Metabolic profiling to dissect the role of HLA-B27 in axial spondyloarthritis

Sneha Couvillion[1], John Davis[2], Alec Furst[2], Henry Bringenberg[2], Kimberly Ogle[2], Ema Fale-Olsen[2], James T Rosenbaum[3], Ernesto Nakayasu[1] and **Tejpal Gill[2]**

[1]Pacific Northwest National Laboratories, [2]Oregon Health and Science University, [3]Legacy Devers and Corvus Pharmaceuticals

Background

Axial spondyloarthritis (axSpA) is an inflammatory form of spinal arthritis that can cause severe and chronic pain, and affects more than 3 million Americans. The association between axSpA and HLA-B27 is one of the strongest disease-genetic factor associations for a complex genetic disease. Studies from our group and other have shown the presence of gut microbial dysbiosis in axSpA however, most studies have revealed disparate bacterial species associated with SpA. Since bacterial function is redundant, this implies that perturbations of common metabolic functions can be involved in disease pathogenesis. In this study, *we aim to perform a comprehensive analysis of microbial metabolites in the fecal and plasma samples associated with HLA-B27 in healthy individuals and axSpA patients in comparison with the HLA-B27 negative healthy individuals*. This study provides a unique opportunity to define the local and systemic disease pathogenesis of axSpA through examination the of small molecules representing functional activity of both microbiome and the host (metabolome). Furthermore, combining biomarker development with novel computational approaches could lead to use of metabolic features for disease diagnosis, prognosis, or treatment in axSpA.

Metabolome and Spondyloarthritis

Diverse gut microbes, but common metabolic pathways correlate with dysregulated immune response in HLA–B27–induced experimental spondyloarthritis

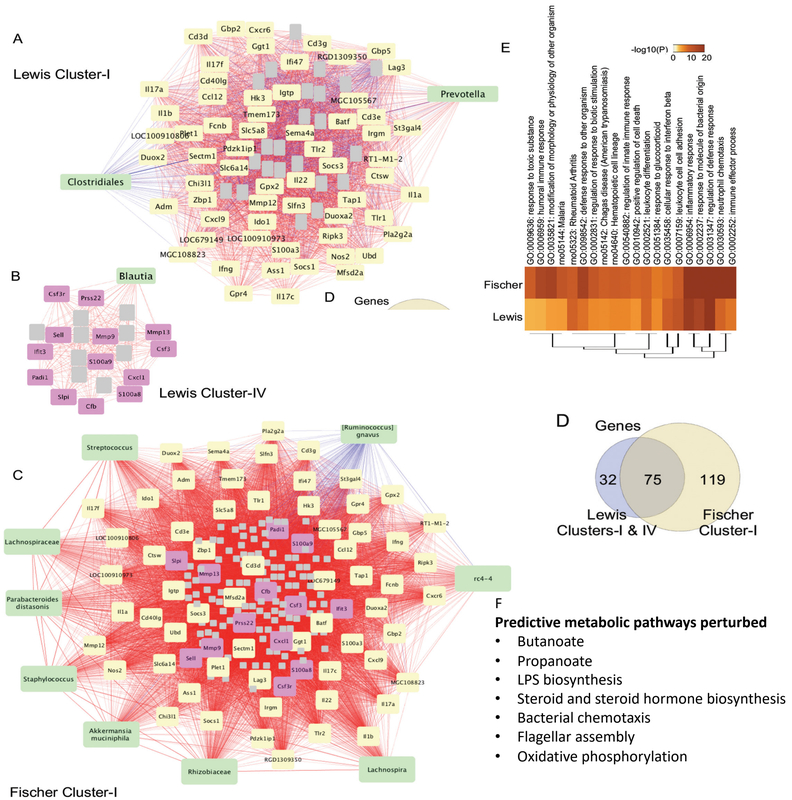

Figure 1: Gene–gene, gene–microbe, and microbe–microbe correlations were calculated using Cytoscape. **A–C**, Lewis and Fischer cluster I. Yellow and purple nodes represent gene clusters that overlap between Lewis and Fischer rats, while gray nodes represent non-overlapping gene clusters. and are not labeled. Lines represent positive correlations (**red**) and negative correlations (**blue**). **D**, Euler diagram showing the genes that overlap between Lewis clusters I and IV and Fischer cluster I. **E**, Heatmap representing the pathways determined using Metascape. **F,** PICRUST2 Inferred microbial metabolic pathways perturbed with inflammation.

Immune targeted fecal IgA-coated fecal microbes contribute to enriched inflammatory microbial metabolites and pathways and metabolites in axSpA patients

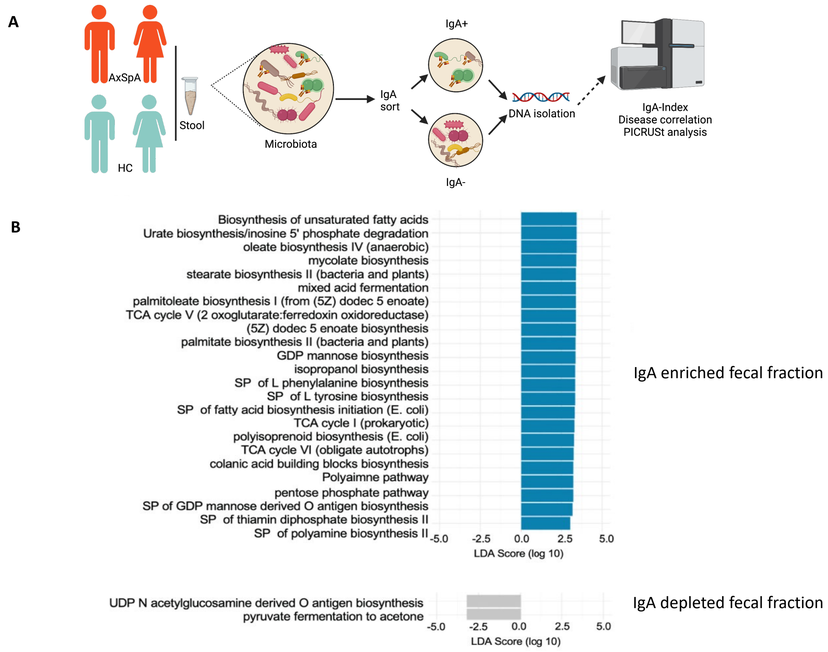

Figure 2: A, Representative methodology flowchart. B, Linear Discriminant analysis (LDA) effect size analysis showing differentially abundant MetaCyc metabolic pathways in IgA enriched and IgA depleted fecal fractions comparing axSpA patients with HCs. MetaCyc pathways and KEGG metabolites (KOs) with a class level alpha<0.05 and subclass alpha<0.05 are considered significant. SP (superpathways), TCA (tricarboxylic acid), GDP (guanosine diphosphate).

Metabolic Profiling in various Spondyloarthropathies

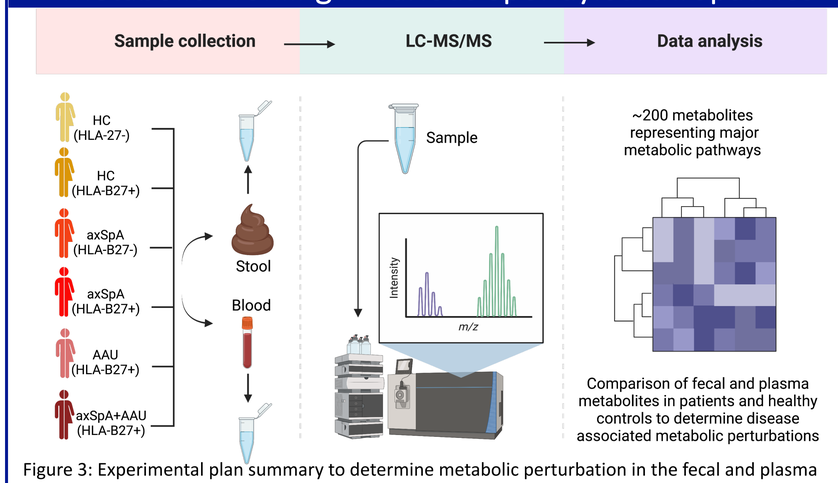

Figure 3: Experimental plan summary to determine metabolic perturbation in the fecal and plasma samples in patients with various spondyloarthropathies. We will use paired fecal and plasma samples from axSpA patients without a history of uveitis, patients with axSpA and acute anterior uveitis (AAU), AAU patients without AxSpA, HLA-B27 negative axSpA patients, along with HLA-B27 positive and negative healthy controls to determine their metabolome, and its association with HLA-B27 and disease. Metabolic profiling will be performed by LC-MS (Liquid chromatography-based mass spectrophotometry) of metabolites isolated from fecal and plasma samples. Based on our studies in an experimental model and inferred analysis from fecal samples of axSpA patients, we expect to detect distinct perturbations in the metabolic profiles which will correlate with the presence of HLA-B27 in healthy individuals and axSpA patients.

Metabolic profiling of fecal samples reveals altered abundance of metabolites in patients with axSpA with and without AAU in comparison to healthy controls

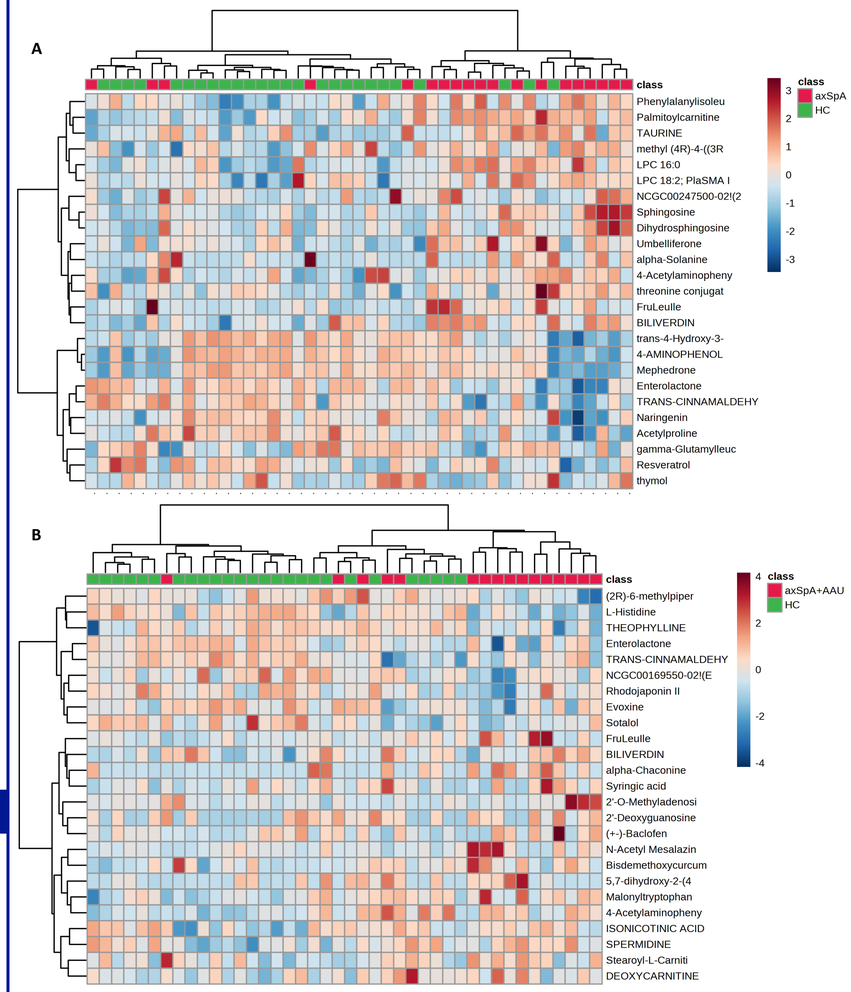

Figure 4: Heat map depicting abundance of fecal metabolites from A: patients with axSpA in comparison with healthy controls; B: patients with axSpA with AAU in comparison with healthy controls. The healthy controls are green and Patients are represented in red.

Summary/Future Directions

- Fecal metabolic proofing revealed altered abundance of metabolites in axSpA patients with and without AAU in comparison with healthy controls. Metabolic analysis on paired plasma samples is underway.
- Future studies involve correlating the fecal and plasma metabolic perturbations to determine disease associated changes in the local and systemic metabolic profile.
- Putative disease associated metabolites will be tested in the rat model of SpA to determine their role in disease pathogenesis.

In Conclusion, metabolic perturbation may be underlying disease pathogenesis and have the potential to yield therapeutic targets for the treatment of SpA

Acknowledgements

This study was supported by the Spondylitis Association of America (TG-PI) OHSU-PNNL Innovation award (TG-PI), GRAPPA and SPARTAN research awards (TG-PI).

Figures made with BioRender.com