subcutaneous AT by liposuction and characterized T cell phenotypes by flow cytometry (N=7 paired samples).
Results: GSK decreased sEH activity in plasma (47.3% vs placebo; P=0.008) and in AT (58.8% vs placebo; P=0.002). GSK also decreased serum F2-isoprostanes (P=0.03), which are markers of oxidative damage and inflammation. In seven paired AT samples, T helper (Th) 1 cells producing the pro-inflammatory cytokine IFNγ were reduced by treatment with GSK as compared with placebo (% of total lymphocytes: Placebo 13.6% ± 6.9, GSK 11.0% ± 5.6, P=0.03 Wilcoxon Signed Rank). In this small sample, we did not detect significant differences in the percentage of other IFNγ-producing cells (natural killer: Placebo 19.0% ± 9.0, GSK 13.3% ± 4.9, P=0.18; CD8: Placebo 12.0 ± 11.0, GSK 6.1 ± 4.6, P=0.61). In addition, we did not detect any change in Th17, Th2, or regulatory T cells.
Conclusions: In a pilot study of seven individuals treated with placebo or an sEH inhibitor, we found that the sEH inhibitor decreased pro-inflammatory Th1 cells as compared with placebo in matched AT samples. Understanding the contribution of the EET/sEH pathway to inflammation in obesity could lead to new strategies to modulate AT and systemic inflammation and reduce the risk of CVD.

Adipose Tissue, Appetite, and Obesity
NOVEL MECHANISMS CONTROLLING ADIPOSE TISSUE PHYSIOLOGY AND ENERGY BALANCE

The Effects of Diet Induced Obesity (DIO) on Skeletal Muscle Transcription in MuRF1 KO Mice

Andrea Gail Marshall, PhD[1], Jennifer E. Norman, Ph.D.[2], Michael S. Chementi, Ph.D.[1], John C. Rutledge, MD[2], Sue C. Bodine, Ph.D.[1].
[1]The University of Iowa, Iowa City, IA, USA, [2]The University of California, Davis, Davis, CA, USA.

Background: As obesity and Type II Diabetes rise globally, it is important to understand the similarities and differences in the response of metabolic tissues between males and females. We wanted to evaluate the impact of prolonged diet induced obesity (DIO) on the skeletal muscle transcriptome of our MuRF1 KO (KO) mice. **Methods:** RNA was isolated from the gastrocnemius muscle of male and female WT and KO mice that were fed either standard chow (Envigo 2918) or a 45% HFD (Research Diets D12451) for 22 weeks (n = 4). RNA was enriched for mRNA prior to library preparation. RNA sequencing was performed using 150 bp paired-end reads (~ 31.6 M reads per sample). Differentially expressed genes (DEGs) were identified using DESeq2 with an FDR set to 5%. **Results:** At baseline (chow diet), both male and female KO mice had DEGs compared to their WT counterparts (male, 1174; female, 105). Most DEGs were found to be unique by sex (male, 1151; female, 82), though 23 genes were found to be changed in common. After obesity was induced by 22 weeks of 45% HFD feeding, KO animals showed a greater transcriptional response than their WT counterparts. Males had 1821 DEGs (v. 179 in WT) while females had 4425 DEGs (v. 2090 in WT). In males, 78 genes were changed in common between WT and KO in response to DIO, with 76 of those genes changing in the same direction (Slc282a and Gm15427 did not). In females, 1445 genes were changed in common between WT and KO, with all but 2 genes (Pla2g7 and Zfp385b) changing in the same direction. In both male and female KO animals, oxidative phosphorylation and ribosomal pathways were most significant, though the direction of change in the DEGs was opposite. **Conclusion:** In skeletal muscle, sex highly influences the genes and pathways changed in response to DIO. Even among common pathways identified, the response between males and females differed. Loss of MuRF1 results in common and unique transcript changes in and between males and females under normal conditions and in DIO.

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NOVEL MECHANISMS CONTROLLING ADIPOSE TISSUE PHYSIOLOGY AND ENERGY BALANCE

The Pesticide Chlorpyrifos Promotes Obesity by Inhibiting Diet-Induced Thermogenesis

Bo Wang, Ph.D.[1], Shuman Zhang, MS[1], Andrea Llanos, MS[1], Evangelia E. Tsakiridis, MS[1], Eric M. Desjardins, MS[1], Julian M. Yabut, PhD[1], Alexander E. Green, PhD[1], Emily A. Day, PhD[1], Brennan K. Smith, PhD[1], James S.V. Lally, PhD[1], Amogelang R. Raphenya, PhD[1], Krishna A. Srinivasan, PhD[1], Andrew G. McArthur, PhD[1], Shingo Kajimura, PhD[2], Jagdish Suresh Patel, PhD[3], Michael Gordon Wade, PhD[4], Katherine Mary Morrison, MD,FRCP[5], Alison Holloway, PhD[1], Gregory R. Steinberg, PhD[1].
[1]McMaster University, Hamilton, ON, Canada, [2]UCSF/Barnett Briggs Medical Lib, San Francisco, CA, USA, [3]University of Idaho, Moscow, ID, USA, [4]HEALTH CANADA HPFB, Nepean, ON, Canada, [5]McMaster University Medical Center, Dundas, ON, Canada.

Obesity is a major risk factor for type 2 diabetes (T2D), non-alcoholic fatty liver disease (NAFLD) and cardiovascular disease that arises from a caloric surplus of as little as 10–30 kcal per day. And while increased consumption of energy dense foods and reduced physical activity are commonly thought to be the major contributors to this caloric imbalance, diet-induced thermogenesis is a quantitatively important component of the energy balance equation. In adult humans, recent studies have indicated that diet-induced thermogenesis requires the activation of brown adipose tissue (BAT), however, the determinants regulating this process and why they may differ between individuals are not fully understood. We hypothesized that environmental toxicants commonly used as food additives or pesticides might reduce diet-induced thermogenesis through suppression of uncoupling protein 1, the defining protein of human BAT thermogenesis. Through a screening approach of pesticides/toxicants chosen from the Toxcast chem Library, we discovered that the organophosphate insecticide chlorpyrifos potently suppressed the expression of uncoupling protein 1 (UCP1) and mitochondrial respiration in brown adipocytes at concentrations as low as 1 pM. Chloropyrifos-induced suppression of brown adipocyte thermogenesis was also observed in mice fed a diet high in fat and housed at thermoneutrality where it promoted greater obesity, non-alcoholic fatty liver disease and insulin resistance. Reductions in thermogenesis by chlorpyrifos were associated with impaired activation of the β_3-adrenergic receptor and protein kinases critical for

regulating UCP1 and mitophagy. These data indicate that the commonly used pesticide chlorpyrifos, at doses found within the food supply, suppresses the activation of brown adipose tissue, suggesting that its use may contribute to the obesity epidemic.

Adipose Tissue, Appetite, and Obesity
NOVEL MECHANISMS CONTROLLING ADIPOSE TISSUE PHYSIOLOGY AND ENERGY BALANCE

TNF Alpha-Induced SOX2 Expression Promotes Hepatic Steatois in Diet-Induced Obesity Model

Chen Shen, PhD, Jin Hong Chen, MS, Haram Oh, BS, Ji Hyun Park, MD,PhD.
Jeonbuk National University Medical School, Jeonju, Korea, Republic of.

Diet-induced obesity can cause metabolic or inflammatory damage on liver. Nonalcoholic fatty liver disease (NAFLD) begins with the fat accumulation in hepatocyte, but can lead to hepatocellular carcinoma (HCC). Sex-determining region Y-box 2 (SOX2) is a critical transcription factor involving regeneration and pluripotency. The expression level of Sox2 is correlated with progression of HCC, and anti-inflammatory effects of Sox2 in mesenchymal stem cells have been found. However, the expression of Sox2 by inflammatory cytokines in hepatocyte in NAFLD or the role of SOX2 in fat accumulation has been rarely reported. Here, we found that high-fat diet feeding, with or without high fructose in drinking water, significantly upregulated SOX2 in the livers of mice. In vitro, treatment with free fatty acids (FFAs) and fructose increased SOX2 expression in FL83B cells compared with the vehicle-treated group. Furthermore, overexpression or knockdown of SOX2 in FL83B cells promoted or suppressed, respectively, triglyceride synthesis and lipid accumulation after FFAs stimulation. The expression levels of several lipogenesis-related molecules were found to be altered by SOX2 expression. In addition, among several cytokines, only the treatment of tumor necrosis factor-alpha (TNFα) increased the SOX2 expression compared with the vehicle-treated control. Further, upregulation of (TNFα) by FFA/fructose was observed, and TNFα and FFA/fructose induced SOX2 expression was abolished by pretreatment of a TNFα inhibitor. Collectively, our findings suggest that TNFα-SOX2 signaling pathway in hepatocyte may be one of targets for early prevention of the development of NAFLD.

Adipose Tissue, Appetite, and Obesity
NOVEL MECHANISMS CONTROLLING ADIPOSE TISSUE PHYSIOLOGY AND ENERGY BALANCE

Unraveling Secretory Mechanisms that Control Pentraxin 3 Secretion in Adipocytes During Inflammation

TE-YUEH LIN, PhD, XiaoLi Chen, MD, PhD.
University of Minnesota, St Paul, MN, USA.

As a soluble pattern recognition receptor, Pentraxin 3 (PTX3) plays an important role in innate immunity and obesity-associated metabolic inflammation. PTX3 is abundantly expressed and secreted in adipocytes in response to lipopolysaccharide (LPS) stimulation. Appropriate regulation of PTX3 secretion is critical for maintaining inflammatory homeostasis. This study aims to unravel the mechanisms that control PTX3 secretion in adipocytes during LPS-induced inflammation. Upon 6h treatment of LPS, PTX3 expression and secretion were significantly induced in 3T3-L1 and stromal-vascular (SV) differentiated adipocytes, but to a lesser extent in SV cells or 3T3-L1 fibroblasts. However, LPSdoes not significantly stimulate PTX3 expression and secretion in macrophages. Using chemical inhibitors of conventional and unconventional protein secretion, we explored the mechanisms for controlling LPS-stimulated PTX3 secretion. 3T3-L1 adipocytes were treated with LPS for 6h in the presence or absence of various inhibitors blocking protein secretion from the Golgi complex (Monensin and Brefeldin A), mitochondrial oxidation (**carbonyl cyanide 3-chlorophenylhydrazone** [CCCP]), autophagy-lysosome (chloroquine and 3-methyladenine) and inflammasome (Bay 11–7082 and wedelolactone) activation, or exosome synthesis and trafficking (GW4869, manumycin A, calpeptin, and Y-27632). There were no significant effects of all inhibitors except for Monensin, Brefeldin A, and CCCP on intracellular and secreted levels of PTX3 in adipocytes. We found that Monensin and Brefeldin A significantly blocked LPS-stimulated PTX3 secretion, resulting in cellular PTX3 accumulation in adipocytes. Disrupting mitochondrial membrane potential by CCCP caused the reduction in PTX3 secretion from adipocytes. Additionally, we detected PTX3 in exosomes isolated from LPS-treated adipocytes. Inhibiting exosome synthesis by Manumycin A attenuated LPS-stimulated PTX3 secretion in both adipocyte culture media and isolated exosomes but not in the non-exosomal fraction of media, suggesting the involvement of the exosomal pathway in PTX3 secretion. However, the levels of exosomal PTX3 were significantly lower than that of the non-exosomal PTX3, and only 4.3% of secreted PTX3 was detected in the exosomal fraction of cultural media. Inhibiting the Golgi complex pathway blocked both the exosomal and non-exosomal secretion of PTX3 in adipocytes. After further fractionation of isolated crude exosomes by the iodixanol density gradient centrifugation, we showed that the majority of PTX3 was found in the non-extracellular vesicular (EV) fractions; only a small portion of secreted PTX3 overlapped with the exosomal marker CD63 in the small EV fractions. We conclude that PTX3 is secreted mainly through the conventional protein secretion pathway and minimally through the exosomal or EV pathway in response to LPS stimulation.

Adipose Tissue, Appetite, and Obesity
THE RELATIONSHIP BETWEEN COVID-19 AND ENDOCRINOLOGY

Antiandrogens Target TMPRSS2 and Reduce SARS-CoV-2 Virus Entry in Lung Cells

Damien A. Leach, PhD[1], Mohr Andrea, PhD[2], Ralf Zwacka, PhD[2], Stathis Giottis, PhD[2], Laura Yates, PhD[1], Clare Lloyd, PhD[1], Greg N. Brooke, PhD[3], Charlotte Lynne Bevan, PhD[1].
[1]IMPERIAL COLLEGE LONDON, London, United Kingdom, [2]University of Essex, Colchester, United Kingdom, [3]Essex University, Colchester, United Kingdom.