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PBDEs and Gut Microbiome Modulate Metabolic Syndrome-Related Aqueous Metabolites in Mice

David K. Scoville¹, Cindy Yanfei Li¹, Dongfang Wang^{2,3}, Joseph L. Dempsey¹, Daniel Raftery³, Sridhar Mani⁴, Haiwei Gu^{*5}, and Julia Yue Cui^{*1}

DKS, CYL, JLD, and JYC: ¹Department of Environmental and Occupational Health Sciences, University of Washington, Seattle, WA 98105, USA;

DW: ²Department of Laboratorial Science and Technology, School of Public Health, Peking University, Beijing 100191, P. R. China;

DW, DR: ³Northwest Metabolomics Research Center, Department of Anesthesiology and Pain Medicine, University of Washington, 850 Republican St., Seattle, WA 98109, USA;

SM: ⁴Albert Einstein College of Medicine, Bronx, NY, 10461, USA;

HG: ⁵Arizona Metabolomics Laboratory, School of Nutrition and Health Promotion, College of Health Solutions, Arizona State University, Phoenix, AZ 85004, USA

*Primary laboratories of origin.

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Send correspondence to:

Julia Yue Cui, PhD, DABT
Department of Environmental and Occupational Health Sciences
University of Washington
4225 Roosevelt Way NE, Seattle, WA 98105
Email: juliacui@uw.edu
Tel: 206-616-4331

And

Haiwei Gu, PhD
Arizona Metabolomics Laboratory
School of Nutrition and Health Promotion
College of Health Solutions
Arizona State University
Phoenix, AZ 85004
Email: haiweigu@asu.edu

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List of Abbreviations:

3-Indolepropionic acid: 3-IPA; alpha-1,6-mannosyltransferase: Alg12; acetonitrile: ACN; binary alignment map: bam; branched short chain fatty acid: BSCFA; branched chain amino acid: BCAA; conventional: CV; cytochrome P450: Cyp; dopa decarboxylase: Ddc; *de novo* lipogenesis: DNL; endoplasmic reticulum: ER; fragments per kilobase of transcript per million mapped reads: FPKM; false discovery rate: FDR; germ free: GF; Gene Expression Omnibus: GEO; glucokinase: Gck; glutamate-cysteine ligase catalytic subunit: Gclc; glycine N-methyltransferase: Gnmt; large intestinal contents: LIC; Luminal endoplasmic reticulum associated degradation: ERAD-L; multiple reaction monitoring: MRM;

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NAD(P)H dehydrogenase, quinone 1: Nqo1; nuclear factor erythroid 2 like 2: Nrf2; polybrominated diphenyl ether: PBDE; phosphatase and tensin homolog: PTEN; pregnane X receptor: PXR; small intestinal contents: SIC; sequencing alignment map: sam; short chain fatty acid: SCFA; tuberous sclerosis 1: TSC1.

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ABSTRACT

Polybrominated diphenyl ethers (PBDEs) are persistent environmental toxicants associated with increased risk for metabolic syndrome. Intermediary metabolism is influenced by the intestinal microbiome. To test the hypothesis that PBDEs reduce host-beneficial intermediary metabolites in an intestinal microbiome-dependent manner, nine-week old male conventional (CV) and germ-free (GF) C57BL/6 mice orally gavaged once daily with vehicle, BDE-47, or BDE-99 (100 $\mu\text{mol/kg}$) for four-days. Intestinal microbiome (16S rDNA sequencing), liver transcriptome (RNA-Seq), and intermediary metabolites in serum, liver, as well as small and large intestinal contents (SIC and LIC; LC-MS) were examined. Changes in intermediary metabolite abundances in serum, liver, and SIC, were observed under basal conditions (CV versus GF mice) and by PBDE exposure. PBDEs altered the largest number of metabolites in the LIC; most were regulated by PBDEs in GF conditions. Importantly, intestinal microbiome was necessary for PBDE-mediated decreases in branched chain and aromatic amino acid metabolites including 3-indolepropionic acid, a tryptophan metabolite recently shown to be protective against inflammation and diabetes. Gene-metabolite networks revealed a positive association between the hepatic glycan synthesis gene alpha-1,6-mannosyltransferase (Alg12) mRNA and mannose which are important for protein glycosylation. Glycome changes have been observed in patients with metabolic syndrome. In LIC of CV mice, 23 bacterial taxa were regulated by PBDEs. Correlations of certain taxa with distinct serum metabolites

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further highlight a modulatory role of the microbiome in mediating PBDE effects. In summary, PBDEs impact intermediary metabolism in an intestinal microbiome-dependent manner, suggesting that dysbiosis may contribute to PBDE-mediated toxicities including metabolic syndrome.

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INTRODUCTION

Polybrominated diphenyl ethers (PBDEs) have been added to a myriad of consumer products as flame retardants (EU, 2001). While there are 209 predicted PBDE congeners stemming from the degree of bromination and stereochemistry, three commercial formulations were manufactured and used in the US (ATSDR, 2017). One mixture, known as commercial pentaBDE and sold under several trade names, was primarily applied to furniture cushion foam and carpet pads. OctaBDE and DecaBDE mixtures, also sold under different trade names, were primarily applied to plastic components of computers, televisions and other electronics (ATSDR, 2017).

Despite the fact that commercial formulations are no longer produced in, or imported into to the US, PBDEs are ubiquitous in human serum and breast milk samples. Recent data from the Methods Advancement for Milk Analysis (MAMA) Study showed that the median serum concentration of BDE-47 was 18.6 ng/g lipid and 3.9 ng/g lipid for BDE-99 (Marchitti *et al.*, 2017). Median breast milk concentrations for BDE-47 were observed in the same study to be 31.5 ng/g lipid, and 6 ng/g lipid for BDE-99. One route of exposure is from household and work-place dust that is contaminated from PBDE-containing products (Hites, 2004; US-EPA, 2012; ATSDR, 2017). In addition, PBDEs are environmentally persistent and bio-accumulative, facilitating human exposures

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through diet, especially dairy, fish, and meat (Schechter *et al.*, 2004; Schechter *et al.*, 2010; ATSDR, 2017). Two commercial PBDE formulations (e.g. DE-71 and Bromkal 70-5DE) were analyzed for individual congeners and consisted of 36-42% BDE-47 (tetra-brominated), as well as 42-48% BDE-99 (penta-brominated), thus BDE-47 and BDE-99 account for ~80-90% of total PBDEs in these mixtures. (Sanders *et al.*, 2005; La Guardia *et al.*, 2006). BDE-47 and BDE-99 are also two of the most prevalent PBDE congeners found in human samples (Sjödin *et al.*, 2001; Schechter *et al.*, 2003; Imm *et al.*, 2009; Marchitti *et al.*, 2017).

Furthermore, recent epidemiological and animal studies have associations between PBDEs and signs of diabetes and metabolic syndrome. In two independent case-control studies, serum concentrations of BDE-47 in participants with diabetes were significantly associated with increased risk of diabetes (Zhang *et al.*, 2016). In a study among obese individuals, adipose tissue concentrations of both BDE-47 and BDE-99 were significantly elevated in insulin resistant participants compared to insulin sensitive participants (Helaleh *et al.*, 2018). BDE-47 and BDE-99 concentrations were also associated with adipose tissue concentrations of insulin and interleukin-6. BDE-47 was associated with alkaline phosphatase, which is a biomarker for dysfunction in hepatobiliary secretion and has also been shown to interfere with rodent glucose homeostasis (McIntyre *et al.*, 2015; Zhang *et al.*, 2016). Rats exposed to DE-71, one of the commercial PBDE mixtures primarily made up of BDE-47 and BDE-99 as well as other minor constituents, exhibited changes in liver gene expression

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patterns that matched patterns associated with metabolic syndrome (Sanders *et al.*, 2005; Dunnick *et al.*, 2012). However, research into mechanisms for PBDE-mediated changes in intermediary metabolism is ongoing .

The dynamic and diverse populations of microbes that populate the lower gastrointestinal tract, collectively known as the intestinal or gut microbiome, are known to interact with host intermediary and xenobiotic metabolism (Nicholson *et al.*, 2012; Li *et al.*, 2017). We have previously shown that the intestinal microbiome modifies PBDE-mediated effects on bile acids as well as the expression of genes involved in xenobiotic biotransformation (Li *et al.*, 2017; Li *et al.*, 2018). However, little is known regarding PBDE-mediated intermediary metabolic changes in rodents and a potential modulatory role for the intestinal microbiome. Thus, in this study we integrated transcriptomics, targeted metabolomics, and microbial 16S rDNA profiling to evaluate the effects of PBDEs on intermediary metabolism and test our hypothesis that PBDEs down-regulate beneficial intermediary metabolites in an intestinal microbiome-dependent manner.

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MATERIALS AND METHODS

Chemicals: BDE-47 (cat. no.: N-105220-10mg, CAS: 5346-43-1) and BDE-99 (cat. no.: FF-BDE-099N-80MG, CAS: 60348-60-9) were purchased from Chem Service, Inc (West Chester, PA), and Accustandard, Inc. (New Haven, CT), respectively. Phosphate-buffered saline (pH 7.4, cat no.: 10010023) was purchased from Thermo Fisher Scientific (Waltham, MA). De-ionized (DI) water was provided in-house by a Synergy Ultrapure Water System from EMD Millipore (Billerica, MA). The un-labeled metabolite standards were purchased from Sigma-Aldrich (St. Louis, MO) and Fisher Scientific (Pittsburgh, PA) and labeled standards were purchased from Cambridge Isotopes (Tewksbury, MA) (Zhu *et al.*, 2014). Ammonium acetate (cat. no.: A11450, cas no.: 631-61-8; LC-MS grade) and acetic acid (cat. no.: A11350, cas no.: 64-19-7; LC-MS grade) came from Fisher Scientific (Pittsburgh, PA). Acetonitrile (ACN, cat. no.: 1.00029, cas no.: 75-05-8) and methanol (cat. no.: 1.06035, cas no.: 67-56-1) both LC-MS grade), ammonium bicarbonate (cat. no.: 09830, cas no.: 1066-33-7; purity >99.99%), and all other chemicals and reagents, unless indicated otherwise, were purchased from Sigma Aldrich (St. Louis, MO).

Animals: As previously reported (Li *et al.*, 2017; Li *et al.*, 2018) and briefly summarized here, eight-week-old male C57BL/6J CV mice were purchased from the Jackson Laboratory (Bar Harbor, ME). Mice were acclimated at the University of Washington for one week prior to experiments. The initial GF mice, which are

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on a C57BL/6J background, were purchased from the National Gnotobiotic Rodent Resource Center (University of North Carolina, Chapel Hill, NC). Breeding and colony management of GF mice was performed by the University of Washington Gnotobiotic Animal Core. All mice were housed according to the Association for Assessment and Accreditation of Laboratory Animal Care International guidelines. At nine-weeks of age, the mice (n = 5 per group) were exposed to vehicle (corn oil (CO), 10 ml/kg), BDE-47 [48.5mg/kg (100 μ mol/kg)] or BDE-99 [56.5 mg/kg or (100 μ mol/kg)] via oral gavage once daily for four consecutive days. All mice were euthanized using CO₂ narcosis followed by cardiac puncture 24 hr after the final PBDE dose. During the cardiac puncture, blood was collected from the heart with a 25-gauge needle and put into a serum separator tube. Serum was collected and stored at -80 °C after centrifugation for 10 min at 2000 g at 4 °C. Liver tissue was frozen in liquid nitrogen and stored at -80 °C for further analysis. Small (SIC) and large (LIC) intestinal contents were collected as a pellet first by flushing the lumen with 10mM dithiothreitol (DTT) in PBS (Sigma Aldrich, St. Louis, MO) and then centrifuging the luminal contents at 20,000 g for 30 min at 4 °C. All animal studies were approved by the Institutional Animal Care and Use Committee at the University of Washington.

RNA Isolation and sequencing: RNA isolation and sequencing methods were previously reported (Li *et al.*, 2017). Briefly, total RNA was isolated from frozen liver tissue (n=2-4/group) using RNA-Bee (Tel-Test Inc., Friendswood, TX).

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Construction of cDNA libraries and sequencing was performed as previously reported (Li *et al.*, 2017). Libraries were constructed with an Illumina TruSeq Stranded mRNA kit (Illumina, San Diego, CA) and integrity was quantified using an Agilent 2100 Bioanalyzer. Libraries were sequenced using 50 bp paired end reads on an Illumina HiSeq 2000.

16S rDNA sequencing: The sequencing of the LIC microbial DNA from CV mice (n=3/group) was previously reported (Li *et al.*, 2018). Briefly, DNA from the V4 region of the 16S rRNA gene was amplified and 250 bp paired end sequencing was performed on a HiSeq 2500 sequencer.

Serum preparation for aqueous metabolite measurements: Frozen serum samples were first thawed overnight under 4°C, and 50 µL of each serum sample was placed in a 2 mL Eppendorf vial. The initial step for protein precipitation and metabolite extraction was performed by adding 250 µL MeOH. The mixture was then vortexed for 10s and stored at -20°C for 30 min, followed by centrifugation at 14,000 RPM for 10 min at 4 °C. The supernatants (200 µL) were collected into a new Eppendorf vial, and dried under vacuum using an Eppendorf Vacufuge (Eppendorf, Hauppauge, NY). The dried samples were reconstituted in 600 µL of 40% Solvent A/60% Solvent B (see details in the LC-MS method section).

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Tissue preparation for aqueous metabolite quantifications: The sample preparation is the same for liver, as well as small and large intestinal contents. Briefly, each tissue sample (~10 mg) was homogenized in 200 μ L water in an Eppendorf tube using a Bullet Blender homogenizer (Next Advance, Averill Park, NY). Then 800 μ L MeOH was added, and after vortexing for 10 s, the samples were stored on dry ice for 30 min. The samples were then sonicated in an ice bath for 10 min. The samples were centrifuged at 14,000 RPM for 10 min (4 $^{\circ}$ C), and 800 μ L supernatant was transferred to a new Eppendorf tube. The samples were then dried under vacuum using an Eppendorf Vacufuge (Eppendorf, Hauppauge, NY). Prior to MS analysis, the obtained residue was reconstituted in 600 μ L of 40% Solvent A/60% Solvent B (see details in the LC-MS method section).

Measurements of aqueous metabolites using LC-MS: A robust targeted LC-MS/MS method has been developed (Zhu *et al.*, 2014) and used in a growing number of studies (Barton *et al.*, 2015; Carroll *et al.*, 2015; Gu *et al.*, 2015; Reyes *et al.*, 2015; Sood *et al.*, 2015; Sperber *et al.*, 2015; Zhu *et al.*, 2015; Deng *et al.*, 2016; Gu *et al.*, 2016). Briefly, the LC-MS/MS experiments were performed on a Waters Acquity I-Class UPLC TQS-micro MS (Waters, Milford, MA) system. Each prepared serum or tissue sample was injected twice, 2 μ L and 5 μ L for analysis using positive and negative ionization mode, respectively. Chromatographic separation was performed on a Waters Xbridge BEH Amide

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column (2.5 μm , 2.1 x 150 mm) at 40 °C. The flow rate was 0.3 mL/min. For positive mode, the mobile phase was composed of solvent A (5 mM ammonium acetate in H₂O with 0.1% formic acid) and solvent B (ACN with 0.1% acetic acid). For negative mode, Solvent A was 10 mM ammonium bicarbonate in H₂O, and Solvent B was ACN. The LC gradient conditions were the same for both positive and negative ionization modes. After an initial 1.5 min isocratic elution of 10% A, the percentage of Solvent A was increased to 65% at t=9 min. Then the percentage of A was kept the same (65%) for 5 min (t=14 min), and the percentage of A was decreased back to 10% at t=15 min to prepare for the next injection. The total experimental time for each injection was 30 min. The metabolite identities were confirmed by spiking mixtures of standard compounds. The extracted peaks were integrated using the TargetLynx software (Waters, Milford, MA). A list of all metabolites on the panel can be found in Supplemental Table 1. Supplemental Tables 2-5 show mean abundance values and standard errors for all metabolites by exposure group that were detectable in each bio-compartment.

Data Analysis

RNA-Seq: Preliminary analyses of the FASTQ files used in this study, which were deposited by our group in NCBI's Gene Expression Omnibus (GEO) (GEO accession number GSE101650) during publication of a previous manuscript, have been previously reported (Li et al., 2017). Abundance was expressed as

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fragments per kilobase of transcript per million mapped reads (FPKM). Differential expression analysis was performed using Cuffdiff (Trapnell *et al.*, 2010). The differentially expressed genes were defined as false discovery rate (FDR) adjusted p -value <0.05 , in at least one of the PBDE-exposed groups compared to the vehicle-exposed group of the same enterotype, which in this study refers to CV as opposed to GF mice, as well as genes that were differentially expressed by the lack of intestinal microbiome under basal conditions. Assignment of genes into intermediary metabolism categories was performed using KEGG metabolism pathways in R 3.4.3 (Kanehisa and Goto, 2000; Kanehisa *et al.*, 2016; Kanehisa *et al.*, 2017; R Core Team, 2017). Mean FPKM standard errors and metabolism categories for intermediary metabolism genes for each treatment group can be found in Supplemental Table 6. Fold changes and associated FDR adjusted p -values for comparisons between PBDE treated mice and enterotype-matched vehicle controls and between CV and GF mice under basal conditions can be found in Supplemental Table 7. Heatmaps were also built in R 3.4.3 (base, gplots package) (Warnes *et al.*, 2013; R Core Team, 2017). Because many gene products have multiple functions, many of the differentially expressed genes classified as intermediary metabolism genes here were reported as drug processing genes previously, mainly cytochrome P450s (Cyps) by Li *et al.* (Li *et al.*, 2017).

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Aqueous Metabolites: Relative abundance values for each metabolite in each bio-compartment were compared using two-way ANOVA followed by Tukey's post-hoc test using R 3.4.3 (base, lsmmeans and multcomp packages) (Hothorn *et al.*, 2008; Lenth, 2016; R Core Team, 2017). Heatmaps were also built in R (base, gplots package) (Warnes *et al.*, 2013; R Core Team, 2017). Gene-Metabolite network analyses were performed using MetaboAnalyst (Chong *et al.*, 2018) (<http://www.metaboanalyst.ca>) using all differentially regulated intermediary metabolism genes with a fold-change > 2 (implemented to emphasize most highly changed genes) and significantly altered metabolites in the liver. Pearson's correlations for gene and metabolite network members were calculated using R 3.4.3 (base, corrplot, and Hmisc packages) (Harrell *et al.*, 2016; Wei and Simko, 2016; R Core Team, 2017). Significant gene-metabolite pair correlation plots and bar plots were created using R3.4.3 (base, ggplot2 package) (Wickham, 2009; R Core Team, 2017).

16S rDNA sequencing: The 16S rDNA sequencing data were analyzed using QIIME v1.9.1 as previously reported (Li *et al.*, 2018). Relative abundance was expressed as percentage of operational taxonomical units (% OTUs) and was examined for PBDE treatment effects using a one-way ANOVA followed by Tukey's HSD post-hoc test in R 3.4.3 ($p < 0.05$) (R Core Team, 2017). In this study, these data were used for investigating potential associations between individual microbial taxa and serum aqueous metabolites. Spearman's

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correlations for differentially regulated microbial OTUs and serum metabolites by PBDEs were performed using R (base, corrplot package) (Wei and Simko, 2016; R Core Team, 2017). Significant gene-metabolite pair correlation plots and bar plots were created using R (base, ggplot2 package) (Wickham, 2009).

Glutathione: Levels of total glutathione in liver were quantified using a fluorescence-based microplate assay, as previously described (White *et al.*, 2003; McConnachie *et al.*, 2007; Weldy *et al.*, 2011; Scoville *et al.*, 2019)

RT-qPCR: Expression of *Alg12*, *Gclc*, and *Nqo1* were measured using RT-qPCR. Total RNA was reverse transcribed to cDNA using the High Capacity cDNA Reverse Transcription Kit (Applied Biosystems, Foster City, CA). The cDNAs were then amplified using Power SYBR™ Green PCR Master Mix (Applied Biosystems, Foster City, CA) using a BioRad CFX384 Real-TimePCR Detection System (Bio-Rad, Hercules, CA). Primers were made by Integrated DNA Technologies (Coralville, IA) and primer sequences can be found in Supplemental Table 8.

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RESULTS

Effects of PBDEs on aqueous metabolites in serum of CV and GF mice

In order to determine the impact of PBDE exposure on the dynamic systems of intermediary metabolism, aqueous metabolites from several bio-compartments were quantified using LC-MS. Of the total 215 aqueous metabolites on the panel a total of 139 were detectable in serum (Supplemental Table 2). As shown in Figure 1, the relative abundances of 13 metabolites in serum were decreased and 2 increased by lack of intestinal microbiome, PBDE exposure, or both, compared to CV control mice.

Six metabolites were decreased in the serum of GF mice under basal conditions including the tryptophan metabolites 3-indoxylsulfate, 3-indolepropionic acid (3-IPA), the phenylalanine metabolites phenylpyruvic acid and 3-phenyllactic acid, the branched short chain fatty acid (BSCFA) isobutyric acid, and the fatty acid amide *N*-acetyethanolamine. Acetylglycine and the saturated long-chain fatty acid palmitic acid were increased in serum under basal conditions in GF mice.

PBDEs differentially regulated 12 serum metabolites. Interestingly, the relative abundances of 3 tryptophan metabolites, namely the microbial metabolites 3-IPA and indole-3-acetic acid, as well as the host metabolite kynurenine, were all decreased by both PBDE congeners, in CV but not GF mice. PBDEs also decreased *N*-acetyethanolamine and phenyllactic acid in CV

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but not GF mice. BDE-47 decreased the relative abundances of the amino acid valine and its non-proteogenic isomer norvaline whereas BDE-99 increased phenylpyruvic acid, isobutyric acid, and the carbohydrate metabolite sorbitol, also in CV but not GF mice. In contrast, the lack of gut microbiota predisposed GF mice to PBDE-mediated decreases in the relative abundance of metabolites derived from the branched chain amino acid (BCAA) valine. BDE-47 increased alpha-ketoisovaleric whereas both BDE-47 and BDE-99 mediated increased alpha-hydroxyisovaleric acid.

Effects of PBDEs on aqueous metabolites in liver of CV and GF mice

In liver, 156 aqueous metabolites were detectable (Supplemental Table 3). As shown in Figure 2, under basal conditions, 11 out of the 16 metabolite changes occurred between CV and GF mice. The relative abundances of the sialic acid *N*-acetylneuraminic acid, the purine metabolite urate, and the amino acid glycine were all increased in GF mice. The relative abundances of isobutyric acid, 3-phenyllactic acid, the amino acid derivative carnitine, the amino acid derivative ketoleucine, the carbohydrate metabolite glucuronic acid, and diet-derived metabolites (benzoic acid, phthalic acid, 4-methoxyphenylacetic acid) were all decreased in GF mice.

In CV mice, BDE-47 had minimal effect on the hepatic aqueous metabolite profiles. However, BDE-99 decreased relative abundances of carnitine, the carbohydrate mannose, the antioxidant glutathione, the homocysteine metabolite

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cystathionine, and the vitamin nicotinamide, all in CV but not GF mice. The lack of gut microbiota predisposed GF mice to BDE-47-mediated decreases in the glutamine derivative acetyl-L-glutamine.

Effects of PBDEs on aqueous metabolites in SIC of CV and GF mice

A total of 147 aqueous metabolites were detectable in the SIC (Supplemental Table 4). As shown in Figure 3, there were 15 metabolites that were decreased in SIC of GF mice under basal conditions, including several amino acids (phenylalanine, tryptophan, arginine, and cysteine, citrulline, asparagine), amino acid derivatives (carnitine, acetyl-L-glutamine, creatine, acetylmethionine, and 4-imidazoleacetic acid); the carbohydrate metabolite sorbitol, as well as other organic compounds (folinic acid, and 4-aminophenol). Conversely, the relative abundance of dextrose (also known as D-glucose) was increased in the SIC of GF mice under basal conditions.

PBDE exposure differentially regulated six metabolites in the SIC of CV mice but not GF mice. Asparagine, creatine, sorbitol, and cysteine were decreased by both PBDE congeners, folinic acid was decreased by BDE-47, and acetyl-L-glutamine was decreased by BDE-99. In contrast, the lack of gut microbiota predisposed GF mice to BDE-47 mediated increases in the relative abundances of amino acid derivatives (3-phenyllactic acid, acetyl-L-tyrosine), the redox active coenzyme NAD, and the glycerol derivative glyceric acid; BDE-99

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mediated increase in carnitine; and BDE-99 mediated decrease in 4-hydroxybenzoic acid.

Effects of PBDEs on aqueous metabolites in LIC of CV and GF mice

A total of 147 aqueous metabolites were detectable in the SIC (Supplemental Table 5). As shown in Figure 4, PBDEs differentially regulated 26 metabolites in the LIC—the highest among the four bio-compartments examined. Interestingly, the relative abundances for the majority of metabolites (24 out of 26) in the LIC were changed by PBDEs only in GF conditions but not in CV conditions. The two exceptions are the tryptophan metabolite indole-3-pyruvic acid, which was reduced in the LIC of CV mice by BDE-47 (the basal relative abundance in GF mice was also lower), and folinic acid, which was increased by BDE-99 in CV but not GF mice.

The lack of gut microbiome predisposed GF mice to PBDE-mediated increases in 17 LIC metabolites. As compared to vehicle-exposed GF mice, both BDE-47 and BDE-99 increased the relative abundances of the amino acids leucine and methionine, amino acid derivatives (4-hydroxyproline, 5-aminoleulinic acid, and acetylcarnitine), the choline metabolism intermediate and methionine precursor betaine, and the coenzyme A precursor pantothenic acid. BDE-47 increased relative abundances of the nucleic acid 2-deoxycytidine and amino cysteine in LIC of GF mice, whereas BDE-99 increased several amino acids (norleucine, isoleucine, L-alloisoleucine, tyrosine, phenylalanine, and

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tryptophan), the tryptophan metabolite indole-3-lactic acid and the carbohydrate metabolite glycerol-3-phosphate.

The lack of gut microbiota also predisposed GF mice to PBDE-mediated decreases in seven metabolites. Both BDE-47 and BDE-99 decreased the relative abundances of carbohydrate metabolites sorbitol and raffinose in the LIC of GF mice as compared to control GF mice. In addition, BDE-47 decreased relative abundances of the nucleic acid adenine and carbohydrate metabolite pyruvate, whereas BDE-99 decreased the NAD precursor nicotinic acid, the histamine metabolite 4-imidazoleacetic acid, and the fatty acid amide *N*-acetyethanolamine in the LIC of GF mice.

Differentially abundant metabolites unique and common to different bio-compartments are summarized in Supplemental Figure 4. Treatment group specific and common differentially abundant metabolites within each bio-compartment are summarized in Supplemental Figure 5.

Liver intermediary metabolism gene expression and gene-metabolite networks

As shown in Figures 5 and 6, 133 intermediary metabolism related genes belonging to one or more of the KEGG pathways (carbohydrate, glycan, amino acid, vitamin and co-factor, nucleotide, and energy metabolism) were differentially expressed due to either PBDE exposure, lack of gut microbiota, or both in liver. Specifically, carbohydrate metabolism and glycan biosynthesis and

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metabolism were represented by 38 and 13 differentially expressed genes, respectively (Figure 5A). Amino acid metabolism was represented by 70 differentially expressed genes (Figure 6A) and vitamin and co-factor metabolism contained 44 genes (Figure 6C). Nucleotide metabolism and energy metabolism contained 11 and 10 differentially expressed genes, respectively (Supplemental Figure 1). Metabolism category labels for genes in Figure 5A, Figure 6A, Figure 6C, and Supplemental 1 can be found in Supplemental Table 6.

To integrate the liver intermediary metabolism transcriptomic and metabolomic data and identify potential gene-metabolite regulatory networks impacted by PBDEs, network analysis was performed using Metaboanalyst, with all differentially regulated intermediary metabolism related genes and aqueous metabolites in liver. As Metaboanalyst was built for human genes, Ensembl Biomart was used to retrieve human homologs.

The Network Explorer module revealed three networks using the list of differentially regulated liver genes and metabolites. One of the networks generated by Metaboanalyst centered on the carbohydrate metabolite mannose with connections (representing interactions from literature found by Network Explorer) to the carbohydrate metabolism gene glucokinase (GCK), the glycan synthesis gene alpha-1,6-mannosyltransferase (ALG12), and the carbohydrate mannose (Figure 5B). In addition, a significant correlation was observed in our data between ALG12 expression and the relative abundance of mannose (Figures 5C), and BDE-99 decreased both in CV but not GF mice (Figure 5D).

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Expression of Alg12 measured using RT-qPCR matched RNA-seq expression patterns across treatment groups (Supplemental Figure 2).

The second gene-metabolite network that was generated by Metaboanalyst centered around glutathione (Figure 6B), which is synthesized from three amino acids (glycine, glutamate, and cysteine). Glutathione was connected to the amino acid metabolism genes dopa decarboxylase (DDC), cytochrome P450 1A2 (CYP1A2 representing the mouse Cyp1a2), CYP3A5 (representing several mouse Cyp3a isomers), and glutamate-cysteine ligase catalytic subunit (GCLC representing the mouse Gclc), which is a glutathione synthesis gene utilizing glutamate and cysteine as substrates (Figure 6B). Expression of Cyp1a2 and Cyp3a11, which was the most highly expressed Cyp3a isoform, have been validated using RT-qPCR as previously reported (Li *et al.*, 2017). NAD(P)H dehydrogenase, quinone 1 (NQO1), which is classified as a vitamin and cofactor metabolism gene as well as a prototypical target gene of the anti-oxidative stress sensor nuclear factor erythroid 2 like 2 (Nrf2), is also connected to glutathione (Figures 6B and 6C) (Ross and Siegel, 2018). Levels of liver glutathione were also measured independently using a fluorescence-based microplate assay. Interestingly, the pattern across exposure groups was similar to that found using the LC-MS based aqueous metabolite panel, suggesting that although the transcriptomic changes in glutathione synthesis/metabolism related genes occur following PBDE exposure and the absence of gut microbiome, the liver serves as an efficient repertoire to buffer the oxidative stress by maintaining

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the total glutathione levels (Supplemental Figures 3A and 3B). To validate the RNA-Seq results, RT-qPCR was performed for the mRNA expression of *Gclc* and *Nqo1*, and the expression patterns were consistent among the two assays across exposure groups (Supplemental Figures 3C, 3D, 3E and 3F).

The third network that was generated by was centered on the vitamin nicotinamide, which was connected to vitamin and cofactor processing genes *CYP2D6* and *CYP3A5*, which are represented by several mouse *Cyp2d* and *Cyp3a* isoforms, respectively (Figure 6D).

Correlations between serum metabolites and gut microbiota

To investigate the intersection of PBDE regulation of serum metabolites and the gut microbiome at the individual taxa level, the relative abundances of 23 gut microbial taxa that were changed by PBDEs were tested for correlation with PBDE-regulated serum metabolites. Serum metabolites were prioritized because the changes in this bio-compartment could have systemic effects. Of the 23 taxa, 19 were significantly correlated with at least one serum aqueous metabolite (Figure 8).

Regarding amino acid metabolism, the microbial tryptophan metabolite 3-IPA was positively correlated with the largest number of taxa (8), all of which belong to the phylum Firmicutes, and apart from one taxon from the Cloistridia class, all belong to the class Bacilli (Figure 8). Four of the Bacilli belong to the Bacillales order and three are Lactobacillales. Within the Bacillales order, two

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taxa were in the *Staphylococcaceae* family, one is a *Bacillaceae*, and the other is unresolved at the family level and beyond. Both *Staphylococcaceae* are *Staphylococcus* at the genus level, but neither is resolved at the species level. The taxon from the *Bacillaceae* family is *Bacillus* at the genus level and also unresolved at the species level. Within the Lactobacillales order, two are in the *Lactobacillaceae* family, and the other taxon is unresolved at the family level and beyond. Both *Lactobacillaceae* are *Lactobacillus* at the genus level. One is the *reuteri* species and the other is unresolved at the species level. The taxon in the Clostridia class is Clostridiales (order), *Lachnospiraceae* (family), and unresolved at the genus and species levels.

Another microbial tryptophan metabolite, namely indole-3-acetic acid, is positively correlated with 2 microbial taxa and inversely correlated with one (Figure 8). The first positively correlated taxon is Firmicutes (phylum), Clostridia (class), Clostridiales (order), *Lachnospiraceae* (family), *Butyrivibrio* (genus), and unresolved at the species level. The second positively correlated taxon is Firmicutes (phylum), Erysipelotrichi (class), Erysipelotrichales (order), *Erysipelotrichaceae* (family), and unresolved at the genus and species level. The inversely correlated taxon has the same taxonomy as the *Erysipelotrichaceae* (family), belongs to the *Allobaculum* genus, and is unresolved at the species level.

The phenylalanine metabolite phenyllactic acid is positively correlated with seven gut microbial taxa (Figure 7). Two of the seven taxa are the same

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Staphylococcus genus as described above. An additional two are the same *Lactobacillus* genus taxa, one identified as *reuteri*, and the other unresolved at the species level. The fifth taxon associated with phenyllactic acid is Bacillales at the order level and is also previously described above. One of the remaining taxa associated with phenyllactic acid is Actinobacteria (phylum), Coriobacteriia (class), Coriobacteriales (order), *Coriobacteriaceae* (family), *Adlercreutzia* (genus), and unresolved at the species level. The other taxon is Firmicutes (phylum), Clostridia (class), Clostridiales (order), *Mogibacteriaceae* (family), and unresolved at the genus and species levels.

BCAAs valine and norvaline, are positively correlated with the same taxon from the *Butyrivibrio* genus as indole-3-acetic acid (Figure 7). They are both also inversely correlated with the same taxon from the *Allobaculum* genus as indole-3-acetic acid (Figure 7).

The fatty acid amide *N*-acetyethanolamine is positively correlated with 4 microbial taxa (Figure 7). The full taxonomy of three have already been described; *Adlercreutzia* (genus), *Lactobacillus* (genus – unresolved at species level), and *Mogibacteriaceae* (family). The 4th taxa associated with *N*-acetyethanolamine has the same taxonomy as the *Lactobacillus* (genus), and also unresolved at the species level, but is distinct from the *Lactobacillus* (genus) associated with 3-IPA and 3-phenyllactic acid.

Three metabolites, namely the branched short chain fatty acid isobutyric acid, the phenylalanine metabolite and phenyllactic acid precursor phenylpyruvic

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acid, and the carbohydrate sorbitol, are all positively correlated with a microbial taxon that is Firmicutes (phylum), Clostridia (class), Clostridiales (order), *Lachnospiraceae* (family), *Dorea* (genus), and unresolved at the species level (Figure 7). These same three metabolites are also inversely associated with *Akkermansia* (genus) *mucinophila* (species), which belongs to Verrucomicrobia phylum, Verrucomicrobiae class, Verrucomicrobiales (order), and *Verrucomicrobiaceae* family.

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DISCUSSION

Although the commercial pentaBDE (comprised mainly of BDE-47 and BDE-99) and octaBDE formulations of PBDEs were voluntarily taken off the market in 2004 and decaBDE by 2014 (ATSDR, 2017), PBDEs are still readily detectable in human samples. While concentrations of pentaBDE constituents have previously been observed to decline since their withdrawal, the trend may have flattened out and concentrations may now be on the rise (Ma *et al.*, 2013; Zota *et al.*, 2013; Hurley *et al.*, 2017; Parry *et al.*, 2018; Cowell *et al.*, 2019). This is likely due to the persistent and bio-accumulative nature of PBDEs, the recycling of PBDE-containing products worldwide, and the potential for higher brominated PBDES to degrade in the environment into lower brominated forms (Bezares-Cruz *et al.*, 2004; Zeng *et al.*, 2008).

As originally reported in Li *et al.*, 2017, male mice were dosed once daily for four consecutive days with 48.5mg/kg BDE-47 and 56.5 mg/kg BDE-99 (100 μ mol/kg). While these doses are high compared to estimated human PBDE exposures, they were selected to allow for comparison of acute xenobiotic metabolism gene expression changes with previously published studies (Pacyniak *et al.*, 2007; Sueyoshi *et al.*, 2014; Li *et al.*, 2017). In this study, metabolomic data from archived samples collected by Li *et al.* was integrated with previously reported liver gene expression (Li *et al.*, 2017) and intestinal microbiome data (Li *et al.*, 2018) to explore acute PBDE effects on intermediary metabolism.

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Limitations of this study include sample size, the evaluation of only male mice, and that is mainly associative, all of which stem from the use of archived samples and previously reported datasets. As reported in Li *et al.*, 2017, male mice were used exclusively due to changes in hormone levels due to the estrous cycle. Such hormonal variation is also relevant to this study as it could independently impact glucose metabolism and other intermediary metabolic pathways (Mauvais-Jarvis *et al.*, 2013). Sample size for the three data sets used in this study was also a limitation. However, even with relatively small sample sizes, we were able to identify statistically significant PBDE and gut-microbiome associated changes in intermediary metabolites, expression of associated genes, and correlations among metabolites and several previously reported human health and disease relevant microbes (Li *et al.*, 2018). Importantly, future mechanistic studies will be needed to fully validate the observed findings of this study.

Tryptophan pathway and involvement of PBDEs in metabolic syndrome

Notably, serum 3-IPA, which is a tryptophan microbial metabolite known to be protective against diabetes and inflammation, was decreased by both PBDE congeners in CV mice but not in GF mice (Figure 1). This highlights the potential involvement of the tryptophan metabolism pathway in PBDE-mediated metabolic disorders because 3-IPA is produced by intestinal bacteria and accumulates in the blood to modulate systemic effects (Dodd *et al.*, 2017) such as metabolic

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syndrome. Specifically, 3-IPA is associated with reduced risk of type-II diabetes in epidemiology studies and is suggested to preserve pancreatic beta cell function (de Mello *et al.*, 2017). These observations in humans were further supported in rats because 3-IPA improved glucose metabolism evidenced by reduced fasting plasma insulin and the homeostatic model assessment index of insulin resistance (Abildgaard *et al.*, 2018). In humans, BDE-47 was associated with diabetes in two independent community-based epidemiological studies (Zhang *et al.*, 2016), and total PBDEs were linked to increased risk of diabetes during pregnancy (Eslami *et al.*, 2016; Smarr *et al.*, 2016). PBDE-exposed rats developed hyperglycemia and aberrant expression of genes involved in glucose homeostasis (Zhang *et al.*, 2016; Krumm *et al.*, 2018), corresponding to disturbed glucose and insulin metabolism in adipose tissue, disturbed lipid metabolism in liver, and increased ketone concentrations in blood (Cowens *et al.*, 2015). PBDEs disturbed glucose signaling in hepatocytes (Søfteland *et al.*, 2011). *In vitro* studies showed that PBDEs disrupted insulin-producing pancreatic beta cells (Karandrea *et al.*, 2017). Therefore, PBDE-mediated decrease in 3-IPA may be part of the toxification mechanisms for the pathogenesis of diabetes by disrupting glucose and insulin signaling.

Another potential involvement of 3-IPA in metabolic syndrome is inflammation—an important contributor to diabetes (Donath and Shoelson, 2011; Goldfine *et al.*, 2011)—and 3-IPA is known to suppress inflammatory genes by antagonizing aryl hydrocarbon receptor functions (Yisireyili *et al.*, 2017). In

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addition, 3-IPA is a pregnane X receptor (PXR) ligand in intestine and activation of PXR reduces intestinal inflammation (Cheng *et al.*, 2012; Venkatesh *et al.*, 2014). Conversely, PBDEs have been shown to affect the immune system during development (Zhou *et al.*, 2006; Martin *et al.*, 2009; Bondy *et al.*, 2011; Liu *et al.*, 2012; Koike *et al.*, 2014; Lv *et al.*, 2015), as well as promote inflammation and oxidative stress (Zhang *et al.*, 2013), which are key contributors for the pathogenesis of diabetes (Farah *et al.*, 2008; Vikram *et al.*, 2014; Domingueti *et al.*, 2016; Hojs *et al.*, 2016). Therefore, PBDE-mediated decrease in IPA may also contribute to the pathogenesis of diabetes by promoting inflammation.

Branched chain amino acid metabolites, PBDEs, and adipose tissue metabolism

Isobutyric acid is a branched short-chain fatty acid (BSCFA) derived from the BCAA valine and can be produced by gut bacteria or by host (Liebich and Först, 1984; Zarling and Ruchim, 1987; Cole, 2015). Shown to potentially benefit adipocyte metabolism, isobutyric acid was decreased in this study in GF mice under basal conditions and by BDE-99 in CV mice (Heimann *et al.*, 2016). While there is controversy as to a potentially beneficial or detrimental role for BCAAs in obesity and type 2 diabetes, preliminary evidence suggests a potential protective role for their metabolites (Lynch and Adams, 2014; Heimann *et al.*, 2016).

In obesity, adipocytes exhibit inter-related changes including increased basal rates of lipolysis and insulin resistance, which are thought to contribute to

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systemic insulin resistance (Girousse *et al.*, 2013; Arner and Langin, 2014; Morigny *et al.*, 2016). Treatment of rat adipocytes with isobutyric acid reduced lipolysis and improved basal and insulin-induced glucose uptake (Heimann *et al.*, 2016). Non-branched short chain fatty acids (SCFAs) have also been observed to have similar effects on rat adipocytes (Heimann *et al.*, 2015). In contrast, PBDEs have been associated with increased rat adipocyte lipolysis and decreased glucose oxidation (marker of uptake and metabolism) (Hoppe and Carey, 2012). Furthermore, PBDEs have been associated with insulin resistance among obese humans (Helaleh *et al.*, 2018).

The decreases of the potentially beneficial valine metabolite isobutyric acid and, among GF mice, its upstream metabolic intermediates alpha-ketoisovaleric and alpha-hydroxyisovaleric acid, suggests that PBDEs may promote metabolic syndrome in part by interference with valine/BCAA metabolism.

PBDEs, protein glycosylation, and metabolic syndrome

Liver intermediary metabolism gene expression and aqueous metabolite data were integrated using the Network Explorer module in Metaboanalyst (Chong *et al.*, 2018). One of the resulting networks, consisting of the genes ALG12, GCK, and the carbohydrate mannose, stood out due to a correlation found in the study data between ALG12 and mannose. As part of the *N*-glycosylation pathway, ALG12 is responsible for the addition of the 8th mannose

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residue to a growing oligosaccharide that ultimately possess 9 mannoses and 3 glucoses, all of which are added in the endoplasmic reticulum (ER) lumen (Burda and Aebi, 1999; Thiel et al., 2002; Aebi, 2013). As an *N*-glycan, this molecule is subsequently attached to the nitrogen of the side chain of asparagine in a protein by oligosaccharyltransferase (Burda and Aebi, 1999; Aebi, 2013). *N*-glycans on proteins participate in the luminal endoplasmic reticulum associated degradation pathway (ERAD-L) that detects and degrades misfolded proteins (Quan et al., 2008).

Epidemiological studies have found that patients diagnosed with metabolic syndrome have an altered *N*-glycan profile with some components, including those with 7 and 8 mannose residues, being positively associated with signs of metabolic syndrome and some inversely (Lu et al., 2011; McLachlan et al., 2016). In this study, BDE-99 reduced Alg12 expression and the relative abundance of mannose in CV but not GF mice, suggesting that a PBDE microbial metabolite could also contribute to these changes. To our knowledge, the potential for PBDEs and/or PBDE host/microbial metabolites to interfere with protein glycosylation has not been explored. However, future glycomics studies will be necessary to evaluate this potential novel mechanism of PBDE toxicity.

Microbiota correlations with serum metabolites

The present study showed that PBDE mediated decreases in *Lactobacillus reuteri* correlate with serum decreases in the tryptophan metabolite

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3-IPA. Even though *L. reuteri* is not known to produce 3-IPA, it does produce indole-3-lactic acid, which is a 3-IPA precursor (Dodd *et al.*, 2017). PBDEs also reduced serum indole-3-acetic acid, which is another tryptophan metabolite produced by *L. reuteri* (Cervantes-Barragan *et al.*, 2017). Interestingly, in mouse models where *L. reuteri* is introduced in combination with tryptophan supplementation, indole-3-acetic and indole-3-lactic acid have been shown to promote the development of intestinal T-cells that help immune tolerance to antigens in the diet (Cervantes-Barragan *et al.*, 2017). However, given the small sample size, microbe-metabolite associations should be viewed as preliminary until follow-up studies with larger numbers are completed.

Summary

Taken together, the present study demonstrates that oral PBDE exposure profoundly altered many important intermediary metabolites involved in amino acid, carbohydrate, and lipid metabolism, in a bio-compartment specific manner. While liver metabolites were more influenced by enterotype than PBDEs, metabolites from large intestinal contents were more influenced by PBDEs than by enterotype and such influence occurred almost exclusively in GF conditions. Metabolites from serum and SIC were regulated by both enterotype and PBDEs. Two interesting regulatory patterns were discovered: 1) the presence of gut microbiota is necessary for PBDE-mediated decreases in metabolites, such as those from the tryptophan pathway in serum and those from the metabolism of

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amino acids and carbohydrates in the SIC; 2) lack of gut microbiota predisposed mice to PBDE-mediated changes metabolites including the majority of those from amino acid and carbohydrate metabolism in the LIC. Correlation analyses among targeted metabolomics, hepatic RNA-Seq, and intestinal 16S rDNA sequencing further established potential mechanisms for PBDE-mediated regulation of intermediary metabolites. Together our observations highlight the potential involvement of dys-regulation of intermediary metabolism as part of the mechanisms underlying the multi-organ toxicities of PBDEs and unveils the important role of gut microbiota as a key internal modifier for PBDE-mediated host response. Future follow-up studies will include mechanistic investigation using male and female wild-type and nuclear receptor knockout mice to assess the impact of key metabolites, identified in this study on PBDE toxicity, dysbiosis, and potential interactions with nuclear receptors. Inflammation, glucose homeostasis, and the effects of high fat diet will also be explored.

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AUTHORSHIP CONTRIBUTIONS

Participated in research design: Li, Cui

Conducted experiments: Li, Dempsey, Wang, Gu, Cui

Contributed new reagents or analytic tools: Raftery

Performed data analysis: Scoville, Li, Wang, Gu, Cui

Wrote or contribute to the writing of the manuscript: Scoville, Li, Dempsey, Wang, Raftery, Mani, Gu, Cui

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FOOTNOTES:

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FIGURE LEGENDS

Figure 1. Two-way hierarchical clustering dendrogram of aqueous metabolites measured using LC-MS that were differentially regulated across treatment groups in serum of GF and CV mice following exposures to BDE-47 and BDE-99. Euclidian distance and complete linkage were used to generate the dendrogram. Red indicates higher standardized mean metabolite relative abundances (z-scores) and blue lower. Asterisks (*) represent statistically significant differences between corn oil-treated and PBDE treated groups within enterotype (adjusted p-value <0.05, Tukey's HSD post-hoc test). Caret signs (^) represent statistically significant baseline differences between CV and GF mice. Percent change from CVCO are shown for treatment groups with significant changes.

Figure 2. Two-way hierarchical clustering dendrogram of aqueous metabolites measured using LC-MS that were differentially regulated across treatment groups in liver of GF and CV mice following exposures to BDE-47 and BDE-99. Euclidian distance and complete linkage were used to generate the dendrogram. Red indicates higher standardized treatment group mean metabolite relative abundances (z-scores) and blue lower. Asterisks (*) represent statistically significant differences between corn oil-treated and PBDE treated groups within enterotype (adjusted p-value <0.05, Tukey's HSD post-hoc test). Caret signs (^) represent statistically significant baseline differences between CV and GF mice.

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Percent change from CVCO are shown for treatment groups with significant changes.

Figure 3. Two-way hierarchical clustering dendrogram of aqueous metabolites measured using LC-MS that were differentially regulated across treatment groups in SIC of GF and CV mice following exposures to BDE-47 and BDE-99. Euclidian distance and complete linkage were used to generate the dendrogram. Red indicates higher standardized treatment group mean metabolite relative abundances (z-scores) and blue lower. Asterisks (*) represent statistically significant differences between corn oil-treated and PBDE treated groups within enterotype (adjusted p-value <0.05, Tukey's HSD post-hoc test). Caret signs (^) represent statistically significant baseline differences between CV and GF mice. Percent change from CVCO are shown for treatment groups with significant changes.

Figure 4. Two-way hierarchical clustering dendrogram of aqueous metabolites measured using LC-MS that were differentially regulated across treatment groups in LIC of GF and CV mice following exposures to BDE-47 and BDE-99. Euclidian distance and complete linkage were used to generate the dendrogram. Red indicates higher standardized treatment group mean metabolite relative abundances (z-scores) and blue lower. Asterisks (*) represent statistically significant differences between corn oil-treated and PBDE treated groups within

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enterotype (adjusted p-value <0.05, Tukey's HSD post-hoc test). Caret signs (^) represent statistically significant baseline differences between CV and GF mice. Percent change from CVCO are shown for treatment groups with significant changes.

Figure 5. Two-way hierarchical clustering dendrogram of carbohydrate and glycan metabolism genes that were differentially regulated across treatment groups in liver of GF and CV mice following exposures to BDE-47 and BDE-99 (A). Gene expression was quantified using RNA-Seq as previously reported (Li *et al.*, 2017) and can be accessed through NCBI GEO (GSE101650). Euclidian distance and complete linkage were used to generate the dendrogram. Red indicates higher standardized mean metabolite relative abundances and blue lower. Asterisks (*) represent statistically significant differences between corn oil-treated and PBDE treated groups (FDR adjusted p-value <0.05). Caret signs (^) represent statistically significant baseline differences between CV and GF mice. Network of differentially regulated carbohydrate (GCK) and glycan (ALG12) metabolism genes (circles) and the significantly altered carbohydrate metabolite Mannose (square), which was quantified using LC-MS, in liver (B). Mouse orthologs for human genes are listed in mixed case. Pearson correlation of log₂ abundances of Mannose and Alg2 across treatment group means (C). Means and standard errors of Alg12 and Mannose for each treatment group (D). Asterisks (*) represent statistically significant differences between corn oil-treated

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and PBDE treated groups (FDR adjusted p-value <0.05, Cuffdiff). Caret signs (^) represent statistically significant baseline differences between CV and GF mice.

Figure 6. Two-way hierarchical clustering dendrogram of amino acid metabolism genes that were differentially regulated across treatment groups in liver of GF and CV mice following exposures to BDE-47 and BDE-99 (A). Gene expression was quantified using RNA-Seq as previously reported (Li *et al.*, 2017) and can be accessed through NCBI GEO (GSE101650). Euclidian distance and complete linkage were used to generate the dendrogram. Red indicates higher standardized mean metabolite relative abundances and blue lower. Network of differentially regulated amino acid metabolism genes (DDC, NQO1, GNMT, CYP1A2, and CYP3A5) (circles) and significantly altered aqueous metabolites (Glycine and Glutathione) (squares), which were measured using LC-MS, in liver (B). Mouse orthologs for human genes are listed in mixed case. Two-way hierarchical clustering dendrogram of vitamin and co-factor metabolism genes that were significantly altered across treatment groups in liver of GF and CV mice following exposures to BDE-47 and BDE-99 (C). Gene expression was measured using RNA-Seq as previously reported (Li *et al.*, 2017) and can be accessed through NCBI GEO (GSE101650). Euclidian distance and complete linkage were used to generate the dendrogram. Red indicates higher standardized mean metabolite relative abundances and blue lower. Network of differentially regulated vitamin metabolism genes (CYP2D6 and CYP3A4) (circles) and the

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significantly altered aqueous metabolite Nicotinamide (square), which was measured using LC-MS, in liver (square) (D). Asterisks (*) represent statistically significant differences between corn oil-treated and PBDE treated groups (FDR adjusted p-value <0.05). Caret signs (^) represent statistically significant baseline differences between CV and GF mice.

Figure 7. Pearson correlation matrix of differentially regulated gut microbiome OTUs (identified and quantified using 16S rRNA sequencing) and serum metabolites (quantified using LC-MS). Red indicates stronger positive correlations and blue stronger negative.

Figure 1.

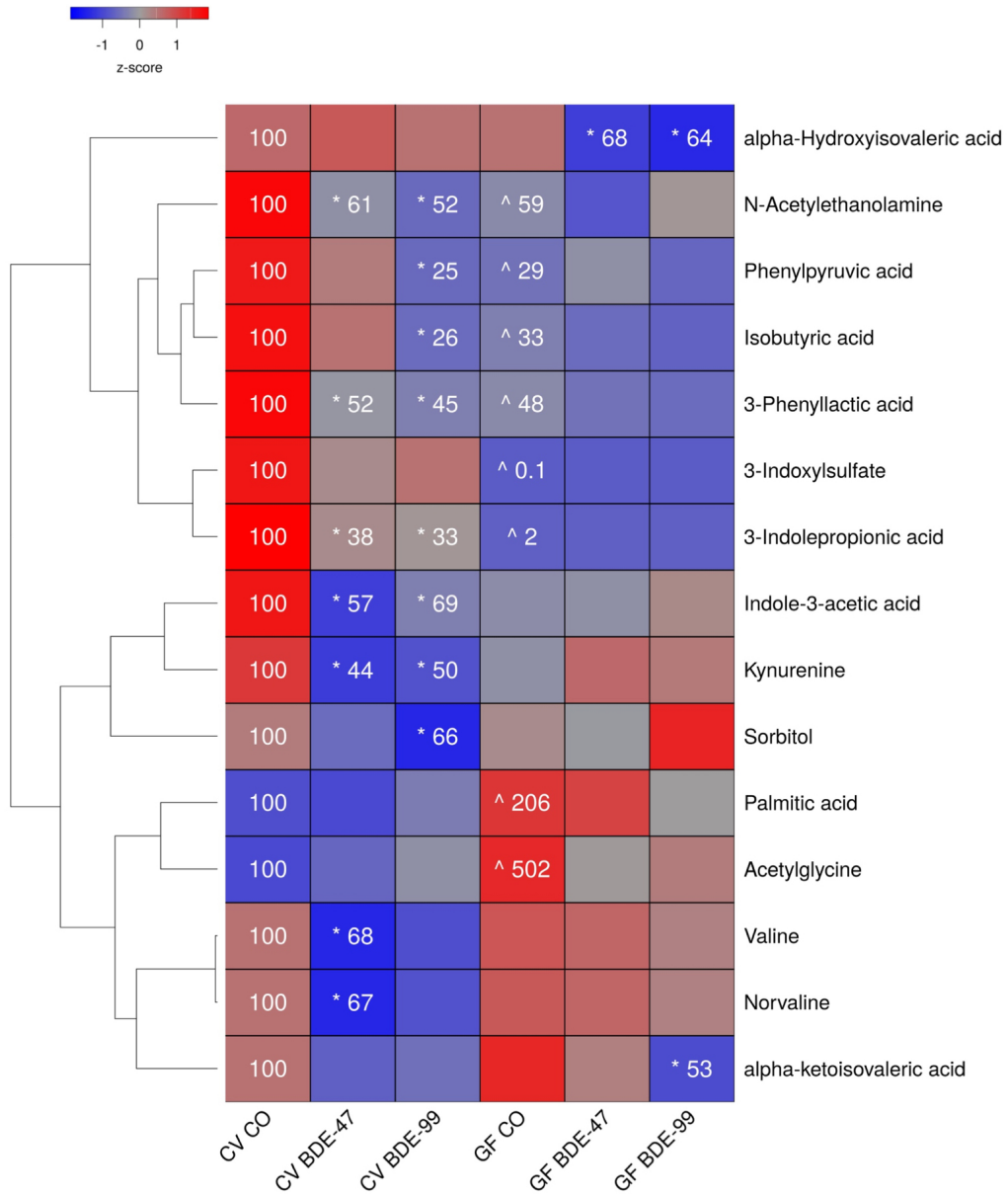


Figure 2.

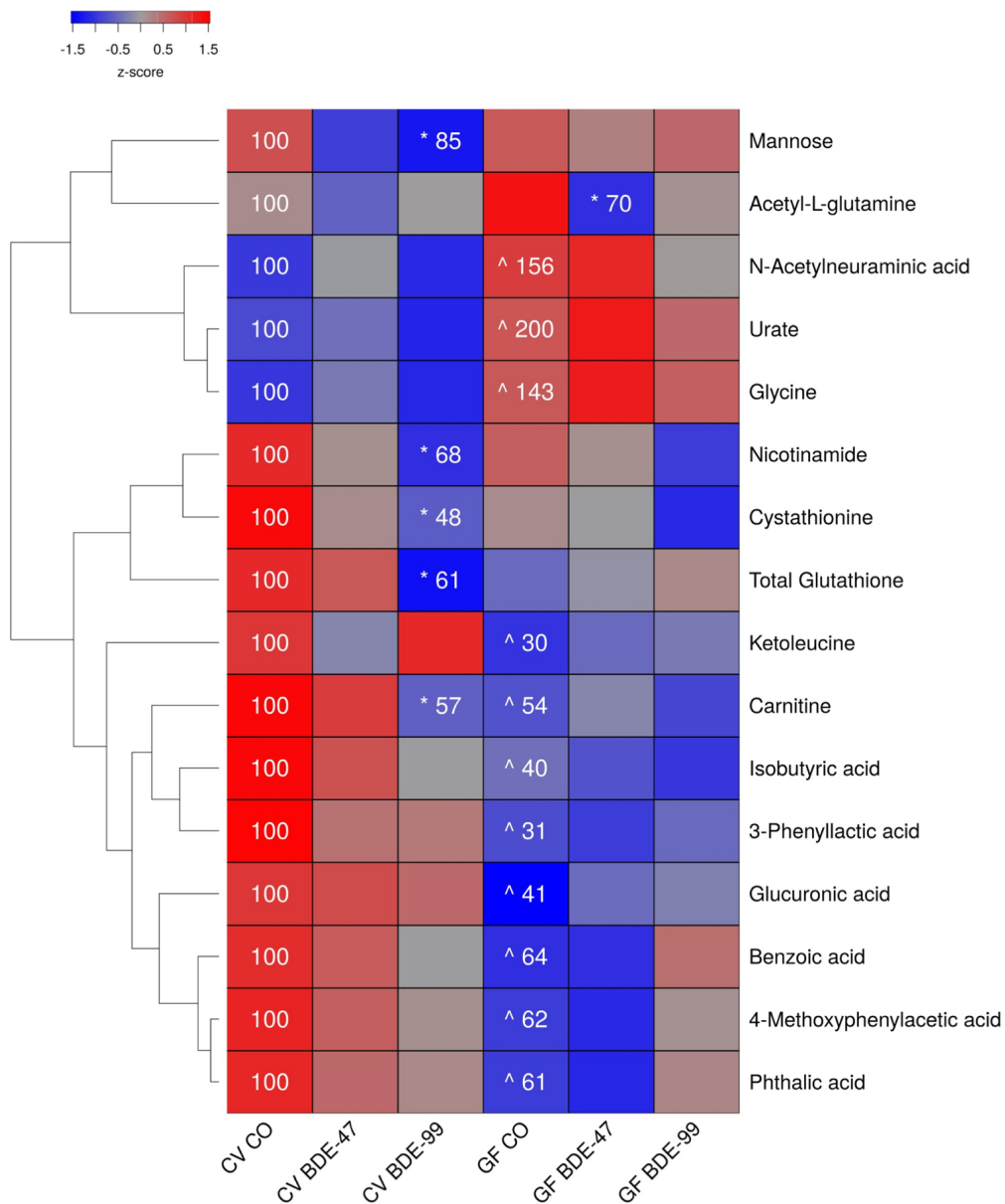


Figure 3.

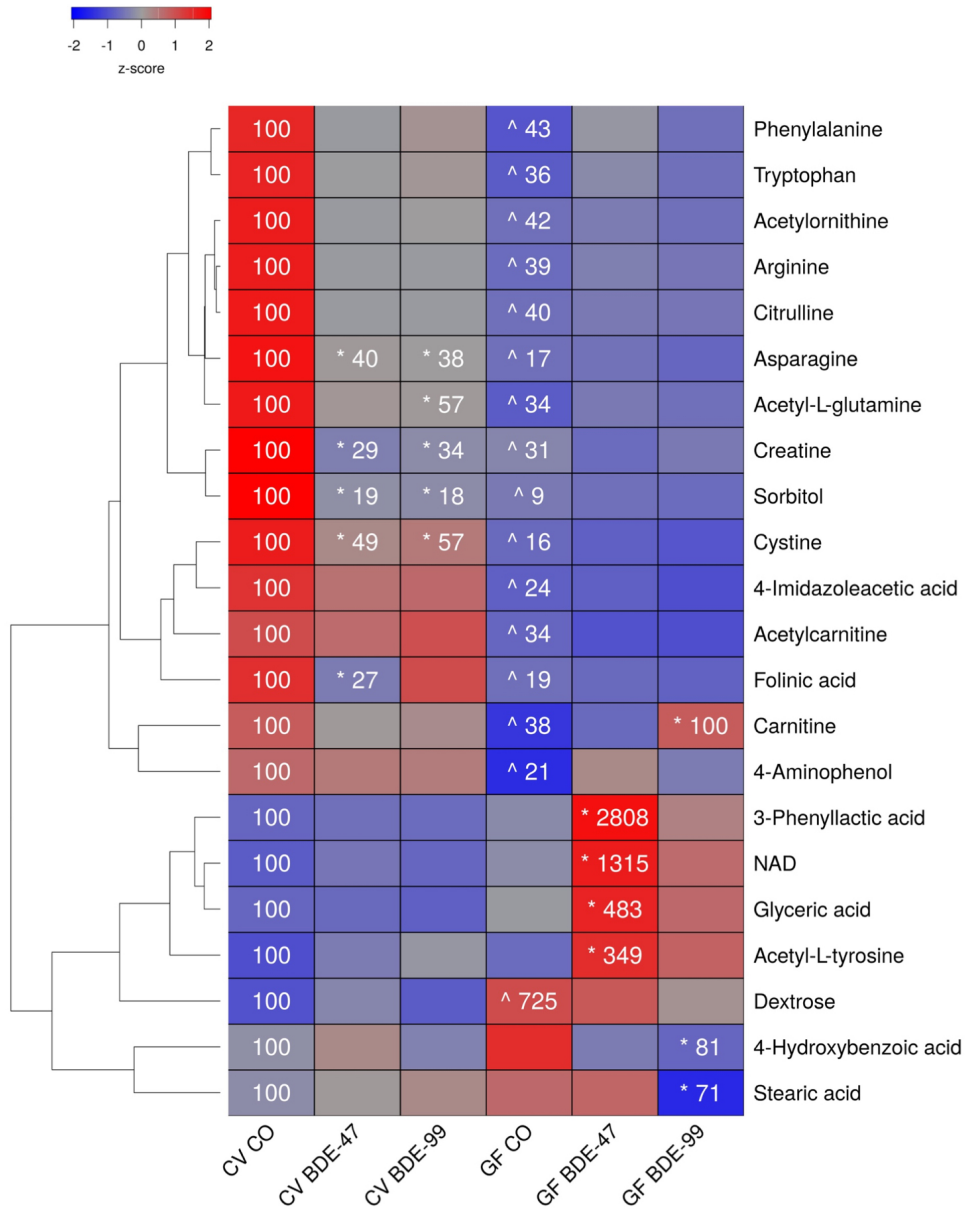


Figure 4.

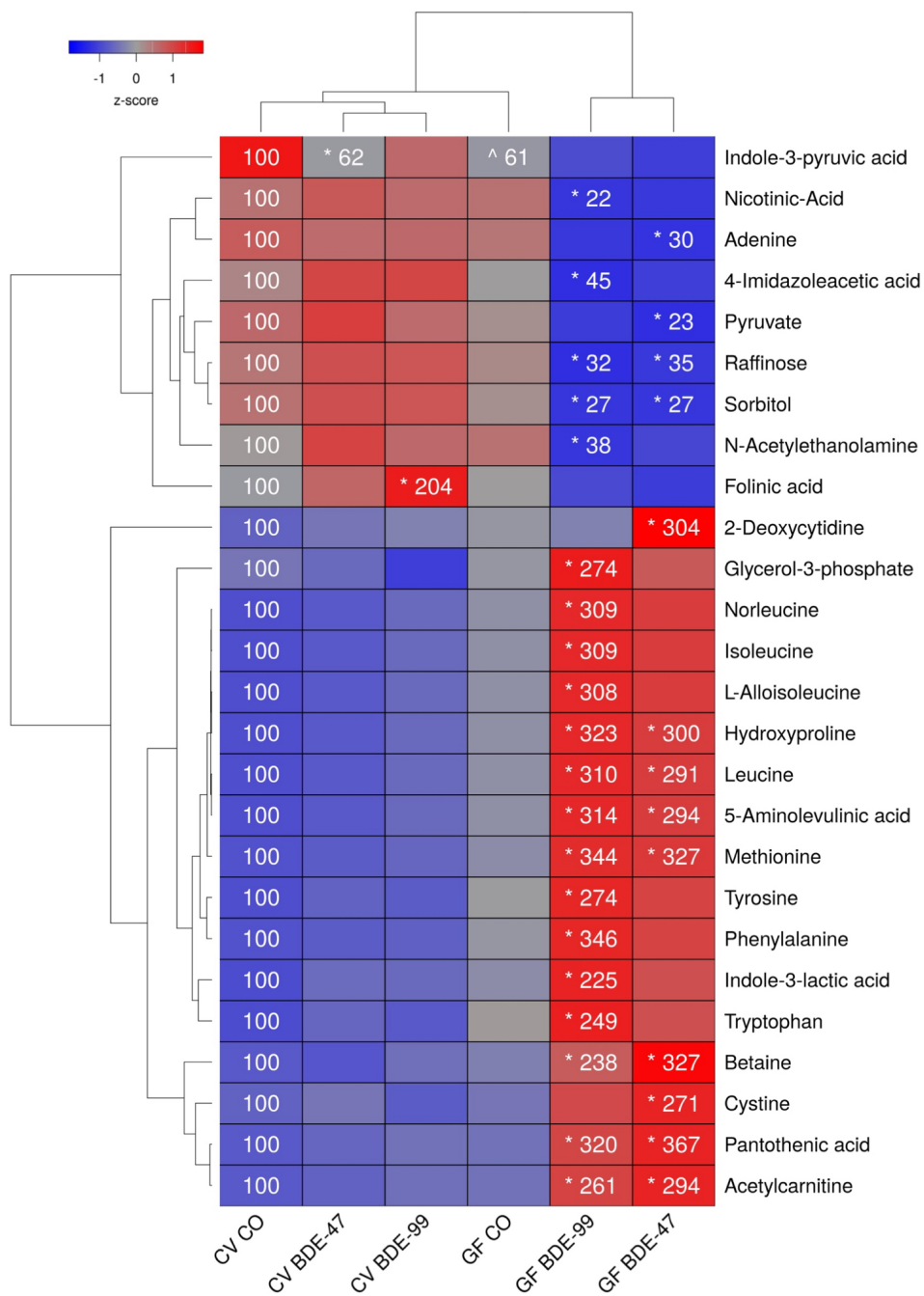


Figure 5.

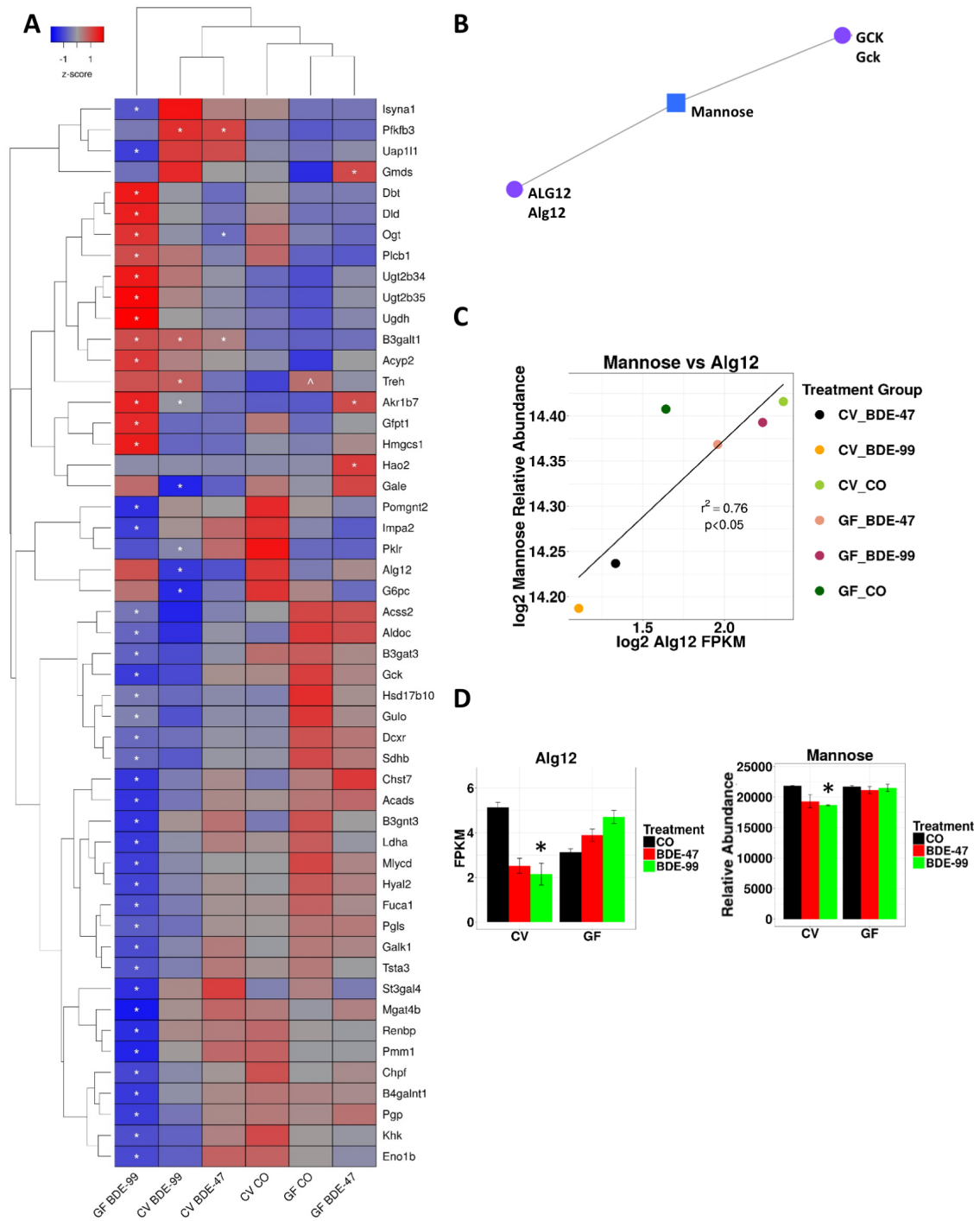


Figure 6.

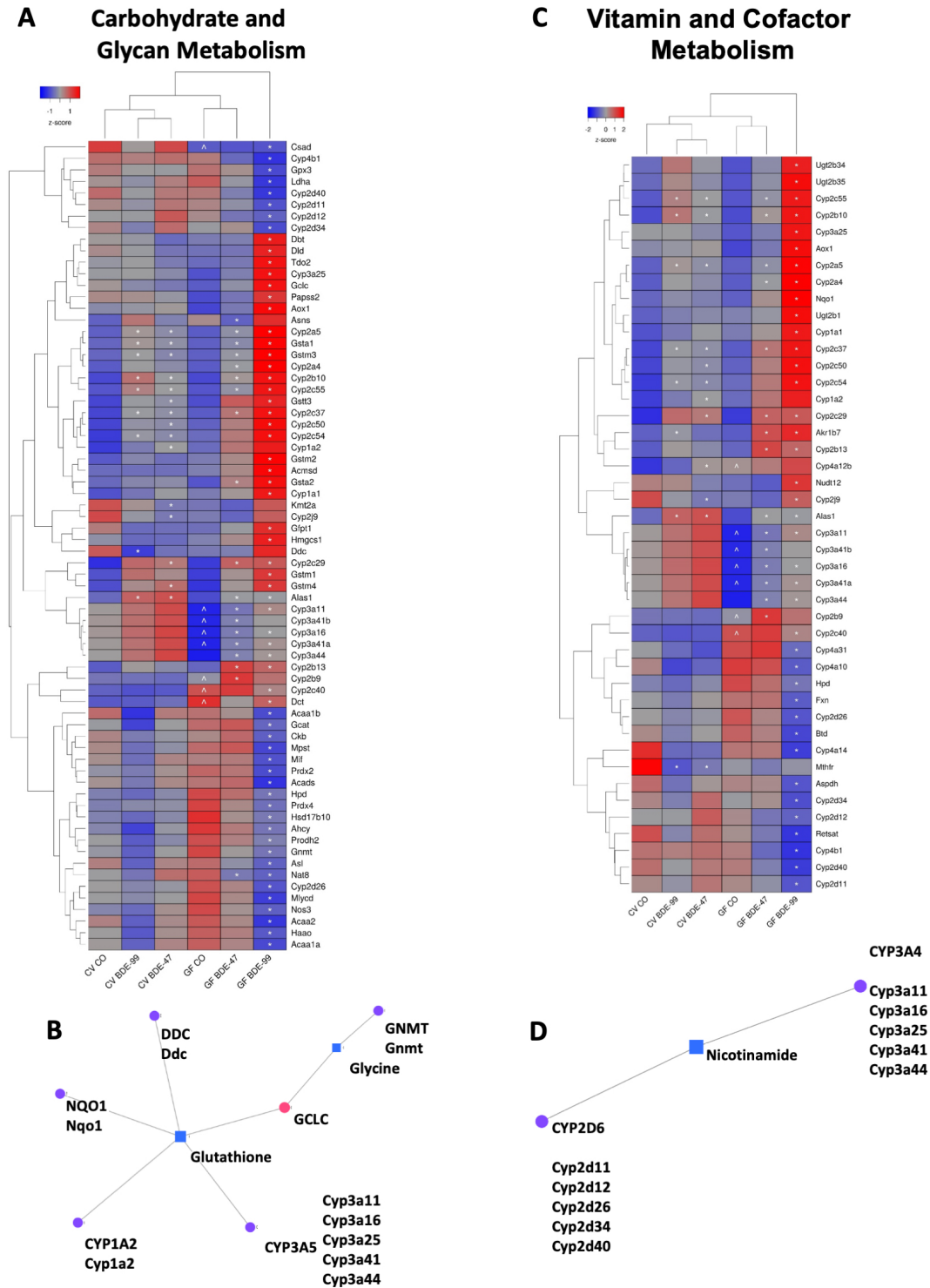
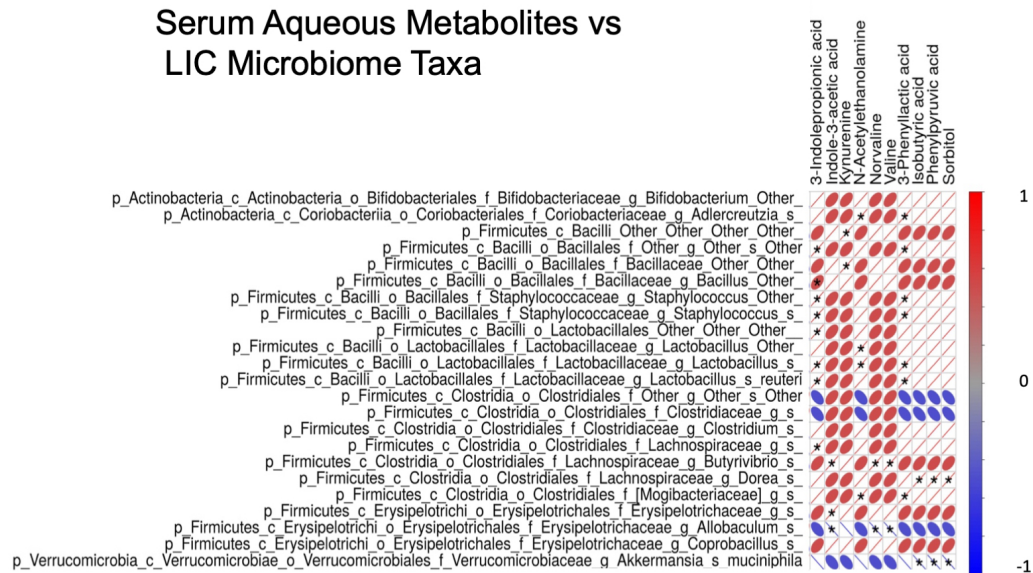


Figure 7.



**PBDEs and Gut Microbiome Modulate Metabolic Syndrome-Related
Aqueous Metabolites in Mice**

David K. Scoville, Cindy Yanfei Li, Dongfang Wang, Joseph L. Dempsey,
Daniel Raftery, Sridhar Mani, Haiwei Gu*, and Julia Yue Cui*

DKS, CYL, JLD, and JYC: Department of Environmental and Occupational Health Sciences, University of Washington, Seattle, WA 98105, USA;

DW: Department of Laboratorial Science and Technology, School of Public Health, Peking University, Beijing 100191, P. R. China;

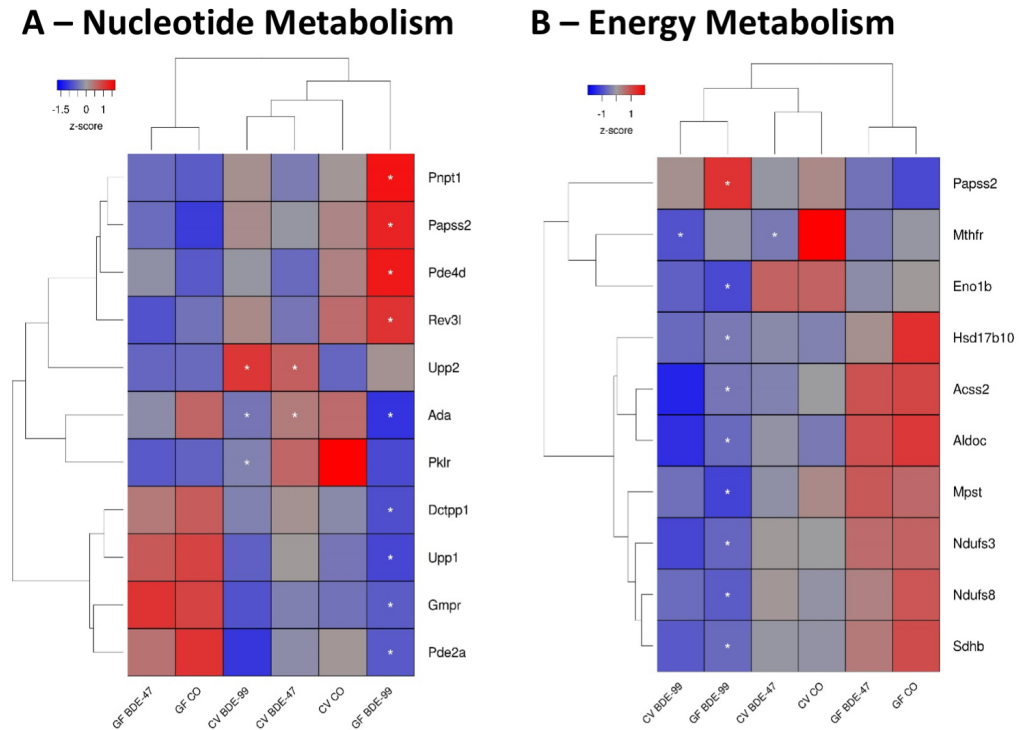
DW, DR: Northwest Metabolomics Research Center, Department of Anesthesiology and Pain Medicine, University of Washington, 850 Republican St., Seattle, WA 98109, USA;

SM: Albert Einstein College of Medicine, Bronx, NY, 10461, USA;

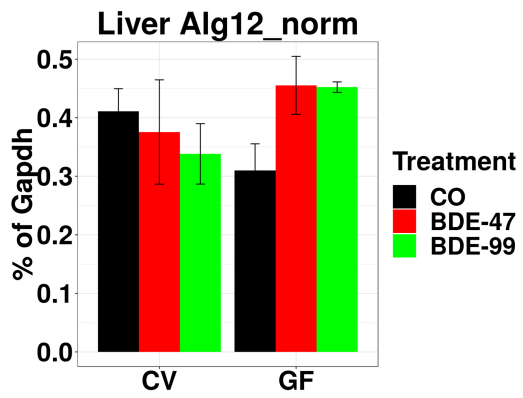
HG:
Arizona Metabolomics Laboratory, School of Nutrition and Health Promotion, College of Health Solutions, Arizona State University, Phoenix, AZ 85004, USA

*Primary laboratories of origin.

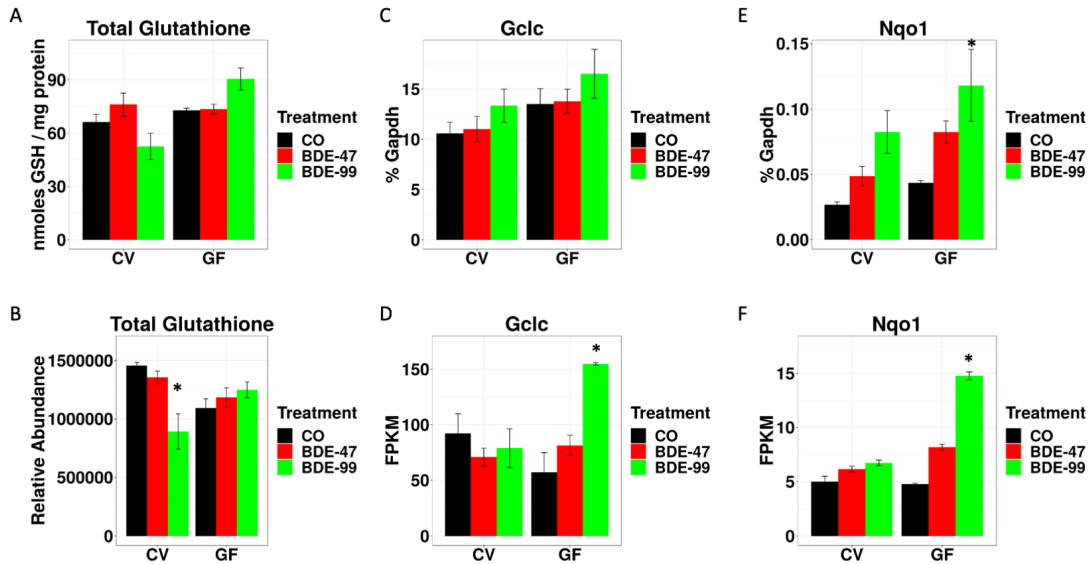
Supplemental Material



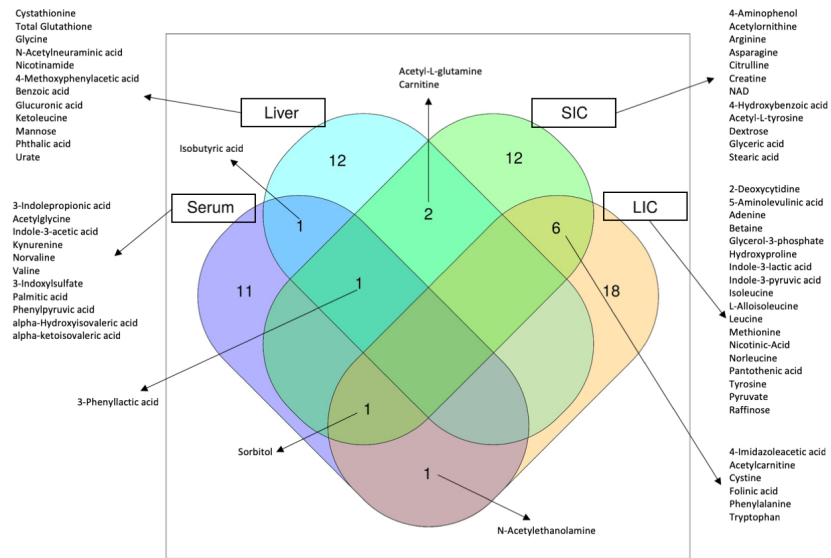
Supplemental Figure 1. Two-way hierarchical clustering dendrogram of nucleotide (A) and energy (B) metabolism genes that were significantly altered across treatment groups in liver of GF and CV mice following exposures to BDE-47 and BDE-99. Euclidian distance and complete linkage were used to generate the dendrogram. Red indicates higher standardized mean metabolite relative abundances and blue lower. Asterisks (*) represent statistically significant differences between corn oil-treated and PBDE-treated groups (adjusted p-value <0.05, Tukey's HSD post-hoc test). Caret signs (^) represent statistically significant baseline differences between CV and GF mice.



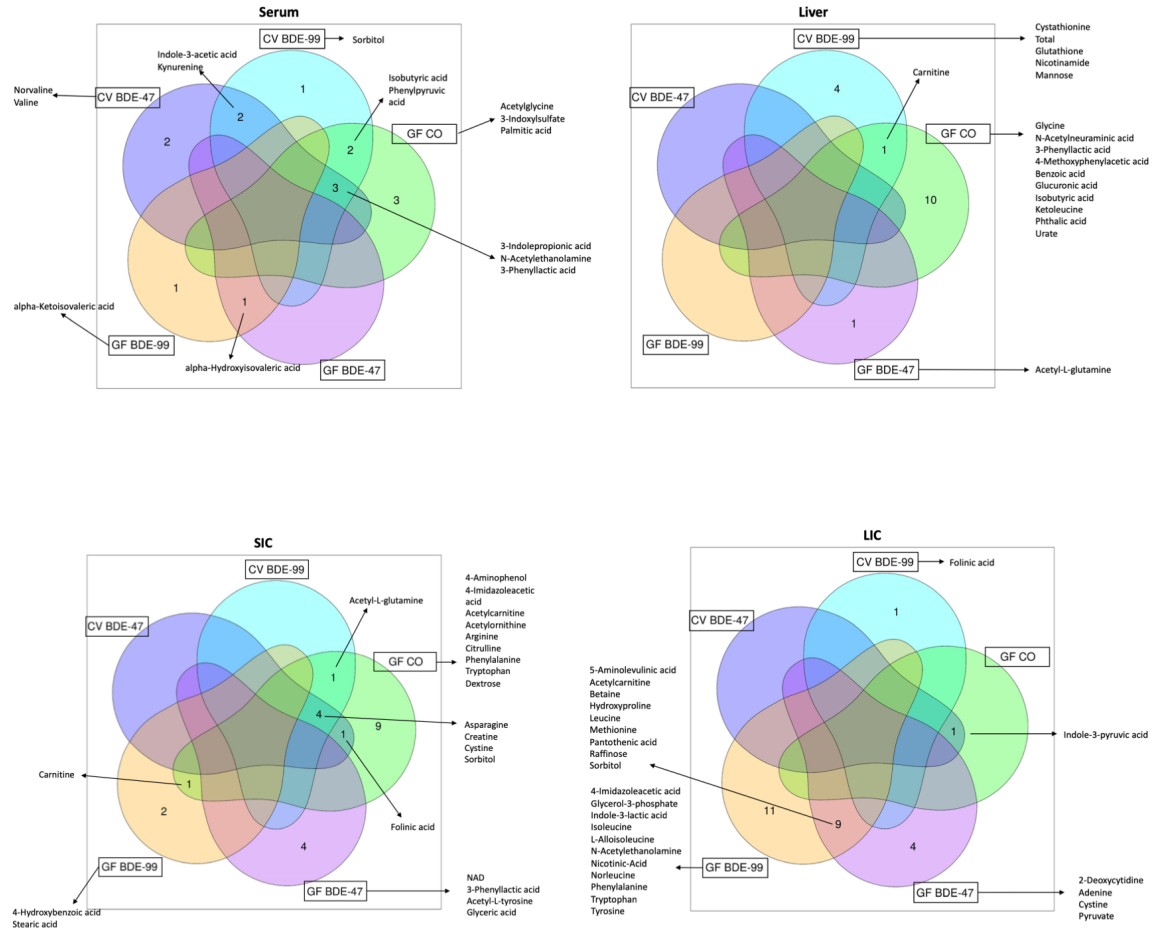
Supplemental Figure 2. Expression of Alg12 measured using RT-qPCR.



Supplemental Figure 3. Levels of glutathione measured by fluorescence (A), and LC/MS (B) shown with means and standard errors. Expression levels of Gclc and Nqo1, measured by RT-qPCR (C,E) and by RNA-Seq (D,F). Asterisks (*) represent statistically significant differences between corn oil-treated and PBDE-treated groups (adjusted p-value <0.05, Tukey's HSD post-hoc test). Caret signs (^) represent statistically significant baseline differences between CV and GF mice.



Supplemental Figure 4. Venn diagram of overlapping and uniquely differentially abundant changed metabolites across the different bio-compartments.



Supplemental Figure 5. Venn diagrams for each bio-compartment indicating overlapping and uniquely differentially abundant changed metabolites for the comparison groups (CV BDE-47 and CV BDE-99 vs CV CO, GF BDE-47 and GF BDE-99 vs GF CO, and between GF CO and CV CO mice).

Supplemental Table 1. All aqueous metabolites detected across bio-compartments.

2-3-Dihydroxybenzoic acid
2-3-Pyridinedicarboxylic acid
2-aminoadipic acid
2-Deoxycytidine
2-deoxyguanosine
2-Hydroxybenzoic acid
2-Hydroxyphenylacetic acid
3-Aminoisobutyric acid
3-Hydroxykynurenine
3-Hydroxyphenylacetic acid
3-Indolepropionic acid
3-Indoxylsulfate
3-Methyl-2-oxovaleric acid
3-Methyladipic acid
3-Phenyllactic acid
4-Aminobutyric acid
4-Aminophenol
4-Hydroxybenzaldehyde
4-Hydroxybenzoic acid
4-Hydroxyphenylacetic acid
4-Hydroxyphenylpyruvic acid
4-Imidazoleacetic acid
4-Methoxyphenylacetic acid
4-Methyl-2-oxopentanoic acid
4-Methylvaleric acid
5-Aminolevulinic acid
5-Hydroxytryptophan
5-Methylcytidine
6-Hydroxynicotinic acid
6-Methyl-DL-tryptophan
6-Phosphogluconic acid
7-ketocholesterol
Acetyl-L-glutamine
Acetyl-L-tyrosine
Acetylcarnitine
Acetylglucosamine

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Acetylglycine
Acetyloronithine
Adenine
Adenosine
Adenosyl-L-homocysteine
Adenylosuccinate
ADP
Agmatine
Alanine
Allantoin
Allopurinol
alpha-Hydroxyisovaleric acid
alpha-ketoisovaleric acid
AMP
Anthranilic acid
Arginine
Asparagine
Aspartic acid
Benzoic acid
Betaine
Cadaverine
Capric acid
Carnitine
Carnosine
cGMP
Citraconic acid
Citrulline
Creatine
Creatinine
Cystathionine
Cystine
Cytidine
Cytosine
D-Galacturonic acid
Dextrose
Dimethylarginine
DOPA
F6P
Folic acid

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Folinic acid
Fumarate
G6P
Galactose
GDP
Gentisic acid
Glucosamine
Glucuronic acid
Glutaconic acid
Glutamic acid
Glyceric acid
Glycerol-3-phosphate
Glycine
Glycoxyamine
Guanine
Guanosine
HIAA
Histamine
Homoserine
Hydroxyproline
Hypoxanthine
Indole-3-acetic acid
Indole-3-lactic acid
Indole-3-pyruvic acid
Inosine
Isobutyric acid
Isoleucine
Isovaleric acid
Ketoisoleucine
Ketoleucine
Kynurenine
L-Alloisoleucine
Lactose
Leucic acid
Leucine
Levulinic acid
Lysine
Malonic acid
Mannose

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Methionine
methyl- α -D-galactopyranoside
Methylguanidine
Mucic acid
N-Acetyethanolamine
N-Acetylneuraminic acid
N-N--Dicyclohexylurea
NAD
NADP
Nicotinamide
Nicotinic-Acid
Nonadecanoic acid
Norleucine
Normetanephrine
Norvaline
Ornithine
Oxalic acid
p-Coumaric acid
Palmitic acid
Pantothenic acid
Pentadecanoic acid
Phenylalanine
Phenylglyoxylic acid
Phenylpyruvic acid
Phthalic acid
Picolinic acid
Pipecolinic acid
Proline
Putrescine
Pyroglutamic acid
Pyruvate
Raffinose
Sarcosine
Sebacic acid
Serine
Serotonin
Shikimic acid
Sorbitol
Stearic acid

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Threonine
Thymine
TMAO
Total Glutathione
Trehalose
Tryptophan
Tyrosine
UDP-GlcNAc
Uracil
Urate
Uridine
Urocanic acid
Valeric acid
Valine
X5P
Xanthosine

Supplemental Table 2. Aqueous metabolites that were detected in serum. Average relative abundance values and standard errors are shown.

Metabolite	CVCO	SE	CVBDE-47	SE	CVBDE-99	SE	GFCO	SE	GFBDE-47	SE	GFBDE-99	SE
2-3-Dihydroxybenzoic acid	17409.92	3338.37	14560.90	3082.42	10044.35	1196.49	9525.48	4050.82	30693.08	17338.64	19222.36	5812.95
2-3-Pyridinedicarboxylic acid	5334.72	464.21	4504.42	426.85	6134.36	1627.32	5951.05	261.62	5177.78	663.76	3807.43	280.80
2-aminoadipic acid	31341.87	1236.34	110555.71	87371.32	24219.23	3051.86	24353.07	4030.45	24251.39	4940.98	23681.49	6707.42
2-Deoxycytidine	59122.34	2878.82	41528.54	10869.47	58945.22	3857.32	42346.99	1638.02	44618.75	4343.42	38994.58	4216.51
2-Hydroxybenzoic acid	3700.15	584.23	3009.60	860.82	2018.16	582.58	1868.59	222.71	3375.75	475.73	3947.23	955.02
2-Hydroxyphenylacetic acid	1102.38	207.02	720.78	122.98	861.79	222.82	1066.29	128.35	1040.02	56.34	608.57	63.96
3-Hydroxykynurenine	1251.84	318.91	2030.91	324.37	1101.80	67.22	1201.84	285.97	1072.83	243.31	1088.93	224.30
3-Hydroxyphenylacetic acid	2483.65	510.43	2025.97	469.27	3541.44	1078.40	1079.89	97.50	1539.75	256.93	1500.40	250.86
3-Indolepropionic acid	7355.24	938.30	2760.55	592.56	2454.80	1031.38	117.27	44.12	139.29	83.20	71.37	20.67
3-Indoxylsulfate	210417.63	51847.72	87677.75	26652.56	113392.01	37051.83	231.92	154.49	470.36	221.08	592.68	125.37
3-Methyl-2-oxovaleric acid	8000.16	516.93	6070.28	1448.01	6312.54	248.80	12875.55	2402.94	11932.58	1373.93	6747.45	1148.30
3-Methyladipic acid	1955.19	259.23	1472.13	423.06	1694.20	267.84	1870.37	387.83	2144.99	525.26	1893.93	182.84
3-Phenyllactic acid	2967.63	476.73	1551.30	228.17	1329.12	257.80	1430.65	198.03	1227.10	267.57	1163.10	255.43
4-Aminobutyric acid	434.57	147.76	12236.64	12065.82	438.98	100.47	238.14	71.85	603.60	277.68	547.04	202.13
4-Aminophenol	1392.80	99.13	4172.67	2482.46	1605.34	267.33	1481.11	139.51	1023.44	57.48	1626.64	182.18
4-Hydroxybenzaldehyde	1260.27	368.87	1083.02	134.08	1280.54	346.90	721.90	17.83	1265.58	287.31	810.06	106.99
4-Hydroxybenzoic acid	3440.52	757.05	2963.23	664.12	1849.99	552.19	1755.95	200.46	3687.12	603.36	3570.39	687.04
4-Hydroxyphenylacetic acid	1137.26	336.48	887.21	136.52	1160.86	185.23	1059.05	246.96	1141.69	69.16	1041.49	54.74
4-Hydroxyphenylpyruvic acid	10079.37	6786.05	1954.64	1165.16	548.86	207.37	860.21	205.22	342.41	74.09	495.59	170.25
4-Methoxyphenylacetic acid	72134.23	1695.60	62846.99	4163.40	84034.99	25943.73	74107.25	3966.04	65256.59	7505.22	45857.38	4884.88
4-Methyl-2-oxopentanoic acid	4288.14	237.53	3663.69	877.60	3158.32	277.75	6358.61	1364.66	5577.55	461.25	3562.66	670.21
4-Methylvaleric acid	39358.05	5650.40	26595.60	4137.04	25362.48	4769.39	34194.03	3744.06	31306.26	4625.64	20072.60	3623.58

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5-Aminolevulinic acid	3071011.00	231226.10	3102472.00	961683.50	2559518.00	322088.60	3307101.00	432742.70	3291751.00	124593.10	3118724.00	102400.00
5-Methylcytidine	1114.10	157.74	779.15	88.28	1522.89	167.74	731.58	101.01	838.13	82.28	1060.70	172.82
6-Hydroxynicotinic acid	10444.65	707.04	6535.35	2118.29	5093.11	1544.70	12238.73	2657.66	11163.49	1709.63	9483.18	1751.08
6-Methyl-DL-tryptophan	2228.04	99.97	2367.77	793.04	1413.19	514.67	1615.74	93.40	1554.34	357.05	1343.81	395.72
7-ketcholesterol	37207.79	15475.29	38301.34	16173.35	30498.95	4543.58	21344.05	2037.39	42840.47	19602.36	24808.71	6528.80
Acetyl-L-glutamine	18405.78	2180.32	92030.10	73872.83	18505.06	1863.13	23573.85	3761.93	24252.89	789.91	20162.65	1931.25
Acetyl-L-tyrosine	4550.25	2521.91	1702.73	963.28	4174.06	2315.85	3846.78	2910.30	4971.07	1339.55	7070.84	1949.25
Acetylcarnitine	284616.12	179891.09	85546.98	57860.97	0.00	0.00	11.73	11.73	131.87	131.87	1445.06	817.83
Acetylglucosamine	2711.82	416.41	10608.49	8637.77	2726.31	403.69	6594.98	3435.97	2331.40	262.40	1819.87	246.42
Acetylglycine	1155.11	335.29	1796.51	222.25	2728.12	488.55	5799.75	1248.96	3222.82	264.43	3836.03	1591.17
Acetylmethionine	23541.39	1438.02	18865.85	3300.97	17434.77	800.87	19574.79	1798.76	20014.30	1155.31	19733.42	609.37
Adenine	16354.47	2705.27	582798.67	532297.92	149304.81	60487.11	35622.26	4735.15	105105.38	37079.75	104875.11	40204.83
Adenosine	68348.87	13040.13	2211231.45	1979440.67	641624.20	322739.45	31525.94	6224.35	362988.63	206787.51	327833.80	170221.82
Adenosyl-L-homocysteine	3638.61	490.16	284158.32	281536.86	8577.72	3021.35	12619.93	2257.49	18517.88	6392.72	22952.76	5864.43
Agmatine	2818.19	196.28	2134.10	215.36	2468.55	258.33	2586.84	281.92	2288.13	72.91	2830.88	238.05
Alanine	139978.20	9815.23	240624.50	149094.02	114124.20	9834.84	124574.90	11750.78	129359.90	18112.25	142204.80	4985.91
Allantoin	52882.18	2296.47	64346.79	23724.68	53744.64	5241.63	50649.14	3902.76	44173.93	7357.79	48228.55	6035.84
Allopurinol	7823.98	1456.46	508224.68	500468.09	21808.26	6037.69	9125.08	2176.59	28052.95	12360.19	20412.09	7917.35
alpha-Hydroxyisovaleric acid	7730.49	434.96	7960.62	582.37	7574.50	414.10	7583.92	168.83	5271.39	420.41	4930.88	518.31
alpha-ketoisovaleric acid	2992.66	363.46	1797.97	225.56	1996.08	173.65	3865.47	577.55	2839.24	454.95	1598.18	158.77
Anthranilic acid	892.63	124.71	1047.04	38.19	715.25	189.81	422.76	140.43	491.38	147.88	567.27	117.88
Arginine	1006965.20	54339.11	781533.10	124136.75	662357.00	46611.98	839479.90	82960.66	868094.80	43129.07	828956.30	20113.05
Asparagine	98693.64	6909.92	147742.35	66991.11	94370.27	10071.69	89847.84	8873.15	91201.51	7894.64	109642.40	16581.94
Aspartic acid	24136.00	1749.23	126950.55	112743.82	22636.87	7372.23	25759.01	6089.43	28412.51	3614.36	34057.47	7926.59
Benzoic acid	6093.74	347.04	5347.84	551.33	6597.67	1478.22	6054.96	250.69	5040.01	828.30	3917.11	494.60
Betaine	7462280.00	146836.50	11204374.00	3019646.60	9159507.00	621585.00	6865704.00	488092.30	6193527.00	277364.60	7698704.00	635791.10

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Carnitine	433541.30	27268.58	1045489.30	561170.00	583222.40	2008.98	439187.30	7091.38	463460.90	21671.58	486735.10	47458.27
Carnosine	1736.13	104.47	9146.20	6999.56	2453.61	340.64	1238.01	110.89	1747.11	428.79	1392.81	191.63
Citraconic acid	5577.01	285.50	4499.67	1419.38	4916.72	202.79	8303.24	1355.83	7988.38	607.17	4715.13	274.65
Citrulline	96694.39	4622.20	76305.10	12528.06	63533.86	4684.05	83413.48	8213.43	82622.14	3639.56	82022.74	1875.40
Creatine	3469.63	281.18	6040.27	2853.94	3013.54	494.57	2832.89	246.45	4196.47	176.86	3220.61	1096.47
Creatinine	13544.77	1008.65	19928.90	5803.68	15824.27	1725.57	14390.17	707.38	15799.02	6694.66	13130.68	2592.61
Cystathionine	2134.61	420.36	20189.59	16936.34	3271.27	363.34	2348.15	194.11	3221.61	699.50	2904.19	174.63
Cystine	18899.59	836.67	19173.24	5748.33	26199.76	3632.75	24357.26	3381.67	19834.48	4243.63	20868.55	3866.17
Cytidine	45536.69	3532.43	78590.60	26779.91	68072.21	10101.89	60534.89	3854.18	57311.90	4776.00	55823.10	6978.68
Cytosine	7877.06	292.36	16029.67	7451.89	11320.29	1774.81	10271.94	807.09	9957.12	1556.00	9609.45	1749.51
Dimethylarginine	38566.41	6743.69	152987.76	121639.17	33820.89	6817.53	43296.92	4397.81	45304.79	2819.31	38623.51	9509.95
DOPA	1004.30	235.16	2652.79	901.91	1410.50	189.74	870.18	113.86	1464.82	44.94	1367.96	546.27
F6P	6785.31	972.34	102415.48	97847.47	6649.12	1076.55	4083.58	313.60	5848.06	986.92	5516.25	549.35
Folinic acid	365.29	99.24	376.62	197.47	162.61	94.67	71.14	27.11	101.37	21.94	160.51	58.56
Fumarate	32156.28	2933.37	24266.61	9101.81	34179.07	6608.82	39614.53	3613.60	39414.28	3220.51	45579.94	4496.39
G6P	6803.13	979.84	114368.08	109796.91	7099.71	1253.02	4378.51	272.62	6228.87	727.71	5412.51	680.55
Gentisic acid	11838.43	2116.64	9598.13	2048.28	6618.69	756.44	6050.70	2382.57	18852.85	9063.25	12928.76	3528.02
Glucosamine	1815.32	615.94	41644.18	41210.99	1031.67	473.10	2454.06	709.13	2478.20	431.42	2153.97	260.86
Glucuronic acid	8289.04	390.43	16151.70	6658.33	10277.76	1307.89	4982.63	186.85	5729.51	438.22	7431.44	527.19
Glutaconic acid	7872.61	536.25	6312.97	1688.43	6342.52	266.47	11832.47	1847.87	11621.79	958.83	7032.77	1389.99
Glutamic acid	247870.20	11583.48	934046.00	814008.08	186249.30	40899.01	183391.90	43527.51	201259.50	13186.38	231280.60	18407.11
Glyceric acid	1915.96	204.51	4132.41	2060.34	3238.84	689.21	2462.09	287.40	3295.60	244.39	3148.58	388.89
Glycerol-3-phosphate	50531.48	7796.07	41803.83	10864.43	41503.37	912.52	25995.15	3334.24	29838.48	1105.12	30144.15	8207.06
Glycine	18198.03	1313.87	27874.13	13269.65	16644.93	1673.89	31025.42	1076.67	26349.07	3347.84	29733.37	1020.36
Glycocyanine	2535.56	253.39	5112.26	3037.77	1768.80	173.86	3372.60	598.76	3840.26	199.29	5209.83	1647.58
Guanine	3326.03	722.00	17695.07	15769.65	5239.83	1703.31	3453.93	691.70	6162.48	2382.58	3261.17	1248.10

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Guanosine	9257.75	3219.07	42683.48	36798.13	13639.83	2941.40	7685.17	1576.63	14950.92	6356.39	8265.91	3174.46
HIAA	1346.79	136.58	872.80	328.21	563.74	118.26	632.63	105.60	1066.82	445.75	1104.90	298.58
Homoserine	420721.90	11851.97	388352.50	93150.81	319162.10	30640.19	440549.40	58815.73	429743.60	26166.70	412105.90	44456.12
Hydroxyproline	836619.80	59503.38	843428.50	259326.52	696426.50	88657.62	904093.80	120751.06	905445.50	35032.55	854573.40	20935.27
Hypoxanthine	8023.43	1610.75	479151.69	472130.25	21291.01	5958.17	7879.91	2538.23	23850.69	11925.82	19441.26	7452.42
Indole-3-acetic acid	9167.41	820.57	5259.81	435.45	6357.90	660.97	6532.07	585.38	6620.88	463.37	7182.46	288.40
Indole-3-lactic acid	7263.06	1141.96	4524.29	459.97	6396.94	1488.39	7370.26	723.28	6559.49	1311.78	7788.87	909.27
Inosine	74841.05	15581.14	4187828.31	4123119.74	188422.04	54031.00	76496.31	19409.56	228704.63	106115.56	179298.16	69686.16
Isobutyric acid	37420.80	5037.56	23246.02	7138.22	9761.13	2510.12	12530.31	3741.36	9596.44	516.90	8351.10	3475.89
Isoleucine	5773013.00	401158.30	5779482.00	1769296.60	4774272.00	612005.00	6232423.00	823479.50	6223698.00	207098.90	5917312.00	169938.40
Isovaleric acid	4433.62	599.86	5254.47	1698.67	3739.17	754.82	6453.01	982.92	6128.07	226.59	3432.81	974.06
Ketoleucine	10323.14	729.21	8616.64	1771.98	8510.34	188.17	16566.19	3139.18	15598.55	1560.23	9219.19	1639.42
Ketoleucine	8943.61	934.82	7084.60	1193.99	7346.31	92.32	13735.52	2716.08	13122.46	1170.27	7214.63	1375.77
Kynurenine	2825.08	229.09	1245.18	272.15	1423.16	350.49	1915.16	281.85	2438.97	220.63	2329.71	103.75
L-Alloisoleucine	6418992.00	444672.80	6451596.00	1964997.60	5337093.00	670658.80	6928816.00	915908.90	6913782.00	249467.60	6576573.00	173611.50
Leucic acid	2008.41	97.79	1855.44	435.11	2002.26	273.78	2258.24	292.77	1477.64	177.16	1061.48	210.84
Leucine	5724311.00	403942.90	5696907.00	1727147.40	4752083.00	583537.90	6170612.00	825389.20	6160563.00	250584.40	5834754.00	164073.20
Levulinic acid	2502.57	403.54	1609.90	338.90	1678.80	257.85	2653.77	738.30	2216.60	188.35	1698.26	117.47
Lysine	940067.50	36758.27	1083175.30	333127.20	708429.30	48295.13	774378.70	71977.25	742258.90	43141.81	757955.40	39786.97
Mannose	27957.70	1853.59	27080.33	3980.61	32006.10	2175.13	30155.35	2303.28	27737.21	2390.61	31904.73	1171.60
Methionine	358689.40	21772.82	382777.70	100698.18	283622.90	33418.13	295194.20	35018.97	303575.10	31585.61	344409.30	10046.58
methyl- α -D-galactopyranoside	7112.25	1153.04	6953.06	1038.55	7498.91	667.12	5883.15	1621.06	8731.08	1449.26	7671.60	1364.41
Methylguanidine	30072.88	1726.17	26709.62	5793.38	21766.35	3289.87	31927.96	3272.23	28385.28	3973.81	27107.79	3085.30
N-Acetyethanolamine	6610.82	216.38	4035.95	641.55	3433.30	344.05	3921.70	577.87	3179.99	247.29	4395.70	441.67
N-Acetylneuraminic acid	533.13	77.80	4466.48	4091.01	710.15	11.05	734.91	37.06	435.05	61.29	617.48	84.01
N-N-Dicyclohexylurea	11003.60	1141.89	10743.49	3281.97	12803.18	2486.21	10589.00	3841.57	7076.11	1407.19	9822.63	2631.05

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Nicotinamide	123785.40	7148.54	3292161.10	3146939.58	139135.00	36819.52	190319.00	29729.38	196058.00	34857.84	140033.90	7664.97
Nonadecanoic acid	335951.00	39439.00	346040.70	47750.97	314424.70	18504.01	296384.50	54413.74	234042.70	26576.54	304918.00	13098.92
Norleucine	5311831.00	364514.90	5296326.00	1639154.30	4432061.00	554869.80	5803524.00	769193.60	5726154.00	216862.20	5433150.00	134754.90
Normetanephrine	1483.49	187.06	2314.28	1217.02	1539.39	402.10	1539.85	238.77	1409.09	724.49	1361.32	357.85
Norvaline	1102034.70	48184.18	740048.70	18105.15	838284.60	94476.68	1161256.00	119282.87	1134100.90	48226.79	1064756.80	11173.26
Ornithine	179289.48	16854.88	378033.36	244784.81	115155.32	18780.02	97938.36	22634.54	86661.33	6940.24	111460.94	17457.64
Oxalic acid	2946.89	496.26	4761.57	1510.81	4578.57	515.97	4570.12	269.52	5570.04	848.99	6066.02	428.39
p-Coumaric acid	3975.33	1340.77	2961.76	433.70	2127.11	960.91	2240.10	742.67	2966.25	542.44	7769.23	2593.38
Palmitic acid	371432.00	16583.40	356358.30	43252.72	462690.00	48082.26	763416.70	57141.39	733584.00	56092.46	544589.30	103867.30
Pantothenic acid	156855.50	24656.91	195042.20	43504.93	152503.20	13383.01	116002.70	9863.39	153030.60	2420.68	139602.20	3160.23
Pentadecanoic acid	666298.20	126039.86	453020.80	85084.40	527487.50	52296.37	425809.60	19779.31	322073.60	29647.52	538881.60	194228.67
Phenylalanine	2675364.00	155166.36	3283232.00	1096423.64	2405454.00	137335.24	2530701.00	196683.74	2621651.00	32970.68	2898959.00	276192.27
Phenylglyoxylic acid	388.42	58.55	310.31	14.67	357.15	35.19	420.70	102.10	487.04	97.38	415.46	66.41
Phenylpyruvic acid	1349.40	323.15	811.64	256.72	338.85	90.09	391.66	126.03	543.89	121.12	335.08	76.95
Phthalic acid	68483.80	3089.48	61840.12	4345.19	79959.07	24156.74	72814.32	2786.32	63026.15	8288.03	42619.28	7677.49
Pipecolic acid	1189812.50	53672.19	2113006.40	864719.96	1258853.60	43253.54	932683.10	61656.82	869463.10	64617.74	1006267.70	42718.03
Proline	510669.70	41837.44	492556.40	83676.58	407427.60	51654.38	396901.40	47115.41	405935.00	31151.83	490628.60	22815.69
Pyroglutamic acid	1126152.40	53055.01	2020174.30	823784.09	1199364.70	43248.22	891851.00	58332.28	823416.80	57707.59	951954.10	36840.52
Pyruvate	1302.47	278.93	3121.79	2148.47	543.38	59.13	470.49	169.40	431.25	130.48	677.01	104.91
Raffinose	20.10	15.28	643.68	504.61	44.93	32.05	1008.83	94.79	756.69	25.69	837.66	38.41
Sarcosine	109427.91	7322.42	191566.09	119273.74	88555.19	7375.25	100099.94	9134.71	103120.58	15316.59	110426.96	4199.25
Sebacic acid	2370.74	97.21	2555.27	631.91	1946.46	114.65	2640.84	195.06	2388.19	231.85	2530.92	115.04
Serine	245080.00	9475.51	281801.30	90724.41	213075.00	15497.47	259018.00	34324.41	254810.70	27424.58	265176.10	19716.08
Serotonin	47170.12	2719.23	49093.28	11007.91	37216.23	6288.93	51946.26	6276.39	51903.50	2209.01	55280.11	3361.36
Shikimic acid	498.57	96.87	1064.53	239.89	987.19	105.47	1251.56	233.68	712.52	127.33	948.97	220.90
Sorbitol	4355.29	76.59	3564.48	360.76	2874.59	163.08	4207.90	271.47	3974.98	308.61	5224.14	267.26

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Stearic acid	139585.50	15301.99	126384.10	29976.27	157697.30	32359.34	212298.60	54597.99	136135.40	15484.76	113655.20	24550.18
Threonine	470205.00	9765.66	447696.10	106517.66	353854.40	35750.53	492802.90	61706.05	478546.30	31232.22	470370.10	51906.68
Thymine	14714.51	3349.21	9422.15	3288.74	11235.01	801.71	7743.67	826.15	7610.03	1646.13	8130.13	1432.79
Total.Glutathione	22083.14	2138.34	210945.51	189883.58	23691.70	761.20	26261.66	116.44	24348.24	3510.50	33876.21	8338.08
Tryptophan	796555.10	50449.36	506833.30	29577.95	627826.90	145433.42	786128.30	74449.33	717277.10	100629.96	823597.60	52986.34
Tyrosine	313397.60	20639.24	233082.50	51325.48	206106.90	27385.29	271869.60	21293.54	250809.70	14446.15	282177.40	20149.66
Uracil	357705.10	45134.01	1242175.00	770455.61	598963.40	52549.73	873808.80	149529.13	928824.50	105135.28	1146424.50	261638.36
Uridine	529627.00	67956.07	1732350.90	986939.35	880962.20	80566.80	1322503.00	223413.50	1384783.80	111333.04	1654102.20	407131.82
Urocanic acid	41422.00	7136.56	29994.94	3820.51	31722.20	5112.32	79605.64	31801.06	49775.25	2336.22	44915.74	10454.78
Valeric acid	4433.62	599.86	5254.47	1698.67	3739.17	754.82	6453.01	982.92	6128.07	226.59	3432.81	974.06
Valine	1170787.20	49948.30	793015.60	21405.13	889446.30	99684.24	1238077.80	121992.38	1204533.90	44096.87	1131445.60	13594.87
X5P	10006.10	325.57	61392.54	54707.51	12143.16	2870.97	8707.79	1077.99	19203.17	4675.56	9937.94	1627.66
Xanthosine	3687.79	1253.75	54516.02	50939.12	10470.16	1348.35	11203.91	2850.63	9483.29	1651.54	13380.20	5398.49

Supplemental Table 3. Aqueous metabolites that were detected in liver. Average relative abundance values and standard errors are shown.

Metabolite	CVCO	SE	CVBDE-47	SE	CVBDE-99	SE	GFCO	SE	GFBDE-47	SE	GFBDE-99	SE
2-3-Dihydroxybenzoic acid	2581.97	475.56	2322.81	153.55	3850.30	2282.75	1199.45	400.85	1350.22	529.78	2965.55	666.12
2-3-Pyridinedicarboxylic acid	5804.56	238.13	5307.57	684.80	5148.92	546.30	3619.13	367.41	3437.69	978.40	5377.87	545.43
2-aminoadipic acid	466707.10	97161.14	299167.40	82217.35	310001.00	110156.79	307632.00	71107.33	331255.00	127317.33	264623.40	66766.89
2-Deoxycytidine	16208.27	4403.91	14269.04	2489.10	16209.71	4249.33	8491.56	1651.96	9697.20	243.40	9880.40	1453.08
2-Hydroxybenzoic acid	2377.92	173.56	2250.37	434.09	1904.42	343.08	1240.18	158.92	2027.76	182.65	2935.60	977.23
3-Aminoisobutyric acid	11661.81	1157.52	16558.81	921.89	14207.40	3302.49	14253.60	2987.60	18388.08	1286.09	18452.95	3349.97
3-Hydroxykynurenine	1605.57	190.91	1758.43	429.70	1866.40	467.27	1930.40	187.76	1473.69	134.32	1662.69	229.90
3-Hydroxyphenylacetic acid	1188.36	277.35	851.43	144.27	2214.52	660.37	524.40	99.37	885.48	305.15	762.54	253.53
3-Indolepropionic acid	816.72	390.19	465.99	90.13	1051.25	205.77	231.40	117.29	436.43	181.11	821.00	131.17
3-Indoxylsulfate	6156.78	5477.17	102.63	57.45	16822.32	16647.56	29.02	12.70	56.09	38.83	102.35	47.30
3-Methyladipic acid	3289.06	182.20	2551.75	174.13	3240.78	577.65	3618.56	208.91	3124.60	407.18	3578.41	308.20
3-Phenyllactic acid	1976.00	434.87	1331.38	90.13	1288.45	212.62	614.87	66.17	520.56	38.71	778.39	66.69
4-Aminobutyric acid	44929.75	4975.47	63287.75	1715.45	55878.48	11368.19	55656.64	10898.19	70253.56	6816.82	74938.62	14173.43
4-Aminophenol	22193.15	1793.19	22064.90	4465.33	14662.78	2556.01	21878.45	6107.04	22121.08	2151.80	17623.76	1393.08
4-Hydroxybenzaldehyde	1485.56	151.17	1643.23	144.28	1626.13	257.43	2420.15	494.48	1698.19	527.94	2179.16	591.87
4-Hydroxybenzoic acid	2352.08	260.36	1964.92	421.35	1586.83	294.24	1287.17	202.25	2030.30	270.62	2900.58	1050.81
4-Hydroxyphenylpyruvic acid	890.10	75.56	979.51	170.00	833.85	14.64	221.76	20.01	426.42	252.61	613.79	158.24
4-Imidazoleacetic acid	43117.12	2406.42	46352.89	3581.26	34406.23	6117.58	41095.48	1116.59	38138.65	2372.62	46198.59	3471.86
4-Methoxyphenylacetic acid	61541.20	5344.72	54761.30	3747.23	49932.22	4545.96	37908.24	5397.20	35787.80	2607.68	49447.55	3475.73
4-Methylvaleric acid	20334.24	2730.41	16524.55	1353.34	20238.77	3556.96	17580.36	1061.50	22653.30	2714.35	25109.64	5184.07
5-Aminolevulinic acid	6487514.00	341947.70	5917114.00	688998.80	6574487.00	878261.50	6403798.00	865963.70	6510070.00	659439.00	5547313.00	212482.00
5-Methylcytidine	671.46	229.06	702.73	181.00	798.78	190.91	904.52	239.00	1119.21	190.34	794.02	311.62

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6-Hydroxynicotinic acid	18402.95	1922.68	18424.80	5606.87	13836.97	584.61	18148.19	1744.99	20870.69	2867.27	17242.13	1905.34
6-Methyl-DL-tryptophan	7906.10	1445.88	7138.71	1212.83	8884.66	2205.62	3646.51	924.93	3519.16	1381.05	2941.80	618.20
6-Phosphogluconic acid	49847.82	3509.13	48922.97	2080.57	54711.93	11264.58	38433.65	5247.23	42025.45	3944.00	38172.88	2786.27
7-ketocholesterol	626081.00	290451.81	258930.40	50953.48	396226.00	275342.90	260433.50	181162.90	286017.00	160097.82	766422.60	355187.40
Acetyl-L-glutamine	421417.90	4592.98	343236.80	31288.31	402769.20	74024.44	541106.20	73565.33	294844.50	28710.73	412553.10	7246.80
Acetyl-L-tyrosine	577.46	55.54	524.99	23.60	902.51	427.03	683.23	284.44	762.03	179.39	663.13	143.65
Acetylcarnitine	96278.65	47871.24	309490.48	216516.23	37751.12	13014.35	44040.47	22725.12	49142.91	10153.33	73873.97	9619.88
Acetylglucosamine	35792.35	1114.82	43466.42	3819.80	45440.10	7655.95	49662.05	2411.92	50202.62	8308.47	42210.25	2066.21
Acetylglycine	800.93	210.28	2056.75	458.89	1704.27	391.46	3151.43	1024.83	2710.94	375.25	2492.14	414.96
Acetylmornithine	414.28	112.92	315.64	101.78	7191.57	6339.76	846.04	222.95	713.39	328.30	619.23	163.63
Adenine	4109460.00	30681.72	4026063.00	307848.06	3498223.00	1021057.7 4	3605038.00	216275.89	4041150.00	257765.77	3575144.00	282188.30
Adenosine	14047201.0 0	561783.20	13810475.0 0	393576.60	12650378.0 0	3131096.7 0	17310018.0 0	750711.70	18553996.0 0	806509.60	18240100.0 0	1077804.0 0
Adenosyl-L-homocysteine	1885562.00	184630.40	1667549.00	115727.90	1237763.00	225905.70	2207777.00	234580.60	2237128.00	215828.70	2348046.00	150499.90
Adenylosuccinate	48700.27	10046.76	52126.27	8886.22	61350.75	10415.64	41404.05	13608.01	43763.25	4262.27	25869.74	3286.32
ADP	1227454.40	72806.58	1295774.50	122570.32	1220834.70	246724.41	1112406.80	78349.60	1052364.80	25457.16	969850.80	39450.59
Agmatine	458.78	50.08	468.58	110.77	530.83	270.92	1511.67	303.83	1459.76	195.66	1222.83	388.28
Alanine	1284340.00	79530.24	1095190.80	78658.57	825486.50	190407.34	992720.20	108861.45	1055105.10	44082.82	894948.60	52072.64
Allantoin	82229.45	9229.62	63180.02	9791.84	79924.46	3032.74	85984.03	5725.05	71842.89	8557.93	74597.93	6781.48
Allopurinol	2748334.00	187995.18	2643310.00	89861.23	2289477.00	374189.92	2539631.00	88266.52	2294731.00	183893.31	2362783.00	152572.23
alpha-Hydroxyisovaleric acid	3993.59	361.84	4818.09	785.48	5957.05	1138.57	2684.97	328.02	2499.68	130.23	2484.73	512.21
alpha-ketoisovaleric acid	848.49	167.42	408.74	204.41	1265.28	48.35	752.68	78.88	832.87	175.46	1144.56	277.35
AMP	676570.90	68880.32	616568.70	45724.67	470219.90	50047.07	574608.20	44947.20	632074.00	32231.77	490083.10	33231.11
Anthranilic acid	1806.44	174.53	2003.62	50.53	1723.08	409.73	1114.98	260.61	2023.43	219.45	2046.61	158.51
Arginine	28250.70	1963.82	22795.25	5157.29	290102.60	262798.81	33772.90	6386.55	24415.81	4183.47	20787.46	1447.00
Asparagine	352811.00	1194.56	347859.80	44470.38	383980.80	52818.01	332473.00	52057.54	347479.80	36034.36	306716.30	20775.18
Aspartic acid	656596.10	74448.63	536358.00	66380.26	600755.10	112909.71	870141.90	148036.02	872759.20	59311.56	681745.40	92424.52

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Benzoic acid	6719.02	190.34	6213.79	240.51	5466.16	319.74	4294.63	339.43	4284.93	377.91	5982.04	764.56
Betaine	28144517.0 0	3940795.0 0	28393369.0 0	1024415.0 0	19848582.0 0	1734751.0 0	18964947.0 0	3910107.0 0	17737798.0 0	2834244.0 0	15191968.0 0	1306973.0 0
Cadaverine	1325.35	615.18	1509.17	370.31	1406.89	638.02	1291.45	229.96	1936.46	375.33	1508.14	593.96
Capric acid	2809.83	218.67	1964.30	434.41	2369.99	613.21	2210.39	173.00	1603.61	250.02	1431.77	110.07
Carnitine	4989271.00	371503.55	4466346.00	106681.06	2844579.00	547363.27	2693969.00	304531.52	3230880.00	50774.85	2583381.00	189724.93
Carnosine	55034.37	12126.27	40412.48	23830.17	43060.15	17278.14	41028.26	15307.70	26070.28	3091.43	22459.03	690.57
Citrulline	1011.57	468.51	971.61	247.90	29049.89	27331.83	1560.00	826.66	1557.53	337.32	1737.69	560.89
Creatine	16255.42	1308.18	15516.83	619.07	16313.91	2802.58	12456.30	2251.55	13538.30	2097.97	11525.12	1044.11
Creatinine	10730.90	595.03	14824.89	3518.79	10874.26	2201.51	9755.47	1439.53	11044.29	858.77	11609.50	1757.80
Cystathionine	157553.99	12220.09	106810.77	16366.23	76046.80	18108.58	107487.34	19520.37	99832.48	10465.80	56482.61	15486.71
Cystine	2208.98	282.84	2294.00	407.79	5902.74	2133.77	3820.08	349.03	3667.79	223.28	2866.33	796.71
Cytidine	181277.70	4968.68	213370.10	5855.79	195299.10	31850.54	177678.80	11277.71	200880.30	14897.99	168098.60	23059.34
Cytosine	36806.82	1576.92	41638.06	1668.28	38544.07	6919.77	35477.47	3415.89	41969.21	3029.99	35859.44	4203.51
Dextrose	16522.61	737.87	16109.10	1641.10	15471.73	1218.22	17293.62	445.07	13612.45	1640.03	15096.41	617.66
Dimethylarginine	549768.40	17215.49	550908.70	79100.56	598939.40	104446.23	473886.50	84680.53	513187.90	78959.70	448355.90	51698.42
DOPA	13830.74	1199.48	17489.07	1287.22	11623.27	3308.50	17750.64	1965.19	18620.85	2354.17	12916.44	1059.04
F6P	428552.30	43434.78	466840.20	62140.92	383504.20	80376.43	452292.10	42409.82	455702.80	43673.91	687674.10	67278.46
Folic acid	4144.68	3501.39	982.25	428.00	2763.31	2002.06	2704.69	1808.88	3549.18	2717.37	942.43	852.03
Folinic acid	2454.66	700.12	1584.49	364.10	3132.18	1355.51	2807.42	463.99	1928.47	372.36	1259.02	335.88
G6P	470109.30	51957.30	453877.80	51565.46	387058.90	78775.15	460301.30	42103.45	478815.70	53785.81	705107.50	42373.44
Galactose	44017.56	2271.22	42464.86	4706.85	43906.85	5653.81	45767.03	224.64	38080.65	1665.01	41537.61	740.75
GDP	158236.10	9302.10	175006.20	16104.12	156348.60	24292.75	117019.20	6047.81	110303.90	10960.47	127757.80	10018.17
Gentisic acid	2412.35	558.19	2010.40	127.30	2948.69	1355.71	1116.00	399.53	1032.75	310.66	1960.24	609.03
Glucosamine	220920.80	12186.73	229143.00	18920.05	191773.30	38727.19	230990.00	13593.15	212093.40	2480.18	224771.80	36243.49
Glucuronic acid	46603.53	1962.76	44165.38	2949.29	41003.09	6573.17	19081.92	108.68	30621.99	1815.15	32500.89	2527.72
Glutaconic acid	2760.97	516.35	2452.57	363.90	2629.76	772.87	1136.26	143.89	1490.63	438.27	1570.12	112.04

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Glutamic acid	2767268.00	500947.60	4176371.00	425440.90	3315934.00	715756.20	2532622.00	131545.20	3411186.00	589598.70	3150964.00	535977.70
Glyceric acid	11323.05	1250.79	9876.97	446.49	10228.85	842.69	10061.22	1192.66	9507.43	471.67	11480.99	689.67
Glycerol-3-phosphate	68664.59	18446.41	67005.67	6265.28	71835.49	6878.52	147433.90	45017.23	118284.25	9507.46	156642.02	88229.54
Glycine	69644.26	1683.41	81198.04	3870.14	67288.96	6486.22	99726.52	3889.01	109956.70	6802.58	98162.36	4170.02
Glycocyanine	17519.86	2870.51	16361.43	2373.42	14978.53	3460.03	24407.22	4631.81	30978.88	5661.31	36772.26	4609.98
Guanine	97833.19	10258.54	90728.77	15803.99	86506.62	18877.00	110387.57	8375.21	95803.64	8306.06	110630.47	14984.01
Guanosine	255072.10	30925.70	252574.80	44429.35	229832.20	50986.77	289317.50	27335.27	245175.30	23480.52	284944.00	34028.01
HIAA	1501.28	351.11	1910.77	724.08	1370.48	445.03	1451.50	651.50	1234.29	461.09	1857.31	359.52
Histamine	25360.74	1700.53	29861.14	4390.38	25142.10	1776.74	26075.58	1223.79	24771.09	1780.85	25452.73	1957.55
Homoserine	754684.70	11185.30	708916.50	31935.07	716500.00	72845.09	722316.60	89338.30	764027.60	83994.23	592280.10	42286.69
Hydroxyproline	1811477.00	86148.72	1648657.00	192232.75	1838956.00	252817.98	1795908.00	248175.60	1799134.00	172161.08	1542018.00	45267.04
Hypoxanthine	2654353.00	198201.21	2575245.00	97324.69	2217063.00	372490.21	2455202.00	93048.60	2213471.00	189576.88	2277232.00	156775.81
Indole-3-acetic acid	3312.30	209.20	4772.61	807.52	4850.40	543.79	3017.38	424.92	3875.67	308.54	3144.87	161.61
Indole-3-lactic acid	2439.99	1600.82	1297.62	416.21	3244.53	1616.82	3385.76	1277.58	2108.12	824.30	1305.06	1017.09
Indole-3-pyruvic acid	1327.00	534.37	1082.05	335.39	1362.59	630.51	639.88	177.88	775.80	235.61	1626.18	718.02
Inosine	23694097.0 0	1796248.5 0	23420575.0 0	1011297.6 0	20552103.0 0	3593058.8 0	22400797.0 0	512769.50	19736851.0 0	1956989.8 0	20501452.0 0	1264520.2 0
Isobutyric acid	43142.63	6009.16	32921.58	7318.79	23056.19	1828.63	17102.90	5190.75	13611.88	1783.00	9790.71	2347.83
Isoleucine	12176627.0 0	620467.90	11087724.0 0	1284562.9 0	12292914.0 0	1570629.4 0	12006485.0 0	1627782.4 0	12181535.0 0	1215453.1 0	10422628.0 0	347469.10
Ketoisoleucine	3686.15	911.86	3121.63	371.08	4323.51	1138.83	1908.07	585.38	2583.62	949.69	2243.37	449.47
Ketoleucine	3631.99	656.87	2088.06	635.88	3810.89	475.94	1072.41	170.46	1760.48	440.53	1948.33	480.32
Kynurenine	1906.66	658.70	3201.57	1402.36	1737.72	120.16	1461.13	524.01	2139.97	575.56	2289.21	182.90
L-Alloisoleucine	13549511.0 0	714495.70	12256809.0 0	1452069.0 0	13636821.0 0	1781606.4 0	13406379.0 0	1853758.7 0	13504076.0 0	1381872.7 0	11556063.0 0	436366.10
Lactose	38023.25	4345.51	36676.50	5385.48	35462.22	5692.80	38788.74	976.04	29991.19	4585.36	31255.42	1236.61
Leucic acid	1003.27	99.40	1019.63	39.19	1527.85	587.40	347.26	19.82	215.52	54.55	939.11	142.82
Leucine	12030178.0 0	650558.90	10930467.0 0	1268818.8 0	12189013.0 0	1588366.6 0	11883817.0 0	1622782.8 0	12011885.0 0	1218631.7 0	10267413.0 0	345733.50

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Levulinic acid	905.72	227.24	588.97	30.77	1626.21	227.39	660.87	179.17	479.85	101.44	1146.55	134.43
Lysine	2173024.00	42557.45	2219040.00	232125.45	2159220.00	212342.11	2347548.00	393911.53	2113633.00	149332.77	2090613.00	108201.40
Malonic acid	5251.13	826.99	5747.17	1187.72	1735.75	1653.50	3734.04	804.67	6503.09	555.03	5650.88	2214.17
Mannose	21859.45	80.48	19305.31	1228.81	18653.69	67.98	21733.63	198.83	21150.80	580.12	21512.37	582.80
Methionine	558940.00	39908.94	607215.50	78704.04	543699.70	159225.58	547314.70	103343.12	538726.90	83405.90	310330.90	124911.68
methyl- α -D-galactopyranoside	3842.91	786.68	4586.55	458.10	4393.40	332.07	3672.52	632.90	3385.05	670.05	4211.52	370.98
Methylguanidine	43288.10	468.10	39243.08	1929.37	40957.09	3677.46	39439.68	4526.70	44900.55	5739.35	34961.04	1897.61
Mucic acid	5740.71	766.26	7581.56	1363.64	9173.60	2342.41	9048.34	1895.04	15861.76	3162.02	15551.11	1297.66
N-Acetyethanolamine	2947.51	433.71	2323.91	328.63	3209.59	1151.48	1726.73	120.93	1584.13	292.11	2364.05	578.84
N-Acetylneuraminic acid	14611.38	1887.22	18524.03	1631.78	13979.33	1368.70	22830.90	985.42	23802.01	1287.75	18919.75	1235.33
N-N-Dicyclohexylurea	16811.61	2370.16	12524.72	1961.82	13191.96	898.26	19907.55	4511.66	13091.70	4729.15	11698.35	2335.98
NAD	169140.20	12648.17	156438.00	5822.38	151599.00	35490.51	179951.00	27303.42	181158.30	10484.56	159715.60	12097.92
NADP	46827.04	5206.03	57686.92	1423.57	64968.23	16454.01	32964.87	3043.69	38296.98	1083.07	38939.29	2298.21
Nicotinamide	19787128.0 0	1050373.7 0	16952797.0 0	299228.50	13505044.0 0	2108469.6 0	18228704.0 0	1177154.5 0	16929529.0 0	995643.40	13841484.0 0	589900.10
Nicotinic-Acid	1774.37	465.54	2538.40	541.74	2540.24	873.28	2415.18	232.43	2431.31	55.64	2169.06	738.85
Nonadecanoic acid	445068.90	27578.83	500688.90	34795.25	454111.00	125640.22	603641.30	167233.14	369292.30	29429.02	411891.00	151833.58
Norleucine	11250557.0 0	598213.60	10145399.0 0	1187692.8 0	11355258.0 0	1458853.6 0	11092502.0 0	1515540.2 0	11241999.0 0	1148861.4 0	9555634.00	324211.50
Normetanephine	9837.44	1979.20	3853.84	2339.46	5926.73	2102.61	4977.49	955.62	5160.15	1918.00	4248.31	377.99
Norvaline	913982.20	19831.03	760975.10	112153.63	913533.40	72395.61	938080.10	81726.98	892549.60	77558.19	777677.60	6929.89
Ornithine	1863707.00	53299.39	1592573.00	340406.88	1393242.00	235928.77	1214880.00	158247.89	1083649.00	69753.37	1127473.00	104665.85
Oxalic acid	2149.93	510.71	2330.44	377.68	2702.68	357.68	1754.29	188.38	1815.93	98.63	1557.28	104.84
Palmitic acid	414097.60	64582.03	261542.30	21679.26	376199.40	46457.49	306245.30	23341.84	239364.50	22702.29	374391.10	39240.03
Pantothenic acid	356703.90	18858.76	372683.00	11960.40	322338.00	33934.96	335021.50	44675.52	322713.00	13563.15	330983.80	7091.32
Pentadecanoic acid	340413.10	51812.17	247014.40	12809.18	262170.10	37545.06	468951.10	101435.90	255833.50	15049.67	219928.90	46831.75
Phenylalanine	6927223.00	147119.30	6872757.00	819787.60	7280278.00	828741.10	6027570.00	929142.70	6313926.00	581914.40	5676429.00	323635.10

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Phenylglyoxylic acid	399.04	135.91	256.39	42.83	694.90	254.77	456.94	31.34	394.23	16.19	567.34	174.06
Phenylpyruvic acid	1023.31	276.90	955.60	97.32	1401.69	342.75	536.71	164.80	1084.50	190.71	912.99	145.22
Phthalic acid	59072.07	5344.08	52084.14	3431.37	48576.87	3312.70	35899.13	5686.58	33847.06	2509.51	48997.16	4496.74
Picolinic acid	2248.55	200.87	2123.11	276.21	2392.87	1000.41	2171.58	445.45	1699.08	609.81	2357.00	931.02
Pipecolic acid	5285465.00	218245.69	5778808.00	132871.94	5686285.00	886746.40	4898692.00	434373.66	4941564.00	243293.83	4915368.00	17768.19
Proline	821186.10	65020.13	735543.20	64150.48	744333.90	50109.03	751352.80	85452.95	773543.50	31932.05	718433.70	18733.07
Putrescine	17516.58	1457.40	21464.76	6390.11	14092.16	2815.50	23619.50	6666.75	22099.09	4108.41	19655.43	165.71
Pyroglutamic acid	5100458.00	204241.41	5513453.00	145073.25	5451450.00	854163.78	4648655.00	386444.35	4730577.00	255749.36	4746774.00	63412.64
Pyruvate	12520.07	1551.34	9851.78	1723.88	12319.93	2003.59	11244.95	1659.60	11267.70	2143.32	14037.11	4424.39
Raffinose	3481.52	258.03	3575.09	1239.12	2989.90	806.09	3453.86	561.03	3379.86	803.86	3278.09	585.74
Sarcosine	1046579.80	75151.79	900824.00	68913.30	687203.40	159076.84	823894.90	87493.34	861428.30	47931.27	733439.60	35725.82
Sebacic acid	2550.53	134.90	2141.71	264.38	2707.88	169.95	2286.96	105.22	2452.87	157.73	2379.03	136.79
Serine	568061.60	33717.45	596208.40	48416.17	598971.00	68500.77	618789.80	74936.74	577123.50	36838.62	468030.30	31174.68
Serotonin	50214.43	6017.88	60646.48	5556.57	50360.85	10111.93	43697.88	1375.66	35487.35	3633.71	47463.04	5044.80
Shikimic acid	3748.89	126.93	3780.22	284.25	2288.36	478.13	2535.51	140.96	3291.82	786.93	2562.75	455.96
Sorbitol	4568.84	738.34	3987.15	218.95	4365.62	89.85	7434.26	880.26	6253.32	1217.26	8668.01	2106.77
Stearic acid	227888.20	43509.23	157743.30	9095.30	185398.80	6060.42	163029.70	16464.27	111519.80	31567.45	196429.80	9580.18
Threonine	843734.30	12897.44	795684.00	37063.18	800878.50	78053.36	821224.30	103374.72	849442.80	91208.14	663362.40	50921.47
Thymine	3280.49	441.44	2882.95	232.86	3346.53	771.73	1387.27	213.11	1526.88	302.68	1471.03	107.00
TMAO	8331.75	558.15	7637.67	417.96	5242.08	1708.60	5556.45	736.97	5609.26	623.40	5065.13	650.25
Total.Glutathione	1457488.70	28810.29	1354721.20	62505.51	892231.30	150511.97	1092043.60	91871.89	1183554.70	82173.61	1249310.40	67691.39
Trehalose	7685.59	441.32	6811.87	1198.73	7032.20	1033.35	7357.51	499.14	5736.73	943.71	6033.44	297.08
Tryptophan	207942.30	129579.42	148049.70	19754.10	281967.40	166017.09	359078.60	136866.21	202036.20	73013.96	130411.90	100982.74
Tyrosine	374764.90	52032.17	342759.90	74058.72	311213.10	35411.97	378655.80	73847.12	338056.70	58399.67	264642.90	34427.56
UDP-GlcNAc	770538.10	54509.62	845846.30	62204.03	690954.40	138456.86	823431.90	46010.97	684889.70	81135.03	681322.40	34349.28
Uracil	3286760.00	441041.00	3268934.00	198349.80	2875221.00	184941.40	3260353.00	464863.70	3313778.00	110090.30	3998974.00	390634.40

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Urate	25680.85	1707.63	31987.15	3275.45	19278.11	827.81	51388.89	4867.14	61162.84	9082.99	48312.15	5491.98
Uridine	4464960.00	694049.70	4565126.00	221631.70	4095088.00	223966.00	4483395.00	547250.10	4513090.00	231334.60	5255109.00	585878.10
Urocanic acid	28463.56	8842.13	77633.71	49050.45	24895.04	7314.60	38142.72	10613.84	23941.14	5086.55	16808.43	2984.24
Valine	957040.60	29170.95	799642.80	123967.63	961812.20	75542.99	992247.90	97460.90	940764.30	88026.07	826754.30	10002.44
X5P	219707.70	9798.67	203045.80	811.37	202388.80	18941.56	221298.70	14551.83	214799.40	18405.91	232745.60	10212.44
Xanthosine	199924.10	37021.40	225343.60	29281.73	271719.20	50237.41	311368.90	34994.85	230378.10	53472.98	283231.00	45422.62

Supplemental Table 4. Aqueous metabolites that were detected in SIC. Average relative abundance values and standard errors are shown.

Metabolite	CVCO	SE	CVBDE-47	SE	CVBDE-99	SE	GFCO	SE	GFBDE-47	SE	GFBDE-99	SE
2-3-Dihydroxybenzoic acid	1683.05	122.78	1036.51	134.23	953.42	112.16	1206.82	240.01	999.86	228.70	658.49	126.15
2-3-Pyridinedicarboxylic acid	3471.23	1057.25	4545.30	52.70	5039.99	135.98	5398.20	517.05	4496.29	1520.13	5751.77	256.77
2-aminoadipic acid	1451.16	605.95	594.10	130.38	792.75	260.34	1343.50	389.58	2290.08	123.14	1900.60	644.83
2-Deoxycytidine	149246.32	111894.36	37046.18	12424.81	44831.98	1983.24	23120.49	7634.35	36391.71	7956.65	25906.06	10963.73
2-deoxyguanosine	39892.98	27340.30	11776.02	3504.44	10509.99	1691.67	9973.15	3127.52	14440.19	2926.38	12340.79	5437.76
2-Hydroxybenzoic acid	897.98	101.70	724.31	100.43	976.31	131.50	1269.62	310.30	872.83	99.94	807.47	13.44
3-Aminoisobutyric acid	973.43	253.15	666.14	279.29	732.37	150.06	666.05	189.30	662.39	360.72	652.53	269.88
3-Hydroxykynurenine	1130.68	26.19	1696.00	102.25	1267.41	648.45	2044.50	276.00	2315.73	247.54	1378.08	176.09
3-Hydroxyphenylacetic acid	344.85	55.64	369.49	64.07	639.66	92.91	605.67	73.90	563.76	32.40	744.68	223.19
3-Indolepropionic acid	393.71	185.19	56.95	21.21	180.42	93.84	239.44	66.67	264.19	117.90	464.71	186.49
3-Indoxylsulfate	2294.97	1447.16	945.99	296.59	629.34	63.43	470.24	236.04	407.56	283.42	310.23	80.42
3-Methyl-2-oxovaleric acid	3817.70	808.83	4980.13	195.94	5732.81	683.88	5868.53	1137.19	5650.41	1211.43	5489.01	241.04
3-Methyladipic acid	1239.15	216.80	1437.59	244.04	1058.87	182.40	1507.93	255.26	1409.18	112.58	1261.00	119.13
3-Phenyllactic acid	127.02	48.16	306.64	75.99	237.24	75.05	762.61	352.80	3566.19	470.15	1576.48	612.90
4-Aminobutyric acid	2783.63	621.09	2217.58	421.91	3079.91	204.22	2172.13	186.68	2741.91	408.83	2371.02	932.31
4-Aminophenol	1691.07	174.94	1564.70	372.73	1528.64	174.52	357.74	110.48	1418.50	298.45	993.92	396.74
4-Hydroxybenzaldehyde	3058.98	748.41	4354.25	1150.17	3352.16	708.49	2782.11	1104.75	3776.65	362.07	3275.76	844.78
4-Hydroxybenzoic acid	871.80	183.82	1012.22	53.20	820.62	99.47	1396.11	211.78	793.02	134.15	702.33	112.07
4-Hydroxyphenylpyruvic acid	219.35	94.27	406.58	36.10	748.97	191.42	948.57	486.88	1122.50	81.60	831.76	311.75
4-Imidazoleacetic acid	6971.27	193.68	4944.61	932.54	5278.49	683.19	1694.75	486.17	1620.29	15.16	1183.37	554.35
4-Methoxyphenylacetic acid	45137.50	12780.09	66865.14	4054.29	69378.56	1077.64	66871.56	6741.63	48387.96	18048.06	65839.48	4234.39
4-Methyl-2-oxopentanoic acid	1752.83	370.56	2298.44	179.69	2567.72	260.72	2728.61	373.02	2729.83	486.75	2517.82	426.25

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4-Methylvaleric acid	13375.41	1008.72	14508.25	603.61	21152.76	4816.49	22548.64	2490.28	35929.87	4525.52	24697.46	1394.66
5-Aminolevulinic acid	6072979.00	762658.50	3838024.00	719591.60	4007779.00	149524.60	3018342.00	405066.90	4730769.00	268302.00	3556895.00	1224861.10
5-Hydroxytryptophan	865.31	308.65	449.86	139.41	277.83	110.39	248.46	166.73	349.33	232.66	461.76	258.94
5-Methylcytidine	1207.73	334.18	1197.56	312.62	2057.51	397.27	2332.71	1225.41	2995.52	215.94	2098.05	588.70
6-Hydroxynicotinic acid	20873.88	5292.25	21078.74	1924.74	18090.10	2873.90	141707.25	60789.01	305247.32	4500.52	271159.00	86101.61
6-Methyl-DL-tryptophan	6518.98	1086.35	4242.53	1253.83	4605.73	699.30	3919.02	272.45	5472.74	533.78	4323.57	1174.62
7-ketocholesterol	35960.57	1953.24	42761.84	2145.28	41259.77	224.87	38854.38	2862.06	42187.07	474.00	43780.27	1092.76
Acetyl-L-glutamine	108194.57	11627.28	63488.61	13244.48	62050.19	7004.23	36951.50	1142.70	47425.80	4059.36	43925.74	13588.06
Acetyl-L-tyrosine	180.10	88.09	282.92	88.96	346.79	18.24	244.40	16.34	629.39	59.48	500.78	58.44
Acetylcarnitine	53170.60	3969.42	45572.37	1794.56	52378.93	8010.67	17971.66	6153.06	12965.11	4061.39	11564.23	3492.99
Acetylglucosamine	2034.25	459.85	3756.81	1076.80	4011.03	781.55	75725.04	37656.16	113659.91	4197.12	107143.80	10718.63
Acetylmornithine	65953.52	11647.09	37071.70	7187.27	38167.75	2418.88	27727.06	2294.79	30450.15	1531.64	28267.91	7959.96
Adenine	45653.37	4345.71	94282.64	21077.76	97772.61	26227.13	1989909.06	1017292.78	2942189.54	496305.33	2104597.94	582458.12
Adenosine	88109.45	5995.31	226581.44	30718.93	246316.08	69356.52	3432153.16	1723022.93	3795530.25	504040.14	1962001.15	716413.78
Adenosyl-L-homocysteine	718.40	718.40	0.00	0.00	0.00	0.00	0.00	0.00	26.14	26.14	41.68	41.68
Agmatine	38447.07	8923.21	19197.25	2661.17	15813.75	2533.19	22880.98	2756.61	22671.05	1463.59	21908.39	5960.70
Alanine	110447.68	18075.96	74543.57	14626.80	75692.79	2874.04	68643.88	12679.67	117176.83	9611.37	108641.70	36172.12
Allantoin	1433.16	366.47	1149.97	229.02	4042.20	3401.58	3466.13	1568.65	9119.36	5349.97	7084.17	4594.09
Allopurinol	22238.63	6801.15	20528.24	1926.48	22553.57	6442.00	48288.62	27749.54	24566.16	4702.59	15520.53	6543.84
alpha-ketoisovaleric acid	660.02	194.36	608.27	69.74	602.55	156.05	1139.97	211.95	1486.39	241.54	1106.61	143.17
Anthranilic acid	1567.55	236.20	965.75	320.00	1164.19	102.92	1471.53	196.11	2019.96	57.84	1874.39	223.60
Arginine	3017438.00	573340.96	1642048.00	322122.58	1632599.00	172870.61	1188192.00	68541.31	1372468.00	86992.47	1263643.00	367493.19
Asparagine	261397.10	58830.76	103832.86	33007.68	100081.64	11314.14	45709.39	4100.52	51097.96	4764.17	33561.80	11690.51
Aspartic acid	221415.10	24274.03	199400.70	30273.11	193050.00	10782.05	245562.80	55618.54	502406.70	36626.77	502054.10	123978.26
Benzoic acid	4903.40	1307.12	7414.92	320.82	7160.73	23.65	7170.75	964.82	5777.79	2052.90	7034.04	269.85
Betaine	1750890.60	168456.54	1328224.70	99835.62	1558675.20	341930.90	1820219.90	330934.93	912008.10	115119.88	1170426.00	222931.55

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Cadaverine	4537.40	708.31	2392.28	196.12	2577.57	452.45	2385.73	346.67	3471.68	443.99	2862.93	661.41
Capric acid	3102.94	600.46	2298.26	501.78	2955.00	526.39	2551.85	885.92	5324.19	523.94	6293.64	2618.17
Carnitine	16637.88	3730.83	12933.78	1034.95	13698.96	1423.19	6293.58	1125.46	9428.66	377.21	16570.34	1778.94
cGMP	180.28	40.29	620.66	322.41	152.91	69.85	8562.43	5120.95	8646.47	330.75	6881.95	2546.19
Citraconic acid	3382.37	674.00	4470.31	450.07	4758.11	436.24	4918.21	724.32	4640.34	589.41	4927.39	154.80
Citrulline	299970.40	53419.85	163073.60	31476.04	164407.00	14074.72	120447.30	8884.01	132082.90	7589.17	126690.10	36457.46
Creatine	7989.50	869.24	2330.74	426.32	2699.17	326.01	2454.90	495.65	1681.06	348.80	2125.53	576.20
Creatinine	15224.97	5961.53	7529.02	461.95	13848.36	2015.14	16639.95	5883.17	16580.11	1988.62	26098.36	6056.32
Cystathionine	216.20	87.42	299.32	152.09	155.23	70.64	61.23	50.81	272.39	49.79	206.82	113.86
Cystine	42944.37	5193.42	21119.20	4841.56	24544.10	2819.84	6678.08	2404.38	4898.22	176.78	2949.42	386.16
Cytidine	165002.80	17713.04	172761.10	22016.96	212757.90	20482.62	505667.50	253247.28	542070.50	46915.27	302161.50	84091.72
Cytosine	32997.81	2378.49	38937.11	4724.51	43464.39	5196.54	112970.01	57768.67	124242.36	12178.02	69753.61	19823.31
D-Galacturonic acid	1780.89	662.55	572.65	82.10	538.00	70.63	1470.63	1201.82	0.00	0.00	897.50	639.97
Dextrose	477.44	192.58	1441.85	599.65	651.31	191.17	3462.39	627.24	3225.56	324.08	2144.49	1122.43
Dimethylarginine	38314.03	7333.65	28506.19	2713.00	23775.61	2311.18	39808.66	13359.29	64794.47	5779.32	63835.06	18604.39
DOPA	1032.99	92.36	772.55	455.62	524.89	39.80	1654.97	654.68	2484.13	295.25	2061.79	559.62
F6P	7113.73	1017.10	7765.99	1661.66	7152.08	450.10	2577.50	1193.01	7692.24	3024.24	6624.13	1545.64
Folinic acid	801.33	110.07	214.34	88.40	690.00	151.40	154.48	30.01	124.12	58.38	103.37	60.81
Fumarate	6695.28	648.11	8199.88	1270.07	10191.18	1968.37	8051.69	4870.34	1647.72	365.50	1908.14	755.17
G6P	7457.20	897.75	7801.59	1436.34	6839.31	930.65	2311.67	953.13	8238.06	3457.67	6557.17	1106.29
Galactose	2548.48	319.31	3503.34	1291.53	1993.35	653.76	8257.70	875.69	8950.48	855.39	5255.92	2442.50
Gentisic acid	1593.80	426.86	1723.09	121.12	1642.08	272.21	952.67	260.79	535.21	70.52	549.03	82.49
Glucosamine	66216.48	19587.72	46834.55	11790.15	66598.39	37133.64	78119.96	47017.59	72818.04	22993.57	71645.15	20619.58
Glucuronic acid	13952.28	805.50	20798.60	1735.09	15817.62	2479.10	6896.32	3667.58	5463.13	1071.08	3710.75	368.46
Glutaconic acid	3874.26	1030.26	4634.77	339.05	5055.34	402.07	5515.19	623.78	5171.16	1116.09	5157.71	480.37
Glutamic acid	1429348.60	186370.16	885692.70	170539.97	935463.10	34176.76	1303600.20	329303.47	2233105.50	155967.63	2212389.70	479732.32

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Glyceric acid	2547.11	64.64	2598.13	442.03	2114.50	451.78	5315.49	2475.74	12291.55	484.80	8552.78	1652.45
Glycerol-3-phosphate	13406.60	3185.08	15290.84	1241.16	17867.07	972.19	13375.98	2531.80	17742.77	276.40	17304.96	4633.41
Glycine	13429.40	2363.66	9040.12	2193.75	9168.19	571.01	7106.17	704.58	9309.06	256.51	8550.99	2157.88
Guanine	43054.46	15808.18	40670.77	4095.92	48165.11	10486.29	167155.38	88465.68	130675.03	23054.21	71324.52	29139.73
Guanosine	118750.60	43209.96	102065.60	12142.20	122100.20	27307.69	426000.10	215797.10	330111.50	54795.99	188680.80	75596.88
Homoserine	286157.70	43447.94	181403.50	37826.62	175503.20	5695.29	146337.90	29083.45	252122.80	13196.42	207026.20	68781.93
Hydroxyproline	1721021.90	234318.77	1059481.10	204254.72	1126440.90	47679.60	846093.10	121084.37	1323588.40	64213.01	999213.40	342299.27
Hypoxanthine	21496.50	5997.35	19956.45	2753.59	21404.47	4493.74	44463.82	26505.53	22740.37	3272.74	14723.12	5649.03
Indole-3-acetic acid	4553.58	370.60	2796.33	547.10	2591.87	99.82	3427.32	826.52	3421.28	321.67	3023.08	806.01
Indole-3-lactic acid	5311.08	1452.08	3227.26	618.38	3128.08	531.77	2008.07	88.57	2742.75	250.53	2030.33	654.27
Indole-3-pyruvic acid	1308.74	172.77	945.48	219.14	981.75	216.31	960.29	103.94	966.23	267.10	846.94	52.72
Inosine	239111.70	67310.67	213465.10	24032.33	230600.60	50842.80	438164.70	248184.07	252360.80	34496.32	178395.20	67681.61
Isobutyric acid	845.72	528.74	1326.85	136.84	1287.02	659.30	2064.94	1432.67	3211.34	1043.44	1243.62	532.89
Isoleucine	11576745.00	1482768.20	7268778.00	1356582.40	7612552.00	248995.40	5722851.00	772582.00	8971662.00	459249.60	6756036.00	2323763.50
Isovaleric acid	2083.80	333.29	2623.87	135.41	3569.66	244.52	3548.28	455.98	2946.15	859.75	10035.87	7518.85
Ketoisoleucine	5225.96	1195.23	6482.45	634.73	7903.57	893.40	8480.08	1090.90	7415.54	1659.61	7938.26	255.54
Ketoleucine	4017.92	979.33	5265.94	248.18	6389.56	553.44	6560.42	1146.59	5652.31	1126.32	5969.95	636.04
L-Alloisoleucine	12850676.00	1574471.90	8135699.00	1528791.80	8562657.00	245741.30	6401741.00	876003.80	10010314.00	535785.10	7543826.00	2589981.90
Lactose	644.57	285.91	2393.68	1192.19	1040.45	240.12	4981.38	667.09	6608.74	1207.60	4966.08	2083.71
Leucic acid	47.04	10.59	114.39	21.27	183.45	77.09	354.81	122.47	2750.65	914.60	1713.65	720.46
Leucine	11409329.00	1388491.80	7169646.00	1343197.10	7539092.00	232976.70	5651859.00	756689.10	8813863.00	449127.50	6651887.00	2266941.90
Levulinic acid	607.13	172.53	569.79	83.56	504.51	26.33	966.54	165.66	835.53	85.45	459.25	34.60
Lysine	1435723.70	215738.49	955560.80	144656.56	965716.60	44129.72	1079669.20	318355.19	2048293.70	82046.64	1929208.20	576691.14
Mannose	938.31	88.46	1001.76	229.89	790.94	326.17	2781.56	1186.20	4878.17	534.11	2885.56	1059.86
Methionine	589386.60	75429.96	368246.60	70760.76	398658.50	29105.94	402784.60	138419.10	833308.00	82326.86	670728.90	224073.65
Methylguanidine	14702.64	1468.99	9451.92	2789.61	8571.48	410.32	7048.58	941.11	12673.84	1322.31	10484.97	3563.97

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N-Acetyethanolamine	1180.01	138.30	728.93	244.72	570.50	167.60	1771.23	572.08	2748.06	287.43	2512.13	501.80
N-Acetylneuraminic acid	1796.40	259.97	1408.89	530.56	1625.46	555.68	9925.97	4979.31	10453.75	434.06	14573.38	1191.52
N-N-Dicyclohexylurea	18268.17	2614.40	19964.98	1040.79	19623.44	1727.87	19180.86	2454.28	14962.30	1142.97	16995.90	1020.35
NAD	244.27	19.08	618.77	66.93	375.16	222.06	944.18	448.33	3212.14	398.57	1999.12	393.80
Nicotinamide	48187.16	38742.62	10940.14	1543.74	12716.69	4030.87	12205.62	865.15	19282.07	3762.38	20803.46	4592.79
Nicotinic-Acid	9086.82	1150.02	10059.80	775.14	10381.88	1950.19	22159.36	7396.05	44287.45	2052.94	38106.11	9821.84
Nonadecanoic acid	635603.50	136918.76	466113.60	81042.70	524218.40	38583.76	1044827.40	612016.23	373490.40	40325.32	404978.20	27472.58
Norleucine	10654976.00	1352194.40	6701028.00	1266816.50	6994193.00	229020.10	5289766.00	719626.80	8206609.00	449129.70	6166355.00	2107227.30
Norvaline	1133867.30	169146.81	741604.20	151689.43	751888.60	22588.24	596118.80	101870.91	934012.20	45830.15	702442.10	262226.43
Ornithine	71833.23	22562.04	49066.95	7275.43	51253.59	4926.76	48212.63	17109.55	125441.24	5405.20	112282.41	27513.57
Oxalic acid	16619.06	3689.55	15405.50	3139.58	15331.89	3935.22	17373.62	4197.04	20482.27	3220.89	20607.77	3651.49
p-Coumaric acid	8844.49	1767.47	4969.36	816.58	7130.48	2288.08	5608.51	1798.68	5680.15	1444.03	13391.37	5653.67
Palmitic acid	393963.80	119814.03	715668.90	12225.16	607062.40	4339.16	516060.50	176426.29	412193.50	21725.60	356053.20	51994.66
Pantothenic acid	58143.95	11698.76	44529.82	4127.29	53487.21	5700.24	146844.23	63762.66	289768.54	10852.00	296501.66	49921.81
Pentadecanoic acid	826175.30	358913.76	376503.00	10798.61	303275.50	9836.66	322577.00	9506.02	396205.20	74296.52	334974.00	34666.69
Phenylalanine	7939498.00	1084143.62	4995707.00	773426.19	5315128.00	196732.67	3382414.00	86573.86	4899530.00	313237.43	3967829.00	1317855.60
Phenylglyoxylic acid	369.49	71.67	449.10	84.25	400.40	111.80	830.79	318.92	960.52	238.00	680.08	227.13
Phenylpyruvic acid	686.17	47.15	1005.73	56.31	1047.63	299.53	1214.36	409.42	2342.41	347.09	1131.59	322.91
Phthalic acid	38797.18	12370.49	60249.44	4282.14	64575.81	1070.09	64922.91	6185.94	46100.92	17143.88	62640.63	4008.36
Picolinic acid	10984.82	1609.18	9925.23	874.56	11084.34	1658.88	27414.59	9134.07	48927.84	2344.85	40967.62	11766.57
Pipecolic acid	649895.80	102424.22	374119.60	83865.19	373084.30	5027.85	351320.90	39373.91	532967.00	50238.32	476926.00	131774.10
Proline	883066.70	220254.60	432768.10	89212.56	492541.70	6482.41	459906.20	102588.13	768263.90	47579.30	690287.50	252899.51
Putrescine	3240.39	911.75	1833.28	434.52	2132.42	1752.23	2605.04	183.16	3655.38	343.37	3382.00	947.69
Pyroglutamic acid	626612.20	96350.94	365385.80	89632.20	361721.00	1861.43	336999.80	39506.29	518929.30	46079.23	458283.90	126433.51
Pyruvate	648.12	128.46	537.30	50.68	464.03	69.59	717.69	102.05	1262.10	322.24	1089.75	112.86
Raffinose	29758.56	13697.75	7832.22	1785.97	9427.25	2874.95	4592.08	3958.12	513.18	117.99	406.41	79.86

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Sarcosine	96008.49	16985.19	60577.93	11528.08	62266.69	2236.61	59160.49	12736.81	101471.29	6937.57	89489.15	28595.91
Sebacic acid	1622.67	198.88	1405.54	165.07	1276.04	185.44	1267.42	171.44	1539.07	59.56	1838.33	109.33
Serine	438255.60	72489.97	305429.80	75827.47	289223.70	6948.15	231439.70	34242.07	346952.50	20564.20	285893.50	88469.26
Serotonin	23395.28	13818.31	46344.94	11792.45	64067.88	13302.78	89286.13	40067.10	197402.35	14627.58	215227.52	61396.32
Shikimic acid	571.82	284.26	105.77	29.96	120.52	35.56	126.26	59.51	152.91	10.72	174.67	102.57
Sorbitol	13948.84	1806.97	2667.09	500.94	2502.68	806.63	1266.65	875.81	556.98	37.90	432.15	69.91
Stearic acid	144219.90	25658.97	154091.30	8288.45	161295.90	2121.76	174570.90	13845.55	175325.50	15855.78	101942.70	6872.10
Threonine	322385.20	50164.59	200522.10	41402.40	199215.50	1737.27	163285.70	36151.04	285808.50	14931.46	235994.50	80347.88
Thymine	4063.58	1980.39	2698.95	216.92	2066.50	351.78	4362.89	1754.06	8624.32	977.65	9593.24	3536.50
Total.Glutathione	3415.16	539.53	4018.13	579.24	3455.21	537.57	1934.61	832.46	2072.61	563.85	3720.04	678.74
Tryptophan	570556.60	105804.30	329499.10	57861.67	349158.70	21416.77	203261.90	6258.80	292879.00	11449.99	241575.40	83119.65
Tyrosine	746968.70	99422.01	462366.00	58353.80	500935.90	12205.02	370530.50	38870.03	588229.70	25616.29	504498.60	166580.56
Uracil	227016.30	42936.68	313424.20	53117.90	304254.60	32192.87	897849.70	441996.75	921152.00	127509.13	713270.60	168372.13
Urate	14082.55	2476.46	14254.65	3204.33	17263.30	1088.22	15526.36	3710.60	16204.30	1519.38	14183.91	3067.38
Uridine	294978.00	50922.77	406204.10	63699.98	421402.00	58354.79	1256054.50	598213.95	1238135.20	160200.37	964642.20	202776.82
Urocanic acid	192309.60	52461.33	200203.90	23242.89	172958.30	28012.39	1340658.10	577402.33	2935776.00	4191.84	2595459.70	831507.33
Valeric acid	2083.80	333.29	2623.87	135.41	3569.66	244.52	3548.28	455.98	3015.52	867.56	10035.87	7518.85
Valine	1190688.20	165802.18	782134.40	164951.00	793071.30	24763.76	626275.40	106812.82	981800.10	45406.02	748014.20	273635.36
Xanthosine	6094.82	2349.76	5052.56	396.39	5835.50	1201.25	22328.14	11848.40	17212.78	3108.21	10057.98	4490.50

Supplemental Table 5. Aqueous metabolites that were detected in LIC. Average relative abundance values and standard errors are shown.

Metabolite	CVCO	SE	CVBDE-47	SE	CVBDE-99	SE	GFCO	SE	GFBDE-47	SE	GFBDE-99	SE
2-3-Dihydroxybenzoic acid	2495.60	305.69	2661.71	396.65	2060.50	343.30	2004.24	997.48	381.74	116.08	465.08	186.76
2-3-Pyridinedicarboxylic acid	4653.08	695.28	4947.41	165.92	5638.86	60.56	4065.26	1168.65	1562.30	235.40	1493.10	207.66
2-aminoadipic acid	4506.08	519.59	6642.08	1171.31	4474.92	1179.86	4144.40	1193.75	2141.27	588.86	2823.29	724.85
2-Deoxycytidine	163164.80	23684.85	194383.10	17959.78	212812.30	11290.53	239874.20	73306.31	496559.60	56648.42	214267.90	56219.20
2-deoxyguanosine	96384.93	19363.86	117701.95	10387.96	100630.56	10418.88	96826.32	9301.47	115868.36	8400.54	47215.48	16316.91
2-Hydroxybenzoic acid	1572.73	504.27	1924.12	91.06	1875.99	636.17	1836.32	458.24	895.37	104.10	913.84	123.28
3-Aminoisobutyric acid	1718.55	295.75	1845.73	171.88	1937.90	96.53	1316.76	527.04	798.44	141.18	622.75	99.66
3-Hydroxykynurenine	2084.27	588.78	2259.63	264.88	2266.06	265.35	1801.39	63.97	1899.97	158.93	2052.43	203.88
3-Hydroxyphenylacetic acid	681.62	191.22	574.19	205.31	1132.07	131.64	732.79	112.38	493.56	59.98	463.17	96.57
3-Indolepropionic acid	369.74	81.43	313.25	122.15	148.41	104.83	216.84	110.44	133.74	81.77	215.86	41.88
3-Indoxylsulfate	1080.85	1011.20	46.58	33.24	89.77	39.15	62.13	38.47	200.34	99.61	151.06	48.69
3-Methyl-2-oxovaleric acid	4086.52	703.85	3771.47	530.26	4676.58	526.94	3033.82	627.30	2213.94	185.06	2931.21	265.12
3-Methyladipic acid	1520.17	99.90	1378.33	211.99	1276.55	139.42	1206.45	230.99	1259.63	38.51	992.64	136.92
3-Phenyllactic acid	52.97	21.38	60.14	26.02	97.04	55.60	47.32	27.31	11.23	7.88	25.88	24.09
4-Aminobutyric acid	6343.09	1109.95	7281.46	1423.19	6831.53	886.52	5663.17	1785.81	2477.03	240.57	2393.70	332.37
4-Aminophenol	6054.00	563.56	5919.33	764.26	6729.12	1257.40	7259.77	1528.66	4515.62	258.17	4428.43	543.00
4-Hydroxybenzaldehyde	5723.60	518.58	11966.62	4153.90	9578.45	2478.81	7895.19	3335.91	3844.03	1117.60	2460.12	676.17
4-Hydroxybenzoic acid	1544.25	585.06	1666.33	158.16	1823.64	476.30	1525.73	482.35	778.50	181.14	787.94	69.05
4-Hydroxyphenylpyruvic acid	421.84	65.87	318.57	117.11	349.49	105.09	393.94	138.64	16.54	16.54	142.63	51.66
4-Imidazoleacetic acid	12606.25	240.62	15952.77	552.95	15983.49	1261.06	11501.76	2296.50	6494.38	471.73	5647.11	341.91
4-Methoxyphenylacetic acid	41997.79	352.56	39612.06	2242.58	46575.66	5248.48	34231.99	5205.15	25919.01	994.19	23283.60	1337.70
4-Methyl-2-oxopentanoic acid	2331.77	394.04	1976.55	19.29	1727.51	222.00	1486.33	271.31	1086.96	159.19	1493.65	135.06

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4-Methylvaleric acid	28403.51	6204.80	19256.49	1293.73	18857.17	2270.51	18813.76	4513.57	15254.48	564.01	14832.51	642.28
5-Aminolevulinic acid	3046085.00	436678.70	3388054.00	332935.70	3891716.00	695301.40	5183512.00	1275809.10	8968358.00	598171.70	9573837.00	957398.10
5-Hydroxytryptophan	1869.59	114.30	1729.42	277.05	2093.47	274.60	1450.26	334.16	1596.77	235.97	1660.87	251.77
5-Methylcytidine	4843.42	1020.45	5106.86	392.57	5282.57	576.41	3398.30	1295.10	1204.70	174.47	1189.32	447.67
6-Hydroxynicotinic acid	22520.05	2814.24	24764.00	1049.90	30714.84	2021.48	20367.29	3767.33	15815.68	331.80	16126.41	3933.26
6-Methyl-DL-tryptophan	5587.99	851.67	8087.55	163.45	7102.83	755.18	8047.29	813.22	6956.89	701.30	7492.32	1074.20
7-ketocholesterol	53267.55	251.65	50941.57	5449.77	41394.03	8820.87	49108.56	3580.58	37166.14	5815.11	28008.16	5094.46
Acetyl-L-glutamine	465584.80	21203.04	543763.30	16150.06	501765.60	24516.70	402983.00	131199.80	144578.60	8264.65	157755.40	16085.71
Acetyl-L-tyrosine	866.51	106.14	902.86	274.96	993.26	168.52	549.84	280.19	64.71	13.28	38.48	21.61
Acetylcarnitine	40333.67	4482.68	44070.84	9344.12	49696.01	4669.77	50821.32	6888.70	118781.10	20248.81	105079.43	8208.33
Acetylglucosamine	11128.08	781.63	13467.60	1620.09	13593.49	720.42	10368.01	2465.71	10154.26	1326.72	8837.72	1363.15
Acetylmornithine	64295.26	5250.44	73477.85	725.23	81046.55	7928.17	86646.66	6772.91	89749.60	5068.50	99884.39	8637.09
Adenine	136886.70	11765.45	126866.97	18007.95	129649.23	15731.56	121759.05	26205.87	41565.84	5405.89	46707.09	9160.22
Adenosine	392398.50	36964.71	324669.70	47123.13	336814.00	46329.15	328352.80	90545.82	160392.80	20998.69	136046.50	32303.87
Adenosyl-L-homocysteine	1667.08	169.78	1449.71	333.67	1966.70	256.36	1658.63	359.84	3210.38	608.65	3436.16	631.49
Agmatine	33949.64	7581.63	38074.09	2515.26	46881.23	1182.11	39882.36	1448.22	32875.13	2565.55	33952.80	1701.71
Alanine	88349.66	10210.55	102694.02	9090.87	96467.84	13402.97	127651.27	6239.79	167629.75	13241.73	173357.94	21440.92
Allantoin	5705.95	535.45	7555.25	1381.93	6206.68	1045.98	4021.15	1454.29	13084.64	5602.67	6753.85	4585.95
Allopurinol	54262.48	5228.34	59298.20	7659.74	59955.09	3796.39	45576.79	8080.59	32051.19	3525.22	39367.25	5219.54
alpha-ketoisovaleric acid	703.71	147.99	423.49	71.82	460.05	23.51	406.16	58.44	664.13	121.79	396.09	46.26
Anthranilic acid	2243.44	161.03	2740.55	187.96	2681.18	273.56	2302.06	623.82	1542.84	230.54	1629.68	355.59
Arginine	2739352.00	188551.88	3131471.00	76346.16	3422472.00	342072.01	3788771.00	338539.35	4065228.00	174578.95	4390339.00	356693.04
Asparagine	655912.40	15954.32	807991.10	45628.75	778330.20	91751.52	721228.80	176640.76	456443.80	40616.99	457954.40	60232.95
Aspartic acid	309624.70	6983.40	391218.60	17828.00	398882.30	22565.66	368238.10	45042.82	406137.70	48175.73	381960.80	57298.90
Benzoic acid	6563.62	294.52	5931.34	145.35	6826.84	422.33	5018.58	1008.24	3162.65	140.08	2994.65	261.40
Betaine	616834.40	103910.09	609798.20	28066.32	761088.10	60039.08	859428.70	124172.10	2015790.40	112382.67	1467210.90	16069.62

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Cadaverine	10301.82	3660.82	13221.40	1849.37	16054.57	1718.51	11425.42	2851.74	4301.74	661.13	4180.98	890.72
Capric acid	1112.40	34.21	1319.42	386.32	1427.10	694.31	2202.40	691.83	4535.49	358.07	3665.90	1635.96
Carnitine	50423.99	3917.30	50591.36	3355.75	55456.64	5888.86	48697.07	14784.95	37920.19	6539.31	34540.27	3335.99
cGMP	1004.33	63.96	898.78	125.71	843.25	234.10	1048.60	386.80	512.02	258.14	232.91	66.15
Citraconic acid	4912.39	806.60	3562.67	166.33	4824.99	300.24	3707.23	853.67	2519.88	185.47	2653.51	71.82
Citrulline	287339.70	17649.63	331787.00	3345.85	361679.70	38807.56	386711.60	28826.61	406406.00	15161.59	438098.90	37632.56
Creatine	19606.81	416.74	24802.91	2343.92	27928.11	4351.08	16813.31	4351.12	10418.10	1339.36	8519.93	809.49
Creatinine	11867.04	2723.63	11158.96	1019.40	9619.12	2372.63	8343.13	577.29	7872.11	1799.13	9425.10	1969.98
Cystathionine	3831.50	597.98	4710.13	443.66	4181.41	122.75	2787.93	1389.03	177.94	5.48	256.11	183.66
Cystine	29151.71	2870.31	34561.00	2310.13	27317.88	5666.36	34312.17	1087.94	79083.58	11957.16	68395.80	13676.15
Cytidine	160076.50	23196.47	128942.90	18046.70	124591.60	14253.35	152592.60	14332.87	226587.30	32799.58	262590.80	58791.54
Cytosine	42788.33	5888.81	29937.80	3763.54	31315.30	5593.27	34781.78	1606.34	38291.40	4431.90	49741.22	11466.80
D-Galacturonic acid	10444.61	1419.57	12055.37	720.39	11616.56	592.68	9220.03	3130.93	2317.26	235.88	2654.94	813.07
Dextrose	151.69	36.03	274.44	20.58	247.00	65.10	268.57	73.59	103.27	49.61	80.24	18.03
Dimethylarginine	333807.82	18955.74	379983.95	16233.40	380297.51	7438.19	269289.81	107210.19	75679.26	11656.23	75516.88	12113.45
DOPA	6485.84	894.25	7824.24	1088.15	8690.55	623.44	5896.46	2045.37	1452.86	333.98	2141.69	238.29
F6P	785.43	125.85	1358.67	109.52	1123.12	203.26	2096.02	1256.31	5143.65	901.88	3437.57	1543.43
Folinic acid	2963.98	464.30	4382.63	204.59	6035.97	707.90	3151.43	900.60	884.42	312.96	1214.25	134.63
Fumarate	1482.93	258.36	1200.79	92.17	1152.93	157.12	2125.72	779.58	3185.65	379.12	3145.14	710.21
G6P	651.40	149.75	1254.04	125.47	1109.68	273.17	1769.36	973.94	5629.57	1046.69	3290.73	1342.05
Galactose	9449.75	922.29	10973.08	430.91	10198.65	161.17	8093.59	2716.39	2567.79	163.11	2670.70	613.59
Gentisic acid	6181.80	1058.44	6642.84	1092.79	5325.70	635.84	6052.93	2763.99	587.40	136.51	662.04	211.18
Glucosamine	259020.67	35127.87	294227.29	12483.09	279023.40	8888.61	206933.34	58267.67	67345.44	5566.72	72818.02	18377.14
Glucuronic acid	49138.21	1884.34	61944.14	7131.30	65464.01	1829.05	48742.84	17897.84	14990.41	1607.59	16280.06	752.49
Glutaconic acid	4061.22	540.32	3236.85	235.04	3941.30	483.88	3100.87	762.35	2292.62	114.56	2605.51	565.68
Glutamic acid	3465472.00	75417.77	4581805.00	230829.54	4419747.00	311183.23	3643456.00	984738.61	2445501.00	235967.31	2347442.00	336632.95

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Glyceric acid	2173.22	77.30	1770.98	207.97	2183.93	232.11	2128.96	235.75	2232.33	190.85	1687.17	169.24
Glycerol-3-phosphate	2822.65	652.52	2411.86	772.33	1234.38	707.65	3714.66	1073.50	5943.22	838.69	7727.43	360.75
Glycine	21368.81	1923.33	23852.58	1779.98	23758.49	2252.69	21937.20	4453.97	20454.29	1573.89	20358.98	3087.34
Guanine	140395.53	11345.82	160873.80	16361.39	142952.77	9594.08	115639.12	26861.61	70595.23	9574.00	92627.73	18753.59
Guanosine	358209.80	28803.44	413092.80	33647.73	356089.20	11357.78	306626.80	68532.23	188393.90	27831.96	246617.40	50518.52
Homoserine	1020361.90	16115.69	1214200.40	82686.37	1155495.70	95690.03	981401.50	288587.35	516422.00	49157.52	528738.30	86125.24
Hydroxyproline	859551.40	120505.68	959832.80	95743.43	1095030.60	197426.72	1482797.10	372195.36	2578665.80	191472.05	2772241.80	282520.78
Hypoxanthine	53316.47	4864.66	56926.77	6731.63	53519.71	2099.79	43259.49	8538.52	32071.26	3621.52	36290.51	4135.39
Indole-3-acetic acid	4522.46	383.28	4565.58	197.17	4081.97	303.21	5039.63	353.12	4924.33	479.58	6444.00	525.09
Indole-3-lactic acid	4825.25	367.78	5730.42	204.48	5623.09	659.68	6736.31	1018.47	9532.96	967.15	10840.37	1182.89
Indole-3-pyruvic acid	2305.11	155.03	1419.99	132.74	1798.38	205.76	1406.78	45.84	872.01	126.06	956.59	146.12
Inosine	517868.60	47670.06	578891.60	67721.35	553791.10	31420.27	455489.10	50398.04	367049.20	32767.07	425056.80	44514.93
Isobutyric acid	2645.57	874.47	3846.79	1258.65	2561.51	1305.29	3383.07	1752.35	84.51	84.51	0.00	0.00
Isoleucine	5846123.00	813795.40	6452984.00	630029.40	7423375.00	1283506.40	9844442.00	2372524.70	16841781.00	1215329.60	18086080.00	1876430.60
Isovaleric acid	1335.91	127.57	1147.06	221.85	1229.89	197.95	1487.91	600.00	3296.35	389.30	2422.29	666.99
Ketoleucine	5625.64	713.97	4556.33	742.35	5265.69	772.61	4395.98	997.51	3366.66	164.26	3455.00	704.52
Ketoleucine	4777.10	735.71	3387.72	557.76	4219.23	387.34	3384.27	872.66	2729.82	259.38	2859.11	281.77
L-Alloisoleucine	6502557.00	932310.90	7236463.00	698980.70	8202470.00	1422894.00	10932456.00	2622594.20	18643299.00	1276000.50	20010892.00	2005231.20
Lactose	110.46	55.35	239.99	46.01	219.90	34.75	223.06	115.68	66.00	39.50	94.99	36.89
Leucic acid	71.95	6.38	91.93	35.07	123.73	46.84	31.53	18.07	2.00	2.00	8.91	4.46
Leucine	5762186.00	829223.30	6369770.00	600651.50	7295245.00	1292119.40	9715254.00	2365084.80	16739214.00	1171939.00	17840437.00	1847556.90
Levulinic acid	583.53	109.04	408.50	112.48	403.37	27.75	456.59	87.36	521.55	71.48	514.58	20.15
Lysine	1990029.00	149438.57	2426243.00	95868.27	2588429.00	275476.23	2447938.00	207682.01	2245328.00	156901.80	2444530.00	296749.68
Mannose	379.80	119.95	462.45	65.18	425.26	22.46	230.46	31.05	137.27	64.33	267.25	115.02
Methionine	289673.00	21859.47	320329.80	34098.27	351939.30	65634.92	503713.90	124685.84	947603.40	90348.53	995539.40	137124.34

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Methylguanidine	55923.37	1737.98	65832.23	3678.88	61680.21	5402.30	54116.11	18358.15	25741.00	3523.53	24053.75	3602.20
N-Acetyethanolamine	2715.07	55.41	4087.53	430.46	3446.89	176.76	3344.13	998.76	1273.05	353.95	1020.18	72.53
N-Acetylneuraminic acid	10678.35	813.62	10795.91	2776.34	11215.04	1050.05	7211.74	2859.08	2387.99	293.14	1649.95	357.39
N-N-Dicyclohexylurea	27209.98	2659.17	31702.39	1695.98	35742.18	2717.82	29416.95	1432.92	33246.30	902.04	27762.94	5897.05
NAD	397.05	215.32	414.75	7.81	578.95	209.49	494.60	190.01	250.69	131.87	582.62	135.17
Nicotinamide	100018.50	8720.50	96404.40	5519.94	105473.20	14302.78	104612.90	11649.42	141911.60	16476.12	128766.00	17596.91
Nicotinic-Acid	16302.51	1442.69	18319.67	2145.22	16657.87	2142.16	16206.24	5585.64	3577.97	228.68	3514.07	461.55
Nonadecanoic acid	326115.90	30370.96	345804.20	62251.83	354356.10	7030.64	546015.00	304215.98	842857.90	37607.14	774251.80	18417.40
Norleucine	5376200.00	749147.20	5912916.00	592984.10	6786388.00	1171824.40	9076619.00	2204785.40	15436979.00	1072328.60	16596198.00	1689782.10
Norvaline	1052365.00	119346.56	1199633.00	105629.73	1187759.00	179995.79	1443473.00	34116.09	1875440.00	153555.15	1919568.00	253772.81
Ornithine	101626.75	11627.40	81102.17	7648.54	90505.21	10627.87	144649.00	4406.28	189329.50	21683.54	199683.31	53367.51
Oxalic acid	7030.11	390.40	8561.06	764.96	7102.77	608.72	7061.14	652.46	10480.08	1934.10	12386.95	3456.37
p-Coumaric acid	2622.53	567.56	1968.64	154.78	2887.09	182.89	2619.82	363.29	2437.00	222.08	2310.08	322.24
Palmitic acid	618695.70	178241.04	466009.20	90711.27	349572.00	63581.50	275501.20	64954.75	230086.60	27383.98	254806.80	54843.27
Pantothenic acid	29578.74	2827.22	34181.15	1328.19	40182.46	4780.58	39836.32	3049.61	108408.59	16744.85	94724.02	19858.51
Pentadecanoic acid	234699.70	24105.71	436264.20	75056.67	264790.80	25053.50	465317.30	233850.36	417930.80	57049.05	398255.60	33707.26
Phenylalanine	3491962.00	456688.20	3958685.00	362168.20	4099817.00	711688.00	6484157.00	1701878.60	10815006.00	744258.90	12072211.00	1087447.60
Phenylglyoxylic acid	705.09	11.63	472.75	26.04	497.54	73.37	444.20	161.64	348.45	72.81	416.77	45.44
Phenylpyruvic acid	905.37	152.05	541.49	82.80	641.09	28.56	802.36	112.07	948.11	39.09	1013.70	124.51
Phthalic acid	38241.98	235.98	36702.84	2697.97	46304.76	5353.12	29183.44	6079.57	17582.07	336.80	16708.07	936.87
Picolinic acid	19108.65	725.40	19036.40	2560.22	17644.13	2162.35	16600.61	5905.70	3960.90	405.90	3983.86	849.60
Pipecolic acid	1213736.50	17562.52	1610342.30	75529.55	1536862.20	177301.88	1400015.40	319517.13	927193.00	78737.13	963748.30	123352.58
Proline	4454127.00	195549.40	5018963.00	490554.10	4605200.00	379033.00	3813135.00	1364382.20	1438060.00	131406.70	1409590.00	183989.10
Putrescine	3633.01	826.55	5577.02	341.08	6336.34	466.34	4163.90	780.64	3326.61	626.75	3740.91	268.35
Pyroglutamic acid	1174250.90	20770.10	1585231.90	82893.89	1519499.70	171767.38	1371443.80	314121.08	918242.60	83202.74	958048.30	119654.51

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Pyruvate	3597.91	305.84	4301.63	172.95	3552.08	148.54	2933.76	940.33	835.65	5.80	1004.03	287.80
Raffinose	160182.47	7499.48	186139.61	9974.82	182769.19	2442.36	144636.99	41178.14	55598.76	5337.11	50462.18	12212.70
Sarcosine	76711.21	9052.82	87954.29	6851.37	82498.32	11058.01	107674.73	5744.67	136890.55	11483.26	149516.72	20161.13
Sebacic acid	1046.94	91.61	1342.24	203.30	1366.98	161.67	1225.29	153.24	1469.72	142.96	1279.33	259.01
Serine	545983.40	31739.10	650085.30	50563.66	623057.00	79085.48	661924.60	59609.23	714937.00	61127.08	717181.70	79104.64
Serotonin	13011.18	1031.59	17094.21	2060.41	15886.50	739.05	15213.54	3921.67	11789.76	462.95	11484.49	1208.71
Shikimic acid	3133.08	252.12	3480.70	179.75	3542.74	144.70	2563.81	812.36	1040.14	74.02	859.43	91.31
Sorbitol	56535.79	2180.79	66861.41	1972.05	65333.45	1206.53	48959.13	16456.34	15309.36	1543.56	15184.67	4114.74
Stearic acid	178431.30	34489.87	150752.83	11660.71	149051.22	12780.96	123232.02	12879.17	85235.96	3387.03	104362.29	15950.90
Threonine	1141555.30	22701.34	1347383.30	89854.81	1288370.00	109112.77	1097516.90	321343.69	575970.70	57799.72	581397.90	91901.68
Thymine	10428.09	2585.77	15986.25	899.65	16476.34	4651.09	11290.61	3895.17	14345.48	1434.20	5497.24	1924.84
Total.Glutathione	4106.90	836.42	4013.03	377.86	4621.17	871.96	4332.95	406.26	3311.71	323.40	4700.88	315.81
Tryptophan	430244.00	39559.45	514096.30	53722.34	471884.50	62913.94	696553.50	69671.55	925567.90	79150.91	1071514.80	105098.79
Tyrosine	412876.80	40253.85	480789.20	36905.19	457887.90	78079.93	691032.00	109551.54	1025985.30	70149.00	1130089.90	127799.13
Uracil	509923.00	20062.96	589937.30	40368.22	577026.00	42803.93	415994.40	94303.80	362124.20	31980.12	370378.90	53321.41

Supplemental Table 6. Expression of genes involved in various intermediary metabolism pathways in livers of PBDE-exposed CV and GF mice. Data were obtained from RNA-Seq experiments, and average expression (FPKM) and standard errors are shown. Gene expression was calculated using Cufflinks.

Category	Gene	CV CO	SE	CV BDE-47	SE	CV BDE-99	SE	GF CO	SE	GF BDE-47	SE	GF BDE-99	SE
Lipid metabolism	9130409I 23Rik	6.71	2.46	6.96	1.53	7.62	2.00	2.55	1.16	4.93	0.63	24.41	3.11
Lipid metabolism & Amino acid metabolism	Acaa1a	175.19	17.24	185.67	21.12	142.62	10.49	206.17	31.44	186.24	15.80	129.29	3.05
Lipid metabolism & Amino acid metabolism	Acaa1b	832.68	80.04	789.37	115.64	397.56	137.45	816.72	98.64	733.71	69.16	462.93	26.91
Lipid metabolism & Amino acid metabolism	Acaa2	668.61	79.93	652.59	45.00	519.45	4.85	738.81	80.26	658.73	43.63	460.66	14.77
Carbohydrate metabolism & Lipid metabolism & Amino acid metabolism	Acads	106.83	14.98	110.39	5.52	91.99	1.08	116.26	18.04	120.27	11.45	70.71	2.19
Amino acid metabolism	Acmsd	3.91	1.01	4.08	0.98	3.93	1.09	5.10	1.55	8.72	1.79	18.09	1.64
Lipid metabolism	Acot1	28.68	4.58	17.10	2.38	6.95	1.08	31.48	2.91	23.83	1.70	5.50	0.35
Lipid metabolism	Acot3	3.55	0.45	2.19	1.02	3.28	0.90	1.88	0.30	3.96	1.51	1.35	0.39
Lipid metabolism	Acot7	14.17	2.32	14.52	1.19	10.74	0.83	14.89	2.99	14.35	1.61	9.12	0.42
Lipid metabolism	Acot8	12.72	1.67	14.03	1.12	11.12	0.47	13.56	1.67	13.77	1.94	9.72	0.11
Lipid metabolism	AcsI4	6.49	2.38	5.06	1.33	6.09	1.12	5.70	3.13	4.00	0.96	13.04	0.74
Carbohydrate metabolism & Energy metabolism	Acss2	77.38	1.92	68.46	6.69	36.58	3.35	107.17	15.58	102.68	12.40	64.00	9.30
Carbohydrate metabolism	Acyp2	1.34	0.24	1.52	0.23	1.67	0.05	0.80	0.26	1.47	0.11	2.16	0.29
Nucleotide metabolism	Ada	1.27	0.18	1.22	0.06	0.99	0.01	1.29	0.24	1.05	0.07	0.77	0.13
Lipid metabolism	Agpat2	174.47	21.39	130.19	20.22	95.53	8.29	159.01	31.92	141.97	13.80	91.89	2.50

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Amino acid metabolism	Ahcy	888.95	156.26	963.39	77.30	592.93	30.98	1446.18	307.38	1139.00	35.29	814.31	53.26
Nucleotide metabolism & Metabolism of cofactors and vitamins	Ak4	9.74	1.77	9.70	3.12	14.36	0.47	5.80	1.64	5.68	0.52	7.30	0.88
Carbohydrate metabolism & Lipid metabolism & Metabolism of cofactors and vitamins	Akr1b7	0.38	0.09	0.71	0.30	1.43	0.15	0.32	0.07	3.12	0.62	3.90	0.39
Amino acid metabolism & Metabolism of cofactors and vitamins	Alas1	72.89	18.37	240.49	76.66	219.09	50.59	32.09	6.97	136.08	23.49	134.05	8.73
Carbohydrate metabolism & Amino acid metabolism & Metabolism of other amino acids	Aldh3b1	1.21	0.13	1.98	0.12	2.01	0.29	0.97	0.15	1.32	0.19	0.90	0.05
Carbohydrate metabolism & Energy metabolism	Aldoc	8.20	1.02	9.14	0.45	5.65	0.23	13.16	1.32	12.36	1.13	7.61	0.48
Glycan biosynthesis and metabolism	Alg12	5.14	0.22	2.52	0.34	2.16	0.49	3.12	0.16	3.89	0.28	4.71	0.29
Amino acid metabolism & Metabolism of cofactors and vitamins	Aox1	25.60	3.22	30.13	2.09	27.86	6.36	13.35	2.08	22.87	2.01	56.60	4.08
Lipid metabolism	Arsa	11.51	1.48	11.34	1.37	14.26	1.01	8.09	1.29	7.46	0.36	6.79	0.18
Amino acid metabolism	Asl	312.41	48.01	303.61	41.07	209.48	9.36	334.80	85.63	261.52	26.99	209.17	8.89
Amino acid metabolism	Asns	0.89	0.33	1.39	0.28	2.48	0.19	2.11	1.03	0.85	0.14	3.68	0.40
Metabolism of cofactors and vitamins	Aspdh	89.80	17.55	76.91	9.17	61.84	0.04	82.53	17.97	80.00	13.72	53.25	2.99
Glycan biosynthesis and metabolism	B3galt1	1.83	0.72	3.80	1.58	4.70	1.52	1.37	0.64	1.76	0.40	5.33	0.27
Glycan biosynthesis and metabolism	B3gat3	23.04	3.39	18.45	2.58	13.55	0.47	24.64	4.85	21.16	2.23	15.15	0.55
Glycan biosynthesis and metabolism	B3gnt3	1.34	0.19	1.63	0.08	1.52	0.10	1.74	0.13	1.48	0.11	1.10	0.07
Glycan biosynthesis and metabolism	B4galnt1	44.05	6.71	41.84	4.11	37.44	1.89	42.01	7.37	41.15	2.78	27.03	0.90
Amino acid metabolism	Bhmt	875.34	109.64	570.50	111.01	653.36	276.76	1409.12	366.54	1159.34	92.04	1275.64	96.37
Metabolism of cofactors and vitamins	Btd	38.58	5.84	36.06	3.08	32.05	1.80	43.58	10.00	39.50	0.81	23.73	0.87

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Lipid metabolism	Cel	0.06	0.05	0.15	0.15	33.42	25.60	0.01	0.01	0.01	0.01	0.00	0.00
Lipid metabolism	Chka	19.43	2.16	6.29	1.78	2.77	0.37	13.62	2.33	9.63	0.65	11.40	1.04
Glycan biosynthesis and metabolism	Chpf	0.82	0.24	0.65	0.04	0.57	0.04	0.62	0.05	0.68	0.05	0.39	0.03
Glycan biosynthesis and metabolism	Chst7	0.61	0.08	0.71	0.04	0.61	0.02	0.74	0.09	0.87	0.04	0.48	0.06
Amino acid metabolism	Ckb	3.82	0.70	3.41	0.59	3.07	0.26	3.78	0.70	4.23	0.70	2.65	0.14
Metabolism of other amino acids	Csad	227.81	18.05	215.00	25.32	112.36	33.49	24.32	4.09	12.91	2.21	13.35	1.28
Amino acid metabolism & Metabolism of other amino acids	Cth	147.29	33.17	115.97	11.03	113.12	0.69	167.40	44.07	168.49	15.00	323.32	6.90
Lipid metabolism	Cyp17a1	2.03	0.36	1.52	0.41	2.20	1.07	3.20	0.84	4.79	1.89	3.34	0.41
Lipid metabolism & Amino acid metabolism & Metabolism of cofactors and vitamins	Cyp1a1	1.09	0.17	1.91	0.29	1.51	0.33	1.58	0.40	2.04	0.21	3.51	0.45
Lipid metabolism & Amino acid metabolism & Metabolism of cofactors and vitamins	Cyp1a2	391.96	38.91	884.13	58.82	719.69	206.33	635.10	112.71	1098.82	98.74	1557.19	108.28
Lipid metabolism & Amino acid metabolism & Metabolism of cofactors and vitamins	Cyp2a4	17.80	2.00	51.16	12.44	63.80	15.82	17.73	0.58	61.55	2.12	178.85	3.27
Lipid metabolism & Amino acid metabolism & Metabolism of cofactors and vitamins	Cyp2a5	227.48	22.22	651.65	167.32	869.87	203.59	216.37	9.39	671.54	64.42	2214.54	60.40
Lipid metabolism & Amino acid metabolism & Metabolism of cofactors and vitamins	Cyp2b10	18.72	1.25	352.58	57.93	550.29	6.82	18.00	3.70	403.26	56.66	963.99	50.05
Lipid metabolism & Amino acid metabolism & Metabolism of cofactors and vitamins	Cyp2b13	0.75	0.14	10.73	1.75	16.58	0.04	2.76	1.66	35.05	19.44	27.80	2.03
Lipid metabolism & Amino acid metabolism & Metabolism of cofactors and vitamins	Cyp2b9	3.29	0.96	1.80	0.85	2.87	0.13	8.54	1.11	27.51	15.20	17.72	4.49
Lipid metabolism & Amino acid metabolism & Metabolism of cofactors and vitamins	Cyp2c29	1358.95	36.01	4370.67	381.61	4671.23	383.40	1605.94	129.41	4820.12	180.39	5023.44	331.18
Lipid metabolism & Amino acid metabolism & Metabolism of cofactors and vitamins	Cyp2c37	193.49	3.86	484.33	30.88	529.16	123.52	280.93	41.67	704.26	88.19	1097.83	74.38

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Lipid metabolism & Amino acid metabolism & Metabolism of cofactors and vitamins	Cyp2c40	31.17	2.09	28.77	1.97	26.57	3.09	74.28	8.31	81.17	14.66	57.66	4.77
Lipid metabolism & Amino acid metabolism & Metabolism of cofactors and vitamins	Cyp2c50	428.37	15.93	1094.53	48.36	1155.31	219.39	690.14	106.68	1537.97	188.07	2583.09	128.07
Lipid metabolism & Amino acid metabolism & Metabolism of cofactors and vitamins	Cyp2c54	197.33	4.27	497.79	20.44	538.88	56.21	386.07	34.88	721.67	85.91	1124.85	40.02
Lipid metabolism & Amino acid metabolism & Metabolism of cofactors and vitamins	Cyp2c55	5.29	1.25	71.18	28.12	98.82	52.81	3.80	1.31	60.33	5.37	200.72	4.68
Lipid metabolism & Amino acid metabolism & Metabolism of cofactors and vitamins	Cyp2c68	79.52	7.10	73.29	5.37	77.77	7.96	144.31	22.66	161.41	15.16	160.21	17.51
Lipid metabolism & Amino acid metabolism & Metabolism of cofactors and vitamins	Cyp2c70	163.19	2.08	186.35	7.91	194.20	8.73	351.01	38.66	413.47	7.32	349.02	5.75
Lipid metabolism & Amino acid metabolism & Metabolism of cofactors and vitamins	Cyp2d11	53.17	10.84	56.61	7.03	46.75	2.11	53.34	13.93	45.75	2.17	34.36	2.45
Lipid metabolism & Amino acid metabolism & Metabolism of cofactors and vitamins	Cyp2d12	65.64	10.13	84.59	12.78	65.10	6.05	71.12	16.71	55.31	6.20	50.17	3.29
Lipid metabolism & Amino acid metabolism & Metabolism of cofactors and vitamins	Cyp2d26	598.42	92.90	578.12	74.99	539.59	43.67	772.39	179.78	655.98	52.65	423.57	15.26
Lipid metabolism & Amino acid metabolism & Metabolism of cofactors and vitamins	Cyp2d34	13.13	1.56	13.91	1.51	11.54	0.26	12.27	1.01	12.81	0.89	10.08	0.81
Lipid metabolism & Amino acid metabolism & Metabolism of cofactors and vitamins	Cyp2d40	78.85	13.64	75.74	11.22	63.17	7.15	75.15	16.20	55.25	3.25	34.10	2.45
Lipid metabolism & Amino acid metabolism & Metabolism of cofactors and vitamins	Cyp2j9	3.05	1.03	1.10	0.25	1.56	0.57	1.14	0.61	0.80	0.06	2.69	0.50
Lipid metabolism & Amino acid metabolism & Metabolism of cofactors and vitamins	Cyp3a11	2650.58	284.60	4119.00	572.44	3393.70	1165.11	273.31	45.65	1854.17	185.95	2994.30	72.10
Lipid metabolism & Amino acid metabolism & Metabolism of cofactors and vitamins	Cyp3a16	88.04	9.02	138.11	18.59	114.66	44.18	7.81	0.98	61.45	5.30	82.33	4.44
Lipid metabolism & Amino acid metabolism & Metabolism of cofactors and vitamins	Cyp3a25	243.23	53.01	218.09	31.57	250.25	63.20	135.64	51.81	204.05	12.91	471.74	32.52
Lipid metabolism & Amino acid metabolism & Metabolism of cofactors and vitamins	Cyp3a41a	59.42	6.19	92.54	12.43	75.93	30.73	6.28	1.17	44.51	5.44	62.24	2.05

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Lipid metabolism & Amino acid metabolism & Metabolism of cofactors and vitamins	Cyp3a41b	56.71	5.72	84.17	10.97	71.95	34.13	5.77	0.90	41.06	4.95	53.88	1.98
Lipid metabolism & Amino acid metabolism & Metabolism of cofactors and vitamins	Cyp3a44	70.69	4.99	111.66	12.23	89.84	30.59	8.66	0.46	57.46	5.80	73.93	2.90
Lipid metabolism & Metabolism of cofactors and vitamins	Cyp4a10	310.99	20.49	193.75	13.84	146.66	17.72	423.42	28.36	395.52	49.73	180.00	29.09
Lipid metabolism & Metabolism of cofactors and vitamins	Cyp4a12b	55.00	3.47	95.93	11.63	72.81	4.73	96.03	9.29	104.62	19.85	123.33	6.77
Lipid metabolism & Metabolism of cofactors and vitamins	Cyp4a14	476.22	16.16	224.95	26.38	215.26	25.17	361.50	26.77	352.95	57.76	147.70	34.43
Lipid metabolism & Metabolism of cofactors and vitamins	Cyp4a31	10.76	0.60	7.52	0.16	8.77	1.16	14.93	0.77	15.98	1.68	8.70	1.33
Lipid metabolism & Amino acid metabolism & Metabolism of cofactors and vitamins	Cyp4b1	11.05	1.02	11.10	1.07	10.72	0.18	10.74	0.43	8.69	0.34	7.38	0.46
Lipid metabolism	Cyp4f13	41.95	6.88	43.72	3.81	34.03	3.00	44.91	8.52	37.91	2.42	27.87	0.63
Lipid metabolism	Cyp7a1	7.44	2.74	12.12	3.40	14.83	0.95	2.70	0.91	3.33	1.07	11.41	0.97
Lipid metabolism	Cyp7b1	51.33	5.04	110.82	26.37	92.00	15.76	91.17	17.28	82.40	17.74	94.82	4.92
Lipid metabolism	Cyp8b1	272.41	32.10	253.00	60.37	120.31	28.67	285.92	63.17	129.57	55.30	164.62	30.70
Carbohydrate metabolism & Amino acid metabolism	Dbt	15.66	5.84	9.11	2.27	14.09	0.02	10.96	5.04	11.22	1.68	30.19	2.50
Amino acid metabolism & Nucleotide metabolism	Dct	0.77	0.17	1.05	0.20	0.82	0.10	2.85	0.50	1.63	0.17	2.26	0.47
Nucleotide metabolism	Dctpp1	38.16	8.69	44.25	5.20	36.79	2.12	54.54	17.33	49.28	10.39	27.72	1.29
Carbohydrate metabolism	Dcxr	114.54	19.29	119.39	18.27	98.04	5.62	171.49	47.87	150.93	24.78	94.38	6.07
Amino acid metabolism	Ddc	45.67	0.68	27.19	5.26	19.21	2.09	28.08	2.19	29.22	1.75	53.64	3.90
Carbohydrate metabolism & Amino acid metabolism	Dhtkd1	9.15	2.26	4.67	0.61	5.83	0.56	6.82	2.53	5.86	1.53	13.32	1.35
Carbohydrate metabolism & Amino acid metabolism	Dld	30.39	10.65	19.35	4.37	26.22	1.84	18.44	8.99	19.11	2.06	51.49	3.39

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Lipid metabolism	Ebp	141.06	20.78	137.28	17.63	107.13	3.97	219.28	50.53	167.83	13.16	111.53	11.37
Carbohydrate metabolism	Echdc1	12.67	4.05	10.42	2.40	10.56	1.34	8.88	3.99	9.90	1.84	22.59	0.90
Lipid metabolism	Elov3	290.87	37.59	188.34	55.05	40.46	17.55	246.42	55.63	129.78	31.24	150.17	12.12
Lipid metabolism	Elov5	114.41	17.67	74.51	7.46	47.51	7.64	115.09	37.89	85.13	14.45	91.18	10.49
Carbohydrate metabolism & Energy metabolism	Eno1b	91.45	11.32	91.47	12.75	57.38	8.17	75.99	13.29	70.02	5.71	50.93	2.98
Lipid metabolism	Ephx2	467.56	54.63	474.71	45.66	357.80	8.10	588.81	98.46	441.39	5.47	343.29	2.56
Lipid metabolism	Etnppl	44.61	10.71	31.85	4.25	28.20	4.77	35.09	12.13	37.75	2.54	77.17	10.48
Lipid metabolism	Fads2	179.85	23.61	215.33	29.82	157.33	28.20	284.22	46.26	201.84	19.56	184.93	10.23
Lipid metabolism	Fasn	60.35	4.16	32.75	2.42	19.34	2.82	31.83	0.71	30.87	3.34	23.67	1.41
Glycan biosynthesis and metabolism	Fuca1	35.01	6.94	34.93	4.01	29.09	0.44	41.03	9.36	35.80	3.96	22.86	1.48
Glycan biosynthesis and metabolism	Fut8	1.20	0.32	1.00	0.21	1.28	0.07	1.11	0.41	1.96	0.22	2.15	0.13
Metabolism of cofactors and vitamins	Fxn	13.22	2.19	13.82	1.14	11.71	0.38	15.70	3.44	15.80	3.48	10.01	0.12
Carbohydrate metabolism	G6pc	118.92	13.30	59.18	15.50	24.41	2.42	84.86	10.12	53.89	7.95	92.16	9.44
Lipid metabolism	Gal3st1	0.83	0.06	0.91	0.14	1.07	0.24	0.31	0.05	0.32	0.01	0.16	0.01
Carbohydrate metabolism	Gale	16.98	1.56	9.54	2.00	4.94	0.41	13.38	1.72	20.82	3.87	17.78	0.34
Carbohydrate metabolism	Galk1	44.03	9.36	52.09	7.28	39.81	1.47	53.09	15.77	50.23	8.72	27.87	1.33
Amino acid metabolism	Gcat	41.12	6.25	41.99	3.51	29.08	1.25	49.34	8.91	51.99	5.16	34.15	1.12
Carbohydrate metabolism	Gck	52.27	1.86	50.61	4.65	28.58	6.05	68.71	13.55	52.77	6.58	25.24	1.33
Amino acid metabolism & Metabolism of other amino acids	Gclc	92.19	17.65	70.81	8.07	78.88	17.41	57.29	17.58	81.33	9.29	154.83	1.12

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Carbohydrate metabolism & Amino acid metabolism	Gfpt1	5.10	1.38	2.78	0.72	2.64	0.21	2.74	1.19	3.93	0.74	7.35	0.43
Amino acid metabolism	Gldc	54.97	5.18	33.07	5.01	29.86	0.33	38.44	5.25	44.61	3.65	68.25	4.18
Carbohydrate metabolism	Gmgs	1.42	0.10	1.48	0.20	2.36	0.07	0.62	0.13	2.11	0.25	1.11	0.13
Nucleotide metabolism	Gmpr	0.68	0.13	0.73	0.02	0.57	0.08	1.12	0.14	1.17	0.21	0.60	0.05
Amino acid metabolism	Gnmt	1766.57	278.48	1820.24	279.10	1289.84	10.34	2605.03	706.73	1884.67	175.52	1498.01	72.83
Carbohydrate metabolism	Gnpda2	0.90	0.35	0.54	0.13	0.69	0.18	0.60	0.32	0.57	0.13	1.55	0.21
Energy metabolism & Amino acid metabolism	Got1	64.07	3.41	65.79	8.13	43.17	1.00	115.32	10.46	79.74	6.10	116.87	1.09
Lipid metabolism	Gpcpd1	8.24	3.00	20.88	7.30	32.43	3.58	10.12	5.79	12.96	3.15	33.12	1.09
Lipid metabolism	Gpd1	175.42	13.43	169.00	23.37	125.08	2.39	171.15	25.28	139.59	3.35	108.03	2.96
Lipid metabolism & Metabolism of other amino acids	Gpx3	4.09	1.60	3.36	0.33	3.45	0.76	4.26	0.82	3.72	0.56	1.75	0.11
Lipid metabolism & Metabolism of other amino acids	Gpx6	0.36	0.03	0.27	0.02	0.36	0.01	2.27	0.28	1.63	0.19	1.56	0.12
Metabolism of other amino acids	Gsta1	16.64	2.63	50.54	8.84	68.91	45.37	10.13	1.52	46.04	8.78	193.95	12.62
Metabolism of other amino acids	Gsta2	46.67	6.31	69.27	10.14	65.13	34.04	34.63	4.21	97.54	17.09	170.69	8.38
Metabolism of other amino acids	Gstm1	2164.30	72.17	4393.48	790.27	5111.82	1417.41	1675.81	272.09	4264.18	642.48	6435.67	63.64
Metabolism of other amino acids	Gstm2	54.97	0.45	82.04	11.81	108.17	6.78	96.89	24.56	159.32	19.38	315.24	9.39
Metabolism of other amino acids	Gstm3	119.71	13.36	342.75	127.73	493.32	252.49	84.61	6.04	443.57	62.22	1558.04	50.20
Metabolism of other amino acids	Gstm4	54.12	0.34	106.27	4.98	94.16	28.71	42.00	4.64	90.75	7.46	130.86	0.52
Metabolism of other amino acids	Gstp1	1779.56	241.56	3050.83	194.72	1941.65	782.74	984.02	177.11	1370.70	218.00	1517.70	35.96
Metabolism of other amino acids	Gstt3	48.89	2.28	90.87	12.22	96.99	2.51	53.25	7.92	136.14	8.37	169.77	6.63

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Carbohydrate metabolism	Gulo	182.37	25.77	183.80	15.51	141.06	2.51	271.99	55.54	208.58	12.13	176.24	10.17
Amino acid metabolism	Haa0	332.80	37.43	343.65	31.19	257.90	1.10	395.55	72.46	362.59	42.28	258.65	5.43
Carbohydrate metabolism	Hao2	0.05	0.01	0.03	0.01	0.08	0.01	0.09	0.04	0.94	0.88	0.07	0.02
Carbohydrate metabolism & Lipid metabolism & Amino acid metabolism	Hmgcs1	61.87	21.66	38.38	12.30	34.82	0.41	51.83	21.49	81.40	11.26	152.06	37.52
Amino acid metabolism & Metabolism of cofactors and vitamins	Hpd	1283.21	143.41	1329.24	143.33	1148.14	31.08	1943.51	448.20	1714.59	142.63	1225.13	76.71
Carbohydrate metabolism & Energy metabolism & Lipid metabolism & Amino acid metabolism	Hsd17b10	180.11	28.70	188.62	33.59	151.66	8.70	339.08	84.22	227.47	19.40	168.94	12.72
Lipid metabolism	Hsd3b5	107.96	15.42	126.91	12.01	99.02	28.65	216.93	22.16	167.97	8.24	115.95	4.69
Glycan biosynthesis and metabolism	Hyal2	21.58	2.36	22.43	1.54	20.63	0.71	25.64	3.90	23.33	1.61	17.01	0.23
Carbohydrate metabolism	Impa2	2.46	0.35	2.00	0.14	1.66	0.22	1.37	0.21	1.06	0.04	0.81	0.06
Carbohydrate metabolism	Isyna1	3.81	0.63	4.01	0.60	7.04	2.86	2.09	0.31	2.10	0.13	1.30	0.07
Carbohydrate metabolism	Khk	381.34	78.62	304.81	55.75	187.01	8.64	271.36	54.73	255.38	44.19	161.27	12.02
Amino acid metabolism	Kmt2a	1.21	0.39	0.48	0.10	0.80	0.09	0.48	0.28	0.33	0.10	1.18	0.20
Lipid metabolism	Lcat	354.27	61.17	378.70	55.39	376.99	44.53	495.29	121.23	412.66	23.42	264.79	7.01
Carbohydrate metabolism & Amino acid metabolism	Ldha	440.09	74.71	473.24	60.56	404.53	49.71	526.99	77.27	411.31	34.51	269.95	7.36
Lipid metabolism	Lpin1	10.08	2.77	12.14	5.44	5.64	0.59	3.02	1.05	3.63	0.96	3.96	0.24
Lipid metabolism	Lpl	2.66	0.90	2.29	0.57	3.04	0.37	4.79	2.29	6.60	0.58	17.43	3.39
Lipid metabolism	Lss	12.98	0.62	11.39	0.67	4.90	0.32	15.88	2.33	21.93	2.01	21.47	3.83
Glycan biosynthesis and metabolism	Man2b2	14.47	1.46	16.34	1.06	16.58	1.93	9.90	1.08	11.98	0.58	8.39	0.37

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Amino acid metabolism	Maoa	2.03	0.35	1.75	0.24	2.11	0.00	0.93	0.32	1.01	0.12	1.09	0.10
Lipid metabolism	Mcat	13.89	2.18	13.86	0.64	11.97	0.17	16.96	2.79	17.26	1.43	10.81	0.29
Glycan biosynthesis and metabolism	Mgat4b	11.71	1.10	12.28	0.77	11.15	0.42	10.57	0.99	11.49	0.93	7.10	0.15
Lipid metabolism	Mgll	50.12	3.94	46.14	2.39	44.07	1.12	41.99	1.45	35.90	0.85	27.27	1.09
Amino acid metabolism	Mif	233.10	56.81	243.56	26.40	170.59	33.42	244.95	62.18	252.56	46.01	140.22	3.97
Carbohydrate metabolism & Metabolism of other amino acids	Mlycd	41.04	6.31	40.90	1.27	38.61	1.25	51.38	7.05	43.72	3.60	29.96	0.96
Energy metabolism & Amino acid metabolism	Mpst	64.79	5.37	58.46	5.00	52.70	4.28	70.65	10.32	73.81	9.73	44.45	2.51
Lipid metabolism	Msmo1	58.27	11.73	61.72	7.27	64.31	5.97	77.09	22.40	108.97	6.85	138.43	32.25
Energy metabolism & Metabolism of cofactors and vitamins	Mthfr	3.54	0.23	1.12	0.26	0.66	0.02	1.47	0.29	1.14	0.15	1.43	0.16
Metabolism of other amino acids	Nat8	21.12	2.44	26.75	5.78	16.74	1.72	29.20	7.94	16.96	1.98	14.50	0.44
Energy metabolism	Ndufs3	115.71	13.53	117.60	8.84	79.28	18.88	140.16	29.14	137.19	19.71	91.77	1.20
Energy metabolism	Ndufs8	115.91	26.55	127.33	14.64	93.14	3.84	165.49	46.60	138.52	21.24	83.78	3.98
Metabolism of cofactors and vitamins	Nmrk1	4.94	0.74	5.48	2.10	8.92	1.76	2.43	0.23	2.32	0.23	3.26	0.26
Amino acid metabolism & Metabolism of other amino acids	Nos3	0.91	0.06	1.01	0.07	0.84	0.02	1.19	0.21	1.00	0.05	0.80	0.08
Metabolism of cofactors and vitamins	Nqo1	5.00	0.49	6.16	0.26	6.73	0.25	4.78	0.08	8.18	0.27	14.77	0.37
Metabolism of cofactors and vitamins	Nudt12	9.40	3.60	6.02	1.20	8.71	0.29	4.86	2.12	3.83	0.61	13.51	0.68
Amino acid metabolism	Oat	175.75	37.51	150.60	13.16	165.29	12.47	312.18	42.18	241.00	25.98	405.49	7.77
Glycan biosynthesis and metabolism	Ogt	7.90	2.40	3.11	0.76	4.81	1.55	4.01	2.28	2.89	0.76	10.70	1.40
Amino acid metabolism & Metabolism of cofactors and vitamins	Pah	332.33	39.07	321.99	37.47	334.91	63.43	367.78	78.94	375.18	41.94	753.59	29.31

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Energy metabolism & Nucleotide metabolism & Metabolism of other amino acids	Papss2	34.62	10.89	29.50	4.93	33.37	1.75	15.88	5.53	23.33	4.24	49.56	0.75
Nucleotide metabolism	Pde2a	8.11	0.76	7.62	1.01	5.85	0.48	10.16	1.08	8.84	0.69	6.53	0.26
Nucleotide metabolism	Pde4b	3.64	0.92	2.56	0.57	2.32	0.47	5.62	1.92	5.46	0.93	9.96	0.57
Nucleotide metabolism	Pde4d	0.69	0.23	0.40	0.09	0.56	0.09	0.34	0.16	0.53	0.07	1.08	0.16
Nucleotide metabolism	Pde9a	11.68	0.42	6.81	1.08	6.11	0.72	10.60	0.54	13.30	1.28	12.39	0.34
Lipid metabolism	Pemt	366.51	68.59	379.29	36.13	293.09	3.77	484.10	102.44	486.57	73.25	291.45	14.84
Carbohydrate metabolism	Pfkfb3	1.17	0.18	3.18	0.70	3.47	0.45	0.68	0.18	0.90	0.11	1.16	0.03
Carbohydrate metabolism	Pgls	59.75	14.35	61.65	10.34	43.58	0.08	69.83	20.36	68.99	16.87	37.88	1.99
Carbohydrate metabolism	Pgp	17.10	3.58	16.05	1.00	12.08	1.12	15.84	3.89	17.36	2.75	8.38	0.39
Carbohydrate metabolism & Nucleotide metabolism	Pkir	92.46	3.64	62.42	8.45	38.22	4.82	28.99	1.90	26.57	2.21	21.61	1.79
Lipid metabolism	Pla2g1b	0.03	0.03	0.04	0.04	6.20	4.73	0.01	0.01	0.00	0.00	0.00	0.00
Carbohydrate metabolism	Plcb1	1.25	0.53	0.66	0.17	1.16	0.04	0.43	0.23	0.34	0.05	1.47	0.04
Carbohydrate metabolism	Plce1	0.82	0.13	0.88	0.10	1.14	0.19	0.52	0.14	0.47	0.09	0.50	0.05
Lipid metabolism	Pld4	4.12	0.81	5.10	0.65	4.45	0.12	5.05	0.86	4.81	0.53	3.42	0.25
Carbohydrate metabolism	Pmm1	2.61	0.20	2.56	0.41	2.18	0.18	2.10	0.11	2.10	0.23	1.19	0.02
Lipid metabolism & Lipid metabolism & Lipid metabolism	Pnlip	0.37	0.31	0.43	0.43	98.97	76.02	0.03	0.03	0.03	0.03	0.00	0.00
Lipid metabolism	Pnliprp1	0.17	0.13	0.36	0.22	37.79	28.86	0.04	0.01	0.33	0.02	0.65	0.19
Lipid metabolism	Pnliprp2	0.03	0.01	0.03	0.02	5.90	4.59	0.00	0.00	0.00	0.00	0.00	0.00
Nucleotide metabolism	Pnpt1	4.49	1.62	3.43	0.82	4.61	0.39	2.63	1.16	2.99	0.56	7.74	0.58

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Glycan biosynthesis and metabolism	Pomgnt2	1.39	0.07	0.97	0.04	1.02	0.06	0.99	0.20	0.88	0.06	0.57	0.07
Lipid metabolism	Ppt2	19.47	2.67	18.35	1.29	17.14	1.17	22.06	4.33	21.43	0.90	15.12	0.46
Metabolism of other amino acids	Prdx2	121.53	17.80	129.54	7.90	112.69	2.22	144.67	22.61	139.70	16.59	96.53	3.15
Metabolism of other amino acids	Prdx4	96.27	15.40	102.37	5.90	99.45	3.98	144.63	32.64	117.35	10.83	89.62	3.18
Amino acid metabolism	Prodh2	83.00	8.31	75.70	11.78	60.52	2.04	110.36	25.96	93.04	13.06	72.41	1.90
Lipid metabolism	Ptgds	0.60	0.09	0.51	0.13	0.80	0.10	0.66	0.19	3.71	1.85	1.51	0.70
Lipid metabolism	Ptges2	11.14	2.17	13.21	1.47	10.78	0.05	14.46	3.59	13.46	1.26	8.24	0.44
Lipid metabolism	Ptgis	0.75	0.21	1.27	0.15	1.34	0.09	0.70	0.16	0.65	0.08	0.37	0.01
Carbohydrate metabolism	Renbp	2.40	0.23	2.29	0.27	2.21	0.29	2.12	0.14	2.08	0.25	1.45	0.08
Metabolism of cofactors and vitamins	Retsat	498.43	95.73	432.41	59.98	281.37	17.41	363.58	78.95	257.21	3.56	118.26	9.79
Nucleotide metabolism	Rev3l	1.14	0.47	0.57	0.17	0.96	0.05	0.54	0.35	0.35	0.09	1.50	0.13
Lipid metabolism	Scd1	1278.96	252.26	803.78	127.74	305.28	116.03	559.26	157.41	372.96	72.50	241.54	34.68
Carbohydrate metabolism & Energy metabolism	Sdhb	209.80	34.07	211.41	17.90	160.19	15.40	290.31	65.52	249.87	26.75	173.01	8.00
Lipid metabolism	Smpd3	0.30	0.09	0.78	0.11	3.79	1.43	0.43	0.15	2.32	0.26	3.93	0.39
Lipid metabolism	Sqle	17.85	1.44	18.60	3.29	13.47	2.00	21.34	5.89	32.87	3.53	31.15	8.12
Glycan biosynthesis and metabolism	St3gal4	30.67	0.95	41.20	3.27	34.64	1.15	35.41	3.07	30.13	2.46	23.38	0.10
Lipid metabolism	Sult1e1	0.02	0.00	0.01	0.01	0.05	0.03	0.10	0.05	0.21	0.05	1.54	0.07
Carbohydrate metabolism	Synj2	0.42	0.02	1.03	0.39	1.18	0.19	0.35	0.02	0.41	0.09	0.44	0.03
Lipid metabolism	Tbxas1	1.06	0.13	1.58	0.22	1.14	0.04	1.33	0.05	1.20	0.03	0.96	0.05

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Amino acid metabolism	Tdo2	478.36	138.23	388.89	71.97	490.71	67.51	338.30	135.48	380.97	33.41	955.16	21.22
Lipid metabolism	Tecr	184.10	27.51	201.50	33.95	141.77	4.18	226.57	57.24	187.98	20.40	131.26	5.23
Lipid metabolism	Tm7sf2	104.05	19.70	99.48	9.60	76.80	0.36	207.24	58.87	124.64	2.33	79.43	11.09
Carbohydrate metabolism	Treh	0.61	0.13	1.11	0.23	2.23	0.95	2.05	0.47	1.41	0.19	2.35	0.13
Carbohydrate metabolism	Tsta3	20.93	4.25	23.05	2.62	17.57	0.88	23.12	6.10	19.99	2.62	13.99	0.20
Carbohydrate metabolism	Uap111	3.19	0.66	4.76	0.41	5.01	0.18	2.87	0.52	3.26	0.44	2.05	0.08
Carbohydrate metabolism	Ugdh	53.81	12.80	68.26	10.31	78.98	17.87	35.07	11.09	63.78	4.46	172.16	5.30
Carbohydrate metabolism & Lipid metabolism & Metabolism of cofactors and vitamins	Ugt2b1	131.71	21.41	212.08	33.52	191.94	56.26	200.54	70.40	238.35	45.63	521.31	33.15
Carbohydrate metabolism & Lipid metabolism & Metabolism of cofactors and vitamins	Ugt2b34	128.43	34.11	177.76	36.49	237.90	27.68	96.47	37.81	169.83	19.70	346.89	8.42
Carbohydrate metabolism & Lipid metabolism & Metabolism of cofactors and vitamins	Ugt2b35	73.11	15.17	100.44	16.62	131.08	22.58	57.68	18.73	101.86	14.60	227.49	4.69
Nucleotide metabolism	Upp1	1.78	0.24	2.20	0.29	1.56	0.05	2.99	0.72	2.80	0.44	1.32	0.15
Nucleotide metabolism	Upp2	37.09	7.11	133.37	68.30	169.40	23.48	40.66	7.55	38.12	5.73	91.03	2.22

Supplemental Table 7. Differential expression ratios and FDR *p*-value of genes involved in various intermediary metabolism pathways in livers of PBDE-exposed CV and GF mice. Data were obtained from RNA-Seq experiments and differential expression was determined using CuffDiff.

Gene	CVBDE-47 / CVCO	FDR p-value	CVBDE-99 / CVCO	FDR p-value	GFCO / CVCO	FDR p-value	GFBDE-47 / GFCO	FDR p-value	GFBDE-99 / GFCO	FDR p-value
9130409I23 Rik	0.23	1.00	0.26	1.00	-0.90	0.77	0.79	0.60	2.77	0.00
Acaa1a	0.27	1.00	0.01	1.00	0.60	1.00	-0.26	1.00	-1.05	0.01
Acaa1b	0.04	1.00	-1.18	0.30	0.18	1.00	-0.27	1.00	-1.28	0.00
Acaa2	0.07	1.00	-0.44	1.00	0.35	1.00	-0.26	1.00	-1.12	0.01
Acads	0.14	1.00	-0.30	1.00	0.35	1.00	-0.08	1.00	-1.19	0.00
Acmsd	0.21	1.00	0.04	1.00	0.78	0.94	0.65	1.00	1.38	0.00
Acot1	-0.62	0.59	-2.01	0.00	0.39	1.00	-0.54	1.00	-2.87	0.00
Acot3	-0.52	0.94	-0.02	1.00	-0.61	1.00	1.03	0.14	-0.76	0.06
Acot7	0.12	1.00	-0.49	0.97	0.23	1.00	-0.12	1.00	-1.12	0.00
Acot8	0.24	1.00	-0.27	1.00	0.38	1.00	-0.15	1.00	-1.01	0.02
Acs14	-0.19	1.00	-0.02	1.00	0.27	1.00	-0.64	1.00	0.76	0.03
Acss2	-0.03	1.00	-1.12	0.05	0.70	1.00	-0.14	1.00	-1.14	0.00
Acyp2	0.32	1.00	0.33	1.00	-0.49	1.00	0.88	1.00	1.10	0.03
Ada	0.03	1.00	-0.45	1.00	0.33	1.00	-0.51	1.00	-1.29	0.01
Agpat2	-0.30	1.00	-0.94	0.24	0.06	1.00	-0.25	1.00	-1.22	0.00
Ahcy	0.18	1.00	-0.24	1.00	0.83	1.00	-0.47	1.00	-1.27	0.00
Ak4	0.11	1.00	0.59	0.79	-0.39	1.00	-0.14	1.00	-0.10	0.81
Akr1b7	1.02	0.42	1.94	0.02	-0.08	1.00	3.41	0.01	3.39	0.00
Alas1	1.87	0.01	1.64	0.00	-0.78	1.00	1.99	0.01	1.60	0.00
Aldh3b1	0.80	0.38	0.64	0.88	0.00	1.00	0.23	1.00	-0.66	0.13
Aldoc	0.24	1.00	-0.03	1.00	0.92	0.66	-0.23	1.00	-1.26	0.00

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Alg12	-0.90	0.16	-1.29	0.04	-0.46	1.00	0.20	1.00	0.14	0.75
Aox1	0.39	1.00	0.12	1.00	-0.63	1.00	0.69	0.85	1.66	0.00
Arsa	0.12	1.00	0.31	1.00	-0.17	1.00	-0.23	1.00	-0.71	0.03
Asl	0.07	1.00	-0.66	0.83	0.29	1.00	-0.45	1.00	-1.11	0.01
Asns	0.69	0.76	1.34	0.12	1.45	0.41	-1.54	0.02	0.24	0.58
Aspdh	-0.13	1.00	-0.63	0.75	0.04	1.00	-0.12	1.00	-1.07	0.01
B3galt1	1.23	0.04	1.47	0.01	0.06	1.00	0.24	1.00	1.53	0.00
B3gat3	-0.21	1.00	-0.85	0.33	0.34	1.00	-0.36	1.00	-1.19	0.00
B3gnt3	0.38	1.00	0.11	1.00	0.54	1.00	-0.31	1.00	-1.06	0.01
B4galnt1	0.03	1.00	-0.31	1.00	0.14	1.00	-0.14	1.00	-1.08	0.01
Bhmt	-0.42	1.00	-0.40	1.00	0.99	0.91	-0.37	1.00	-0.57	0.21
Btd	0.00	1.00	-0.35	1.00	0.40	1.00	-0.28	1.00	-1.35	0.00
Cel	1.02	1.00	8.82	0.01	-3.05	1.00	0.44	1.00	-	-
Chka	-1.49	0.01	-2.82	0.00	-0.20	1.00	-0.60	1.00	-0.69	0.04
Chpf	-0.32	1.00	-0.65	0.92	-0.01	1.00	-0.27	1.00	-1.40	0.00
Chst7	0.30	1.00	-0.08	1.00	0.75	1.00	-0.14	1.00	-1.35	0.00
Ckb	-0.05	1.00	-0.42	1.00	0.23	1.00	0.01	1.00	-1.01	0.01
Csad	0.02	1.00	-1.09	0.18	-2.92	0.01	-1.10	0.07	-1.37	0.00
Cth	-0.17	1.00	-0.34	1.00	0.55	1.00	-0.10	1.00	0.52	0.18
Cyp17a1	-0.35	1.00	0.05	1.00	0.80	1.00	0.48	1.00	-0.36	0.40
Cyp1a1	0.33	1.00	0.04	1.00	-0.11	1.00	0.45	1.00	1.15	0.00
Cyp1a2	1.27	0.01	0.80	0.81	0.91	1.00	0.67	1.00	0.85	0.05
Cyp2a4	0.74	0.59	0.47	1.00	1.11	0.58	2.18	0.01	1.41	0.00
Cyp2a5	1.61	0.01	1.90	0.00	0.23	1.00	1.50	0.02	2.88	0.00
Cyp2b10	4.33	0.01	4.83	0.00	0.24	1.00	4.32	0.01	5.24	0.00
Cyp2b13	3.10	0.12	3.20	0.26	6.37	0.16	3.15	0.01	-3.44	0.00

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Cyp2b9	-1.15	0.13	-0.59	0.90	1.70	0.01	1.49	0.01	0.60	0.11
Cyp2c29	1.79	0.01	1.73	0.26	0.50	1.00	1.49	0.03	1.23	0.01
Cyp2c37	1.42	0.01	1.43	0.03	0.76	1.00	1.26	0.03	1.62	0.00
Cyp2c40	-0.45	1.00	-1.56	0.08	2.88	0.01	-0.07	1.00	-1.80	0.00
Cyp2c50	1.46	0.01	1.38	0.11	0.97	0.76	1.00	0.32	1.48	0.00
Cyp2c54	1.43	0.01	1.39	0.04	1.21	0.32	0.78	0.87	1.12	0.01
Cyp2c55	3.87	0.01	4.23	0.00	-0.07	1.00	3.85	0.01	5.26	0.00
Cyp2c68	-0.01	1.00	0.00	1.00	1.09	0.26	0.05	1.00	-0.13	0.77
Cyp2c70	0.31	1.00	0.22	1.00	1.35	0.19	0.15	1.00	-0.42	0.30
Cyp2d11	0.06	1.00	-0.88	0.54	0.15	1.00	-1.05	0.45	-1.95	0.00
Cyp2d12	0.25	1.00	-0.20	1.00	0.13	1.00	-0.48	1.00	-1.39	0.00
Cyp2d26	0.06	1.00	-0.21	1.00	0.56	1.00	-0.34	1.00	-1.29	0.01
Cyp2d34	-0.32	1.00	-1.32	0.22	-0.08	1.00	-0.53	1.00	-3.10	0.00
Cyp2d40	0.07	1.00	-0.38	1.00	0.21	1.00	-0.67	1.00	-2.07	0.00
Cyp2j9	-1.29	0.03	-0.91	0.43	-1.13	0.58	-0.46	1.00	0.94	0.03
Cyp3a11	0.78	0.43	0.35	1.00	-2.79	0.01	2.55	0.01	2.91	0.00
Cyp3a16	0.64	0.52	0.16	1.00	-3.54	0.01	3.11	0.01	2.80	0.00
Cyp3a25	-0.01	1.00	0.06	1.00	-0.48	1.00	0.48	1.00	1.37	0.00
Cyp3a41a	0.81	0.21	-0.36	1.00	-4.08	0.01	3.81	0.01	4.02	0.00
Cyp3a41b	-	-	-	-	-3.00	0.01	2.12	0.02	-	-
Cyp3a44	0.24	1.00	-0.54	0.96	-1.64	0.21	2.69	0.01	1.56	0.00
Cyp4a10	-0.54	0.71	-1.13	0.11	0.67	1.00	-0.21	1.00	-1.60	0.00
Cyp4a12b	1.03	0.02	0.48	0.93	1.46	0.05	-0.01	1.00	0.04	0.93
Cyp4a14	-0.96	0.08	-1.17	0.18	-0.13	1.00	-0.15	1.00	-1.70	0.00
Cyp4a31	-0.27	1.00	0.15	1.00	0.80	0.89	0.16	1.00	-0.84	0.02
Cyp4b1	0.16	1.00	-0.11	1.00	0.20	1.00	-0.40	1.00	-1.01	0.00

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Cyp4f13	0.15	1.00	-0.40	1.00	0.22	1.00	-0.29	1.00	-1.06	0.01
Cyp7a1	0.86	0.20	1.07	0.13	-1.01	0.64	0.16	1.00	1.64	0.00
Cyp7b1	1.21	0.01	0.80	0.45	1.08	0.42	-0.25	1.00	-0.37	0.32
Cyp8b1	0.03	1.00	-1.26	0.06	0.26	1.00	-1.26	0.12	-1.20	0.00
Dbt	-0.61	0.66	-0.07	1.00	-0.07	1.00	-0.07	1.00	1.04	0.00
Dct	0.67	0.70	0.12	1.00	2.49	0.01	-1.14	0.11	-0.98	0.01
Dctpp1	0.29	1.00	-0.16	1.00	0.71	1.00	-0.27	1.00	-1.46	0.00
Dcxr	0.17	1.00	-0.31	1.00	0.74	1.00	-0.24	1.00	-1.27	0.00
Ddc	-0.62	0.48	-1.29	0.01	-0.44	1.00	-0.05	1.00	0.49	0.17
Dhtkd1	-0.79	0.22	-0.61	0.77	-0.08	1.00	-0.32	1.00	0.56	0.11
Dld	-0.48	0.90	-0.14	1.00	-0.29	1.00	-0.05	1.00	1.06	0.00
Ebp	0.07	1.00	-0.48	1.00	0.81	1.00	-0.47	1.00	-1.39	0.00
Echdc1	-0.12	1.00	-0.20	1.00	-0.08	1.00	0.03	1.00	0.91	0.03
Elov13	-0.48	0.93	-2.93	0.00	-0.05	1.00	-1.02	0.21	-1.13	0.00
Elov15	-0.44	0.93	-1.26	0.03	0.35	1.00	-0.56	1.00	-0.78	0.04
Eno1b	0.13	1.00	-0.97	1.00	-0.01	1.00	-0.06	1.00	-1.43	0.04
Ephx2	0.13	1.00	-0.46	1.00	0.51	1.00	-0.49	1.00	-1.18	0.01
Etnppl	-0.32	1.00	-0.63	0.74	0.02	1.00	0.02	1.00	0.70	0.05
Fads2	0.35	1.00	-0.28	1.00	0.84	1.00	-0.60	1.00	-1.05	0.01
Fasn	-0.75	0.27	-1.66	0.00	-0.63	1.00	-0.15	1.00	-0.87	0.02
Fuca1	0.09	1.00	-0.37	1.00	0.50	1.00	-0.39	1.00	-1.38	0.00
Fut8	-0.10	1.00	0.14	1.00	0.30	1.00	0.69	1.00	0.50	0.20
Fxn	0.14	1.00	-0.27	1.00	0.43	1.00	-0.08	1.00	-1.10	0.00
G6pc	-0.92	0.10	-2.30	0.00	-0.20	1.00	-0.76	0.79	-0.30	0.46
Gal3st1	0.27	1.00	0.32	1.00	-1.00	0.94	-0.25	1.00	-1.60	0.01
Gale	-0.70	0.38	-1.84	0.00	-0.15	1.00	0.59	1.00	0.00	1.00

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Galk1	0.34	1.00	-0.25	1.00	0.43	1.00	-0.16	1.00	-1.37	0.00
Gcat	0.13	1.00	-0.59	0.84	0.50	1.00	-0.06	1.00	-1.02	0.01
Gck	0.07	1.00	-0.89	0.24	0.69	1.00	-0.48	1.00	-1.89	0.00
Gclc	-0.22	1.00	-0.21	1.00	-0.35	1.00	0.41	1.00	1.02	0.01
Gfpt1	-0.72	0.35	-0.90	0.28	-0.51	1.00	0.41	1.00	1.00	0.01
Gldc	-0.59	0.58	-0.89	0.28	-0.24	1.00	0.13	1.00	0.41	0.28
Gmcs	0.15	1.00	0.70	0.79	-0.86	0.94	1.62	0.01	0.32	0.50
Gmpr	0.16	1.00	-0.32	1.00	0.77	1.00	0.11	1.00	-1.21	0.01
Gnmt	0.16	1.00	-0.54	1.00	0.76	1.00	-0.57	1.00	-1.25	0.02
Gnpda2	-0.54	1.00	-0.31	1.00	-0.08	1.00	-0.24	1.00	0.89	0.04
Got1	0.17	1.00	-0.60	0.77	1.12	0.41	-0.64	1.00	-0.42	0.29
Gpcpd1	1.51	0.01	2.05	0.00	0.73	1.00	0.24	1.00	1.29	0.00
Gpd1	0.08	1.00	-0.55	0.94	0.18	1.00	-0.39	1.00	-1.10	0.01
Gpx3	-0.25	1.00	-0.39	1.00	0.17	1.00	-0.32	1.00	-1.77	0.00
Gpx6	-0.23	1.00	-0.01	1.00	2.82	0.01	-0.42	1.00	-0.83	0.04
Gsta1	2.39	0.01	2.64	0.00	-1.04	0.98	3.17	0.01	4.83	0.00
Gsta2	0.64	0.46	0.35	1.00	-0.15	1.00	1.42	0.01	1.73	0.00
Gstm1	1.11	0.08	1.18	0.73	-0.13	1.00	1.28	0.11	1.50	0.00
Gstm2	0.68	0.35	0.95	0.16	1.04	0.51	0.67	1.00	1.29	0.00
Gstm3	1.86	0.01	2.39	0.00	-0.20	1.00	2.76	0.01	4.38	0.00
Gstm4	1.10	0.01	0.75	0.51	-0.13	1.00	1.04	0.12	1.22	0.00
Gstp1	0.86	0.22	0.04	1.00	-0.62	1.00	0.31	1.00	0.19	0.69
Gstt3	1.03	0.04	0.95	0.18	0.42	1.00	1.23	0.06	1.21	0.00
Gulo	0.11	1.00	-0.45	1.00	0.76	1.00	-0.48	1.00	-1.05	0.01
Hao	0.16	1.00	-0.44	1.00	0.45	1.00	-0.20	1.00	-1.03	0.01
Hao2	-0.44	1.00	0.75	1.00	0.40	1.00	3.98	0.01	-0.04	1.00

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Hmgcs1	-0.50	0.89	-0.76	0.58	0.17	1.00	0.56	1.00	1.17	0.01
Hpd	0.17	1.00	-0.23	1.00	0.80	1.00	-0.27	1.00	-1.09	0.02
Hsd17b10	0.19	1.00	-0.33	1.00	1.07	0.64	-0.65	1.00	-1.42	0.00
Hsd3b5	0.33	1.00	-0.20	1.00	1.17	0.46	-0.41	1.00	-1.28	0.00
Hyal2	0.16	1.00	-0.14	1.00	0.45	1.00	-0.23	1.00	-1.03	0.00
Impa2	-0.21	1.00	-0.65	0.88	-0.62	1.00	-0.51	1.00	-1.23	0.01
Isyna1	0.16	1.00	0.81	0.51	-0.77	1.00	-0.01	1.00	-1.04	0.01
Khk	-0.21	1.00	-1.13	0.16	-0.29	1.00	-0.21	1.00	-1.22	0.00
Kmt2a	-1.15	0.02	-0.53	0.92	-0.92	0.84	-0.66	1.00	0.86	0.02
Lcat	0.20	1.00	0.00	1.00	0.67	1.00	-0.36	1.00	-1.35	0.00
Ldha	0.20	1.00	-0.20	1.00	0.46	1.00	-0.46	1.00	-1.42	0.00
Lpin1	0.42	1.00	-0.80	0.42	-1.37	0.09	0.15	1.00	-0.02	0.96
Lpl	-0.05	1.00	0.26	1.00	1.25	0.24	0.39	1.00	1.45	0.00
Lss	-0.06	1.00	-0.63	0.70	0.55	1.00	0.38	1.00	0.03	0.94
Man2b2	0.28	1.00	0.13	1.00	-0.29	1.00	0.13	1.00	-0.72	0.05
Maoa	-0.08	1.00	0.07	1.00	-0.71	1.00	-0.06	1.00	-0.27	0.51
Mcat	0.08	1.00	-0.30	1.00	0.39	1.00	0.01	1.00	-1.01	0.00
Mgat4b	0.17	1.00	-0.14	1.00	0.10	1.00	0.00	1.00	-1.05	0.00
Mgl1	-0.01	1.00	-0.25	1.00	-0.01	1.00	-0.35	1.00	-1.09	0.00
Mif	0.14	1.00	-0.56	0.90	0.30	1.00	-0.09	1.00	-1.32	0.00
Mlycd	0.08	1.00	-0.17	1.00	0.50	1.00	-0.32	1.00	-1.21	0.00
Mpst	-0.03	1.00	-0.36	1.00	0.35	1.00	-0.03	1.00	-1.11	0.00
Msmo1	0.24	1.00	0.17	1.00	0.75	1.00	0.41	1.00	0.44	0.27
Mthfr	-1.54	0.01	-2.45	0.00	-0.98	0.51	-0.50	1.00	-0.50	0.17
Nat8	0.63	0.62	-0.33	1.00	0.74	1.00	-1.25	0.03	-1.47	0.00
Ndufs3	0.12	1.00	-0.64	0.73	0.43	1.00	-0.11	1.00	-1.01	0.01

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Ndufs8	0.22	1.00	-0.43	1.00	0.64	1.00	-0.31	1.00	-1.39	0.00
Nmrk1	0.26	1.00	0.87	0.35	-0.72	1.00	-0.13	1.00	0.01	0.99
Nos3	0.26	1.00	-0.17	1.00	0.64	1.00	-0.38	1.00	-1.01	0.01
Nqo1	0.46	0.97	0.42	1.00	0.19	1.00	0.72	0.77	1.25	0.00
Nudt12	-0.45	1.00	-0.02	1.00	-0.51	1.00	-0.45	1.00	1.05	0.00
Oat	-0.04	1.00	-0.05	1.00	1.17	0.38	-0.46	1.00	-0.03	0.96
Ogt	-1.18	0.01	-0.67	0.70	-0.60	1.00	-0.57	1.00	1.01	0.01
Pah	0.09	1.00	0.00	1.00	0.45	1.00	-0.07	1.00	0.61	0.14
Papss2	-0.05	1.00	0.01	1.00	-0.71	1.00	0.44	1.00	1.22	0.00
Pde2a	0.04	1.00	-0.54	0.89	0.48	1.00	-0.26	1.00	-1.01	0.00
Pde4b	-0.36	1.00	-0.62	0.81	0.99	0.51	-0.15	1.00	0.40	0.27
Pde4d	-0.63	0.74	-0.23	1.00	-0.52	1.00	0.46	1.00	1.14	0.00
Pde9a	-0.63	0.54	-0.97	0.16	0.15	1.00	0.22	1.00	-0.23	0.55
Pemt	0.14	1.00	-0.42	1.00	0.56	1.00	-0.07	1.00	-1.15	0.01
Pfkfb3	1.59	0.01	1.58	0.00	-0.42	1.00	0.27	1.00	0.31	0.46
Pgl3	0.14	1.00	-0.58	0.85	0.36	1.00	-0.07	1.00	-1.31	0.00
Pgp	-0.02	1.00	-0.61	0.84	-0.02	1.00	0.11	1.00	-1.29	0.00
Pklr	-0.43	0.98	-1.31	0.01	-1.40	0.12	-0.24	1.00	-0.86	0.01
Pla2g1b	0.19	1.00	7.42	0.92	-1.70	1.00	-	-	-	-
Plcb1	-0.73	0.53	-0.01	1.00	-1.06	0.63	-0.46	1.00	1.34	0.00
Plce1	0.23	1.00	0.48	1.00	-0.42	1.00	-0.19	1.00	-0.41	0.29
Pld4	0.39	1.00	0.01	1.00	0.49	1.00	-0.19	1.00	-1.03	0.00
Pmm1	0.11	1.00	-0.31	1.00	0.48	1.00	-0.66	1.00	-1.82	0.00
Pnlip	-0.03	1.00	7.81	0.00	-3.80	0.37	-0.01	1.00	-	-
Pnliprp1	0.95	0.90	7.58	0.00	-1.73	0.96	2.50	0.19	3.19	0.01
Pnliprp2	-0.31	1.00	7.40	0.15	-	-	-	-	-	-

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Pnpt1	-0.21	1.00	0.12	1.00	-0.39	1.00	0.14	1.00	1.20	0.00
Pomgnt2	-0.41	1.00	-0.50	1.00	-0.05	1.00	-0.47	1.00	-1.47	0.00
Ppt2	0.01	1.00	-0.33	1.00	0.40	1.00	-0.17	1.00	-1.01	0.00
Prdx2	0.18	1.00	-0.19	1.00	0.45	1.00	-0.15	1.00	-1.03	0.00
Prdx4	0.18	1.00	-0.04	1.00	0.76	1.00	-0.38	1.00	-1.11	0.00
Prodh2	0.00	1.00	-0.52	0.93	0.66	1.00	-0.37	1.00	-1.08	0.01
Ptgds	-0.19	1.00	0.36	1.00	0.39	1.00	2.44	0.01	0.71	0.18
Ptges2	0.34	1.00	-0.15	1.00	0.52	1.00	-0.18	1.00	-1.23	0.00
Ptgis	0.81	0.52	0.72	0.86	-0.01	1.00	-0.16	1.00	-1.31	0.00
Renbp	0.03	1.00	-0.19	1.00	0.25	1.00	-0.33	1.00	-1.20	0.00
Retsat	-0.11	1.00	-0.92	0.57	-0.28	1.00	-0.62	1.00	-2.06	0.00
Rev3l	-0.83	0.31	-0.16	1.00	-0.63	1.00	-0.74	1.00	1.05	0.01
Scd1	-0.49	1.00	-2.06	0.11	-0.84	1.00	-0.72	1.00	-1.66	0.00
Sdhb	0.10	1.00	-0.48	1.00	0.63	1.00	-0.29	1.00	-1.17	0.00
Smpd3	1.59	0.01	3.74	0.00	0.86	1.00	2.41	0.01	2.82	0.00
Sqle	0.19	1.00	-0.41	1.00	0.54	1.00	0.53	1.00	0.15	0.74
St3gal4	0.53	0.85	0.13	1.00	0.45	1.00	-0.33	1.00	-1.03	0.01
Sult1e1	-0.34	1.00	1.16	1.00	2.22	1.00	1.23	1.00	3.79	0.00
Synj2	1.38	0.31	1.46	0.43	-0.18	1.00	0.29	1.00	0.11	0.90
Tbxas1	0.72	0.58	0.07	1.00	0.70	1.00	-0.37	1.00	-1.03	0.01
Tdo2	-0.13	1.00	0.09	1.00	-0.10	1.00	0.07	1.00	1.07	0.01
Tecr	0.24	1.00	-0.46	1.00	0.50	1.00	-0.37	1.00	-1.24	0.00
Tm7sf2	0.02	1.00	-0.53	0.96	1.13	0.63	-0.81	0.89	-1.78	0.00
Treh	1.07	0.13	1.88	0.01	2.05	0.01	-0.59	1.00	-0.20	0.66
Tsta3	0.23	1.00	-0.35	1.00	0.36	1.00	-0.36	1.00	-1.22	0.00
Uap1l1	0.65	0.59	0.55	0.94	0.15	1.00	-0.05	1.00	-1.07	0.00

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Ugdh	0.50	0.81	0.58	0.85	-0.24	1.00	0.76	0.67	1.86	0.00
Ugt2b1	0.82	0.18	0.55	0.96	0.94	0.88	0.14	1.00	0.95	0.02
Ugt2b34	0.63	0.58	0.95	0.39	-0.01	1.00	0.71	1.00	1.43	0.00
Ugt2b35	0.60	0.59	0.86	0.41	0.02	1.00	0.70	1.00	1.54	0.00
Upp1	0.41	1.00	-0.27	1.00	0.96	0.91	-0.19	1.00	-1.63	0.00
Upp2	1.96	0.01	2.20	0.00	0.47	1.00	-0.20	1.00	0.74	0.04

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Supplemental Table 8. Gene names and primer sequences for genes measured by RT-qPCR.

Mouse Gene	Forward primer	Reverse primer
Alg12	CCGTGGTTTATGTGCTCTCG	TCCGAACTGCTGTCTCACTT
Gclc	CAGGTGACATTCCAAGCCTG	CTCCCAGCGACAATCAATG
Nqo1	TATCCTTCCGAGTCATCTCTAGCA	TCTGCAGCTTCCAGCTTCTTG