SUPPLEMENTAL INFORMATION

Genome-Wide Transcriptome Analysis Reveals Intermittent Fasting-Induced Metabolic Rewiring in the Liver

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SUPPLEMENTAL INFORMATION

Supplemental Information contains:

- Two Supplemental Figures and Figure Legends.
- Ten Supplemental Table Legends.

Figure S1 | Experimental Design Workflow.
At three months old, mice were randomly assigned and subjected to *ad libitum* (AL), intermittent fasting for 16-hours (IF16H) and 24-hours (hereby termed as every-other-day (EOD)). The mice were intermittently fasted for a period of three months. After which, the animals were anesthetized and euthanized before the left lateral liver lobe (LLL) was harvested. RNA samples from the liver was isolated, and cDNA libraries were constructed, before sequencing using HiSeq™ 2500 Illumina platforms to obtain 12GB of raw data per sample. Read counts obtained were then aligned to *mus musculus* (mm10) reference genome, and downstream data analysis was performed for liver transcriptome.

Figure S2 | Pearson Correlation Coefficients for Liver
Pearson correlation coefficients of minimal 0.9 value obtained for liver transcriptome samples demonstrated high coverage and reproducibility.

Table S1 | Differential Enriched Gene Ontologies for IF16 against AL in the Liver.
Genes queries were sent to clusterProfiler R package to determine gene ontologies (GO) following gene length biasness correction. GO terms with corrected p-value<0.05 were significantly differentially enriched. Gene identity is presented in Gene symbol format.
Key:
BP: Biological Processes
CC: Cellular Components
MF: Molecular Functions

Table S2 | Differential Enriched Gene Ontologies for EOD against AL in the Liver.
Genes queries were sent to clusterProfiler R package to determine gene ontologies (GO) following gene length biasness correction. GO terms with corrected p-value<0.05 were significantly differentially enriched. Gene identity is presented in Gene symbol format.
Key:
BP: Biological Processes
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Table S3 | Differential Pathways Enrichment for IF16 against AL in the Liver.
Genes queries were sent to clusterProfiler R package to determine pathway enrichment following gene length biasness correction. Statistical enrichment of differentially expressed genes were being interrogated in KEGG pathways database. KEGG terms with corrected p-value<0.05 were significantly differentially enriched. Gene identity is presented in Gene symbol format.
Key:
KEGG: Kyoto Encyclopaedia of Genes and Genomes

Table S4 | Differential Pathways Enrichment for EOD against AL in the Liver.
Genes queries were sent to clusterProfiler R package to determine pathway enrichment following gene length biasness correction. Statistical enrichment of differentially expressed genes were being interrogated in KEGG pathways database. KEGG terms with corrected p-value<0.05 were significantly differentially enriched. Gene identity is presented in Gene symbol format.
Key:
KEGG: Kyoto Encyclopaedia of Genes and Genomes

Table S5 | Reactome Analysis List of Differentially Expressed Genes for IF16 against AL in the Liver.
Detailed pathways and their respective interactors list are presented here.

Table S6 | Reactome Analysis List of Differentially Expressed Genes for EOD against AL in the Liver.
Detailed pathways and their respective interactors list are presented here.

Table S7 | Gene Ontologies & Pathways Enrichment of Top 50 Differentially Expressed Common Genes for IF16 and EOD against AL in the Liver.
Genes queries were sent to gProfiler online software to determine gene ontologies and pathways enrichment without hierarchical filtering. p-value is determined using g:SCS algorithm method provided by the software. g:SCS employed multiple testing correction for p-values obtained from gene ontologies and pathways enrichment analysis. A threshold *t*, which corresponds to the 5% upper quantile of randomly generated queries of input query size, will be multiplied to an initial experiment-wide threshold of *a=0.05* (at least 95% of matches above given threshold are statistically significant), in order to obtain the corrected p-values as shown in the table. Enrichment score is expressed in percentage. Gene identity is presented in Gene symbol format.
Key:
BP: Biological Processes
CC: Cellular Components
MF: Molecular Functions
KEGG: Kyoto Encyclopaedia of Genes and Genomes

Table S8 | List of Top 50 Differentially Expressed Common Genes for IF16 and EOD against AL in the Liver.
Respective $\log_2$ fold change for each common gene that were differentially expressed in both IF16 and EOD as compared to AL was shown. Gene identity is expressed in Gene symbol format. Differential gene expression was achieved using DESeq2. The resultant p-values were then adjusted using Benjamini and Hochberg's test to control false discovery rate (FDR). Genes with adjusted $p < 0.05$ were assigned as differentially expressed. Top 50 common genes were selected by descending ranking of total FPKM average value of each gene.

Table S9 | Gene Ontologies List for Differentially Expressed Genes for IF16 against AL in the Liver.

Genes queries were sent to gProfiler online software to determine gene ontologies and pathways enrichment without hierarchical filtering. p-value is determined using g:SCS algorithm method provided by the software. g:SCS employed multiple testing correction for p-values obtained from gene ontologies and pathways enrichment analysis. A threshold *t*, which corresponds to the 5% upper quantile of randomly generated queries of input query size, will be multiplied to an initial experiment-wide threshold of *a=0.05* (at least 95% of matches above given threshold are statistically significant), in order to obtain the corrected p-values as shown in the table. Enrichment score is expressed in percentage. Gene identity is presented in Gene symbol format.

Key:

BP: Biological Processes

CC: Cellular Components

MF: Molecular Functions

T: Number of Gene Terms in Textual Format

Q: Number of Query Genes

Q&T: Number of Common Genes

Table S10 | Gene Ontologies List for Differentially Expressed Genes for EOD against AL in the Liver.

Genes queries were sent to gProfiler online software to determine gene ontologies and pathways enrichment without hierarchical filtering. p-value is determined using g:SCS algorithm method provided by the software. g:SCS employed multiple testing correction for p-values obtained from gene ontologies and pathways enrichment analysis. A threshold *t*, which corresponds to the 5% upper quantile of randomly generated queries of input query size, will be multiplied to an initial experiment-wide threshold of *a=0.05* (at least 95% of matches above given threshold are statistically significant), in order to obtain the corrected p-values as shown in the table. Enrichment score is expressed in percentage. Gene identity is presented in Gene symbol format.

Key:

BP: Biological Processes

CC: Cellular Components

MF: Molecular Functions

T: Number of Gene Terms in Textual Format

Q: Number of Query Genes

Q&T: Number of Common Genes