

Supplementary Information:

The Fatty Acid β -Oxidation Pathway is Activated by Leucine Deprivation in HepG2 Cells: A Comparative Proteomics Study

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Content:

Supplementary Figure S1: Page 2

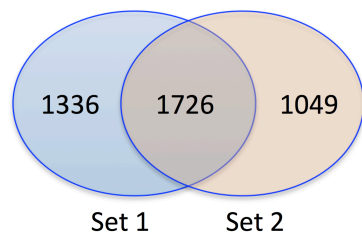
Supplementary Figure S2: Page 3

Supplementary Figure S3: Page 4

Supplementary Figure S4: Page 5

Supplementary Table S1: Pages 6-7

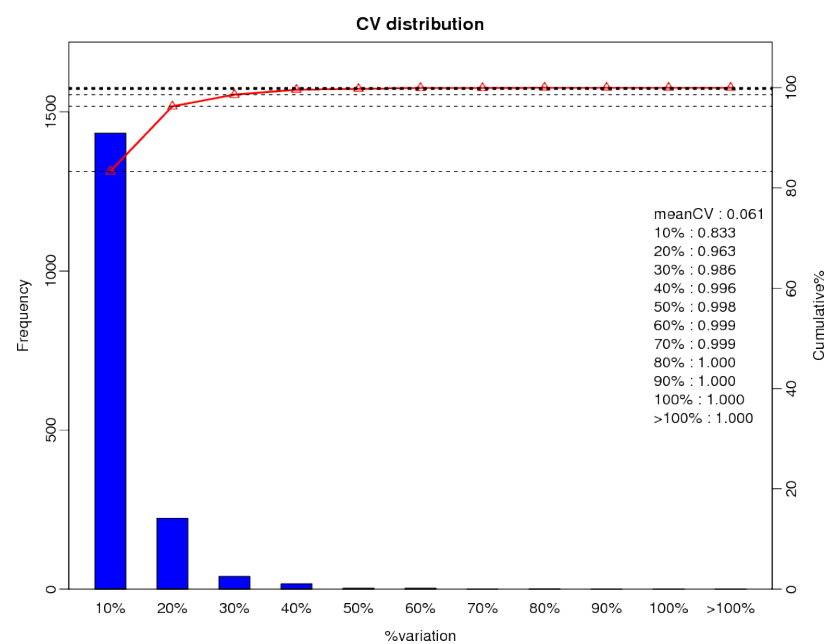
Supplementary Figure S1:



Supplementary Figure S1. Venn diagram of the number of quantified proteins derived from the two sets of iTRAQ experiments.

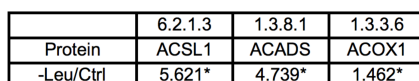
Supplementary Figure S2:

-Leu/Ctrl, Set1 vs Set2:

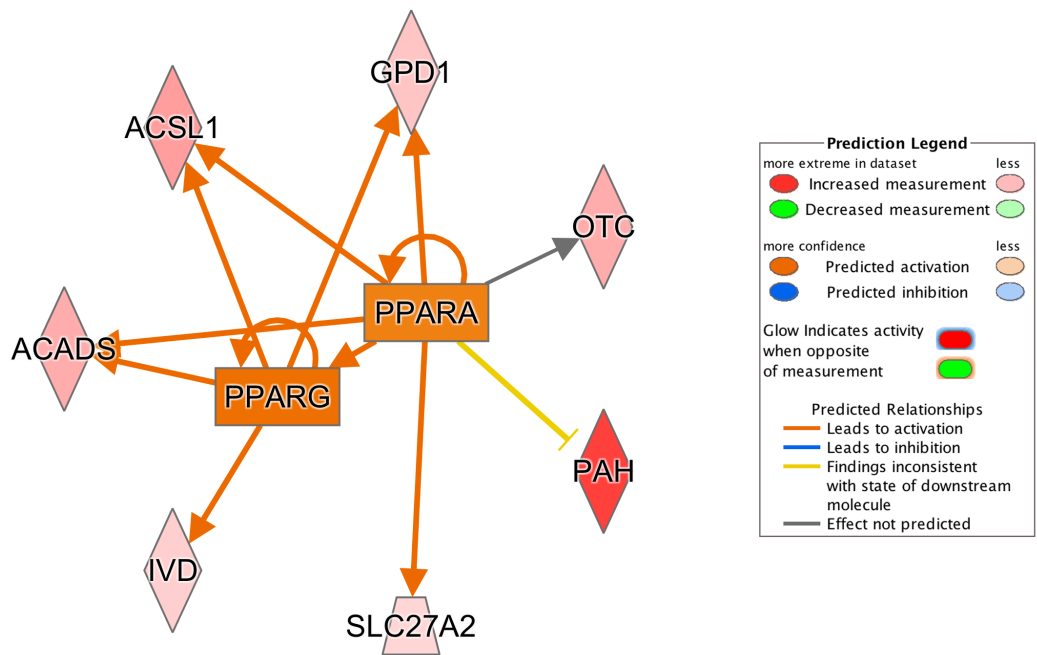


Supplementary Figure S2. The variation between the two iTRAQ runs.

Supplementary Figure S3. The information of “Fatty Acid Degradation” pathway from the KEGG database. Yellow, the proteins differentially regulated by Leu deprivation.



Supplementary Figure S4:



Supplementary Figure S4. Representative upstream analysis results of the differentially expressed proteins in Leu-deprived HepG2 cells using the IPA tools.

Supplementary Table S1. The details of functional characterization of the differentially expressed proteins for -Leu/Ctrl.			
Molecular and cellular function			
Category	p-value	Molecules	# Molecules
Amino Acid Metabolism	1.59E-10	AGXT2, BHMT, DMGDH, HAL, IVD, OTC, PAH, PCCB, QDPR, RGN	10
Small Molecule Biochemistry	1.59E-10	ACADS, ACSL1, ADH1C, ADK, ALDH1A1, ALDH8A1, AGXT2, BHMT, CDC42, CTSB, CYP2A6 (includes others), CYP2D6, DMGDH, GK, GPD1, H2AFY, HAL, HBB, IVD, KRT1, NPC2, OTC, PAH, PCCB, QDPR, RBP1, RGN, SLC27A2, STARD10, XDH	30
Drug Metabolism	6.25E-09	ACSL1, ADH1C, ALDH1A1, ALDH8A1, CTSB, CYP2A6 (includes others), CYP2D6, RBP1, XDH	9
Lipid Metabolism	6.25E-09	ACADS, ACSL1, ADH1C, ALDH1A1, ALDH8A1, BHMT, CDC42, CTSB, CYP2A6 (includes others), CYP2D6, GK, H2AFY, HBB, NPC2, PCCB, RBP1, RGN, SLC27A2, STARD10, XDH	20
Vitamin and Mineral Metabolism	6.25E-09	ADH1C, ALDH1A1, ALDH8A1, CYP2A6 (includes others), CYP2D6, KRT1, NPC2, RBP1, RGN, SLC27A2, XDH	11
Molecular Transport	1.28E-07	ACADS, ACSL1, ADH1C, ADK, ALDH1A1, ALDH8A1, BHMT, CDC42, CTSB, CYP2D6, H2AFY, HBB, NPC2, PAH, RBP1, RGN, SLC27A2, STARD10, XDH	19
Energy Production	7.06E-07	ACADS, ACSL1, ADH1C, ALDH1A1, CYP2D6, GPD1, HBB, RBP1, SLC27A2, XDH	10
Nucleic Acid Metabolism	1.02E-05	ADK, BHMT, CDC42, CYP2A6 (includes others), CYP2D6, GNB2, GPD1, XDH	8
Cell-To-Cell Signaling and Interaction	2.42E-05	CDC42, CTSB, CYP2D6, NPC2, XDH	5
Cell Death and Survival	1.27E-04	ADK, CTSB, HBB, KRT10, NPC2, WDR81, XDH	7
Cellular Compromise	1.27E-04	ACSL1, CDC42, CTSB, CYP2D6, NPC2, WDR81, XDH	7
RNA Post-Transcriptional Modification	1.80E-04	CYP2D6, XDH	2
Free Radical Scavenging	8.16E-04	CDC42, CYP2A6 (includes others), CYP2D6, HBB, XDH	5
Carbohydrate Metabolism	1.27E-03	ACSL1, ALDH1A1, BHMT, CDC42, CTSB, GK, GPD1, RBP1, RGN, XDH	10
Cellular Growth and Proliferation	1.28E-03	ALDH1A1, BHMT, CDC42, CTSB, KRT10, TIMELESS, XDH	7
Cell Morphology	2.04E-03	CDC42, CTSB, KRT1, NPC2, XDH	5
Cellular Assembly and Organization	2.04E-03	CDC42, CTSB, KRT1, XDH	4
Cellular Development	2.04E-03	ALDH1A1, BHMT, CDC42, CTSB, KRT10, NPC2, TIMELESS, XDH	8
Cellular Function and Maintenance	2.04E-03	CDC42, XDH	2
Cellular Movement	2.04E-03	CDC42, CTSB, RGN, XDH	4
DNA Replication, Recombination, and Repair	2.04E-03	ADK, CDC42, GPD1	3
Immune Cell Trafficking	2.04E-03	CDC42	1
Protein Synthesis	2.75E-03	ACADS, ALDH1A1, CDC42, CTSB, HBB, IVD, RIDA	7
Cell Cycle	4.07E-03	CDC42	1
Gene Expression	4.07E-03	RIDA	1
Post-Translational Modification	4.07E-03	ACADS, ALDH1A1, CDC42, CTSB, OTC, XDH	6
Cell Signaling	8.12E-03	CDC42, OTC, XDH	3
Protein Trafficking	8.12E-03	CDC42	1
Physiological system development and function			
Category	p-value	Molecules	# Molecules
Tissue Morphology	1.27E-04	ACSL1, ALDH1A1, BHMT, CDC42, CTSB, NPC2, WDR81, XDH	8
Embryonic Development	9.83E-04	ALDH1A1, BHMT, CDC42, HBB, NPC2, RBP1, RGN, TIMELESS	8
Organ Development	9.83E-04	ADK, ALDH1A1, CTSB, HBB, NPC2, RBP1, RGN, TIMELESS, XDH	9
Organismal Development	9.83E-04	ALDH1A1, BHMT, CDC42, CTSB, H2AFY, HBB, NPC2, RBP1, RGN, STARD10, TIMELESS, XDH	12
Respiratory System Development and Function	9.83E-04	CDC42, HBB, NPC2, RBP1, RGN, TIMELESS	6

Tissue Development	9.83E-04	ALDH1A1, CDC42, CTSB, HBB, KRT10, NPC2, RBP1, RGN, RIDA, TIMELESS, XDH	11
Cardiovascular System Development and Function	2.04E-03	CDC42, CTSB, XDH	3
Nervous System Development and Function	2.04E-03	ALDH1A1, CDC42	2
Tumor Morphology	2.04E-03	CTSB, RGN	2
Connective Tissue Development and Function	2.44E-03	ALDH1A1, CDC42, CTSB, NPC2, XDH	5
Inflammatory Response	3.70E-03	ADK, CTSB, GPD1, HBB, KRT1, KRT10, NPC2, RIDA, XDH	9
Digestive System Development and Function	4.07E-03	ADK, BHMT, CDC42, CTSB, RBP1, RGN, STARD10, XDH	8
Hepatic System Development and Function	4.07E-03	ADK, BHMT, CTSB, RBP1, RGN, STARD10, XDH	7
Organ Morphology	4.07E-03	ALDH1A1, BHMT, CTSB, H2AFY, HBB, KRT1, NPC2, RBP1, RGN, STARD10	10
Reproductive System Development and Function	4.07E-03	CDC42	1
Skeletal and Muscular System Development and Function	4.07E-03	ALDH1A1, CDC42, CTSB	3
Visual System Development and Function	4.07E-03	ALDH1A1, RBP1	2
Renal and Urological System Development and Function	6.09E-03	BHMT, CDC42, TIMELESS, XDH	4
Endocrine System Development and Function	8.12E-03	CTSB	1
Humoral Immune Response	1.22E-02	CTSB	1
Behavior	1.42E-02	CYP2A6 (includes others)	1
Hair and Skin Development and Function	1.42E-02	CTSB, KRT1	2
Hematological System Development and Function	1.62E-02	NPC2	1
Hepatotoxicity			
Category	p-value	Molecules	# Molecules
Liver Steatosis	6.69E-07	ACADS, ACSL1, ADK, BHMT, GPD1, H2AFY, RGN, STARD10	8
Liver Cholestasis	6.24E-06	ACSL1, ADH1C, GK, PAH	4
Hepatocellular Carcinoma	2.29E-03	AGXT2, ALDH8A1, BHMT, H2AFY, HAL, HBB, RGN	7
Liver Hyperplasia/Hyperproliferation	2.29E-03	ACSL1, AGXT2, ALDH1A1, ALDH4A1, ALDH8A1, BHMT, CNOT1, CTSB, CYP2A6 (includes others), CYP2D6, H2AFY, HAL, HBB, INTS1, IVD, KRT10, PAH, RGN	18
Liver Fibrosis	4.07E-03	ADK, CTSB, XDH	3
Liver Proliferation	4.07E-03	CTSB, XDH	2
Liver Damage	1.22E-02	ADK, ALDH1A1, CTSB, RGN	4
Liver Inflammation/Hepatitis	1.22E-02	ADK	1
Liver Necrosis/Cell Death	3.65E-02	CTSB, RGN	2
Liver Cirrhosis	8.09E-02	ADK, PAH	2