent stimulation of the immune system with negative effects on health. These proof-of-concept studies will therefore poise our group to apply for NIH and other funding to characterize the clinical significance of bacterial speciation on multiple diseases and clinical settings.

ARC members and expertise:

Boston University:

Lisa Ganley-Leal, M.S., PhD (co-director). Section of Infectious Diseases. Differential innate immune host responses to microbial antigens in health and disease, with specific work in B cell phenotype and responses in noninfectious systemic inflammation.

Marie E. McDonnell, MD (co-director). Section of Endocrinology, Clinical Diabetes. Linking natural history of insulin resistant type 2 diabetes to innate immunity and systemic inflammation.

Caroline M. Apovian, MD. Section of Endocrinology, Clinical Obesity. Association between obesity, periodontal disease, systemic inflammation and negative clinical outcomes in obesity.

Caroline A. Genco, PhD. Department of Microbiology. Mechanisms of microbial virility that promote systemic effects of local infection, including innate immune recognition.

Frank C. Gibson III, PhD, Section of Infectious Diseases. Application of microbial environment to controlled models for mechanistic studies linking microbiome to systemic inflammation.

Hatice Hasturk, DDS, PhD, Goldman School of Dental Medicine, Department of Periodontology and Oral Biology. Associations between periodontal disease, metabolic disease and systemic inflammation

Alpdogan Kantarci, DDS, PhD Goldman School of Dental Medicine, Department of Periodontology and Oral Biology. Defining the inflammatory signature of the oral microbial environment in periodontal health and disease.

Forsyth Institute:

Bruce Paster, PhD. . Dr. Paster uses 16S rRNA sequencing to identify predominant bacterial species in the oral cavity, and a bacterial microarray, developed in his lab, to determine the microbial profiles of clinical samples of approximately 300 oral bacterial species, including those that are not presently cultivable.

IBIS technologies, Abbot

David Ecker, PhD Dr. Ecker has developed a unique method to detect low level bacteria in blood, namely PCR coupled to electrospray ionization mass spectroscopy (PCR/ESI-MS), which allows identification and quantification of bacterial species present in a subject's blood.

If you are interested in joining this pre-ARC, please contact one of the directors:

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