Originally appearing as: Adams SB Jr, Setton LA, Nettles DL. The role of metabolomics in osteoarthritis research. J Am Acad Orthop Surg. 2013 Jan;21(1):63-4. doi: 10.5435/JAAOS-21-01-63. PMID: 23281473

The Role of Metabolomics in Osteoarthritis Research

Samuel B. Adams, Jr., MD[1]
Lori A. Setton, PhD[2]
Dana L. Nettles, PhD[2]

[1]Department of Orthopaedic Surgery, Duke University Medical Center, Durham, NC 27713
[2]Department of Biomedical Engineering, Duke University, Durham, NC 27710

Corresponding Author:
Samuel B. Adams, Jr., MD
Assistant Professor
Director of Foot and Ankle Research
Department of Orthopaedic Surgery
Duke University Medical Center
Box 2887, Durham, NC 27710
Phone: 919-660-5010
Fax: 919-660-5022
Email: samuel.adams@duke.edu

Metabolomics is the comprehensive analysis of small molecules in a biological system and has generated great interest for identifying novel biomarkers for disease diagnosis and pharmaceutical treatment. Metabolomics has been used to identify citrate and choline as biomarkers for prostate and breast cancer, respectively. In fact, both tests are now clinically available and supported by most health insurance providers.[1, 2]

Metabolites are the end-products of cellular processes, and their levels can be regarded as the ultimate response of biological systems to genotype, phenotype, and environmental conditions. They encompass a diverse group of low-molecular weight compounds including lipids, amino acids, peptides, nucleic acids, organic acids, vitamins, thiols, and carbohydrates,[3] and are commonly analyzed using nuclear magnetic resonance spectroscopy, liquid chromatography/mass spectrometry, and/or gas chromatography/mass spectrometry.

Metabolomics may be well suited for osteoarthritis (OA) research for many reasons including the tremendous heterogeneity in the disease process and recognition that no single bbiomarker can reflect the breadth of temporal and pathological processes associated with OA.[4] Combining several biomarkers into a panel will assuredly increase the discriminatory capability[5], as would occur with metabolic profiling. Secondly, since metabolic perturbations are real-time, they indicate the current disease state, a distinct advantage over current clinical diagnostics and disease monitoring techniques for OA, such as radiography.

Metabolomics has been employed to detect metabolic perturbations in urine, blood, synovium, and synovial fluid (SF) of animal models and patients with OA. Lamers and co-workers[6] used nuclear magnetic resonance to study the urine from guinea pigs that spontaneously develop OA. Disturbances in lactic acid, malic acid,

hypoxanthine and alanine were found to contribute heavily to the metabolic profile of OA. These investigators further studied the urine of humans with and without OA and found distinct patterns in the NMR spectra that could discriminate between groups.[4] Zhai and co-workers[7] employed metabolomics on human serum in a study of patients with and without knee OA. The investigators demonstrated that the ratios of valine and leucine to histidine were predictive of OA, pointing towards interest in the use of branched-chain amino acids (BCAA) as potential biomarkers.[7] Many of these studies of venous plasma or urine identify metabolites that may be related to aging, altered muscle mass, and other factors that may confound the unique signature of a pathological OA joint. For these reasons, SF may yield the most accurate, real-time, and joint-specific metabolic profile.

Metabolomics has been performed on SF from experimentally induced OA in canine knee joints.[8] The nuclear magnetic resonance spectra demonstrated increased concentrations of lactate, pyruvate, glycerol, alanine, isoleucine, hydroxybutyrate, hydroxyisobutyrate, and lipoprotein associated fatty acids. The investigators concluded that the intra-articular environment of OA was more hypoxic and acidotic than in the normal joint, and that arthritic joints may rely in part upon altered lipid metabolic pathways.

We have performed metabolomics on both human synovium and SF. In our first study, the metabolic profile of conditioned media collected from synovium explant cultures was obtained for tissues from patients undergoing total knee arthroplasty (end-stage OA) and from patients undergoing ligament or meniscal repair with little or no evidence of OA.[9] Thirteen metabolites were significantly elevated in the end-stage OA group and included glutamine, succinate, pro-hydroxyproline, amongst others. Despite

results suggestive of a distinct metabolic profile in OA, the synovium culture method does not easily translate into clinical practice.

In a second study, we performed metabolomics on ankle SF of patients with and without ankle OA.[10] Results identified 106 metabolites as significantly elevated in the OA samples and represented perturbations in virtually all metabolic pathways, including amino acid metabolism, carbohydrate metabolism, mitochondrial oxidation, lipid metabolism, peptide, vitamin, and nucleotide synthesis, and redox homeostasis. More importantly, when a rigorous decision tree analysis was applied to the metabolic profiles of the two populations, a 90% discriminatory accuracy was achieved, indicating the potential use of this technology as a diagnostic tool for OA. Studies are ongoing to confirm these findings in a larger population and to generate a narrowed panel of metabolic biomarkers and measurement methods for translation into the clinic.

In conclusion, metabolic profiling of biofluids and tissues can provide a panoramic view of the current physiologic state of a biological system such as the intra-articular environment of an osteoarthritic joint. We envision the role of metabolomics for OA as a clinically applied diagnostic tool in which a sample of a patient's synovial fluid would be analyzed for a panel of metabolite biomarkers, similar to following serial values from a complete blood count. Alterations in the metabolic profile could indicate disease progression or a therapeutic response at a resolution not possible with currently employed clinical techniques.

References

1. Serkova NJ, Spratlin JL, Eckhardt SG. NMR-based metabolomics: translational application and treatment of cancer. *Curr Opin Mol Ther.* Dec 2007;9(6):572-585.

2. Bartella L, Thakur SB, Morris EA, et al. Enhancing nonmass lesions in the breast: evaluation with proton (1H) MR spectroscopy. *Radiology.* Oct 2007;245(1):80-87.

3. Zhang A, Sun H, Wang P, Han Y, Wang X. Recent and potential developments of biofluid analyses in metabolomics. *Journal of proteomics.* Feb 2 2012;75(4):1079-1088.

4. Lamers RJ, van Nesselrooij JH, Kraus VB, et al. Identification of an urinary metabolite profile associated with osteoarthritis. *Osteoarthritis Cartilage.* Sep 2005;13(9):762-768.

5. Garnero P, Rousseau JC, Delmas PD. Molecular basis and clinical use of biochemical markers of bone, cartilage, and synovium in joint diseases. *Arthritis Rheum.* May 2000;43(5):953-968.

6. Lamers RJ, DeGroot J, Spies-Faber EJ, et al. Identification of disease- and nutrient-related metabolic fingerprints in osteoarthritic Guinea pigs. *J Nutr.* Jun 2003;133(6):1776-1780.

7. Zhai G, Wang-Sattler R, Hart DJ, et al. Serum branched-chain amino acid to histidine ratio: a novel metabolomic biomarker of knee osteoarthritis. *Ann Rheum Dis.* Jun 2010;69(6):1227-1231.

8. Damyanovich AZ, Staples JR, Chan AD, Marshall KW. Comparative study of normal and osteoarthritic canine synovial fluid using 500 MHz 1H magnetic resonance spectroscopy. *J Orthop Res.* Mar 1999;17(2):223-231.

9. Adams SB, Jr., Setton LA, Kensicki E, Bolognesi MP, Toth AP, Nettles DL. Global metabolic profiling of human osteoarthritic synovium. *Osteoarthritis Cartilage.* Jan 2012;20(1):64-67.

10. Adams SB Jr. KE, Jones LC, Haile A, Miller SD, Gutyon GP, Schon LC. Inflammatory cytokine composition and metabolic profile of post-traumatic ankle joint arthritis. *Transactions of the Orthopaedic Research Society.* 2012;37:Poster 1788.