SUPPLEMENTAL INFORMATION

Supplemental Figures

Fig. S1 Liver weight and hepatic/plasma metabolites in *Lrh-1*$^{AAV8\text{-}GFP}$ and *Lrh-1*$^{AAV8\text{-}Cre}$ mice on SD.

Fig. S2 *Lrh-1*$^{AAV8\text{-}Cre}$ fed SD exhibit normal rate of VLDL secretion and improved plasma cholesterol profile on HFD.

Fig. S3 *Lrh-1*$^{AAV8\text{-}Alb\text{-}Cre}$ mice fed HFD do not exhibit significant changes in lipid metabolism.

Fig S4. Volcano plot of DEGs in *Lrh-1*$^{AAV8\text{-}Cre}$ fed HFD.

Fig. S5 Validation of DEGs and expression of known risk alleles for NAFLD in *Lrh-1*$^{AAV8\text{-}Cre}$ mice fed standard diet.

Fig. S6 Additional phospholipid totals showing elevated LPL in *Lrh-1*$^{AAV8\text{-}Cre}$ livers.

Fig. S7 Additional genes complemented by hLRH-1, hepatic/plasma cholesterol and plasma ALT measurements in *Lrh-1*$^{AAV8\text{-}Cre+hLrh1}$.

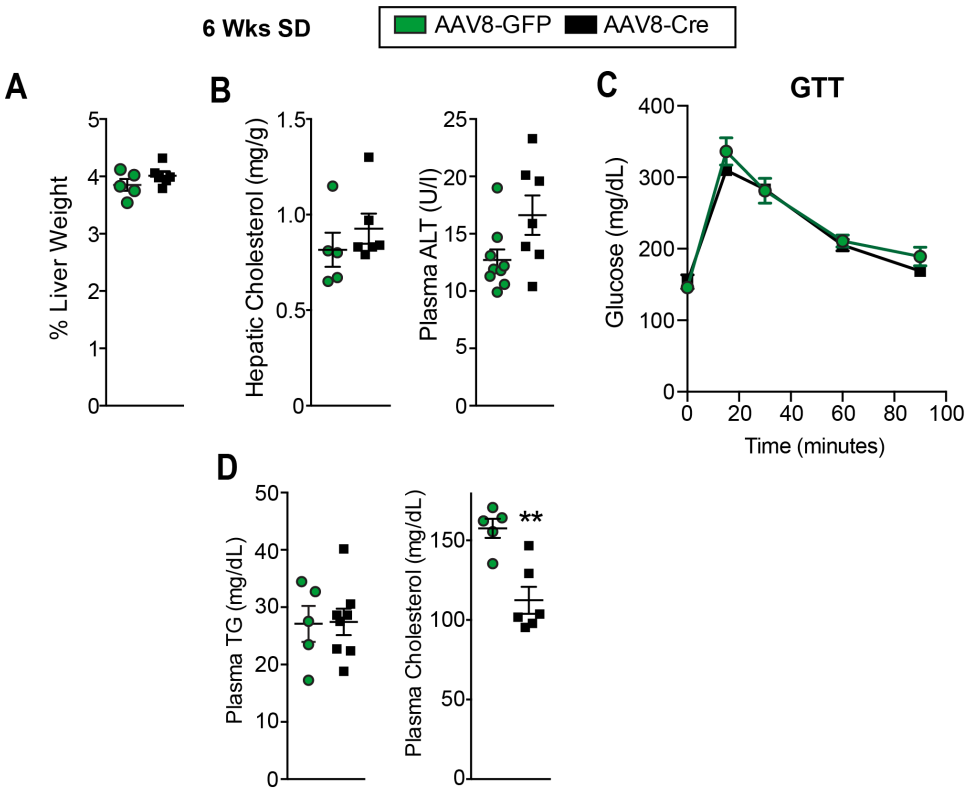
Fig. S8 Profile of early changes in gene expression and plasma cholesterol levels after acute ablation of LRH-1 in livers of *Lrh-1*$^{AAV8\text{-}Cre}$.

Supplemental Tables

Table S1 Sequences of forward and reverse primers used in this study.

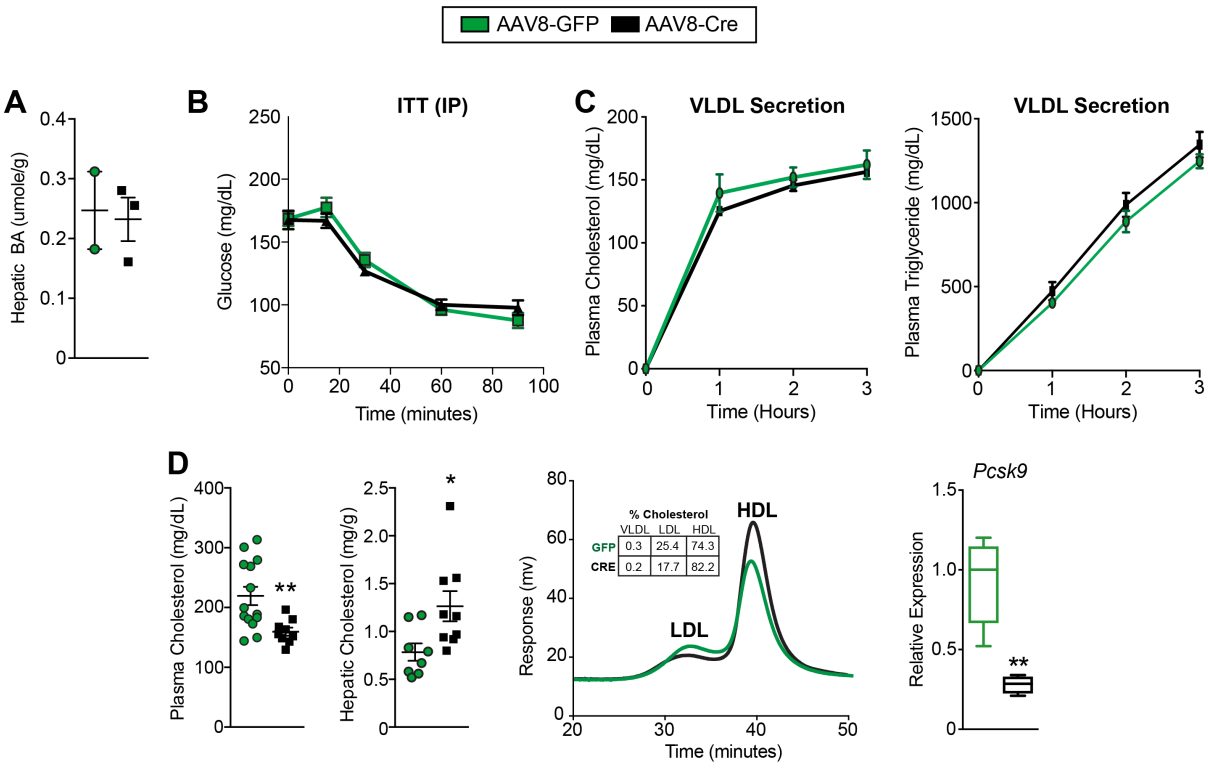
Supplemental Data Files

Data Set 1 - Significant DEG changes on SD and HFD after loss of LRH-1 on separate tabs.



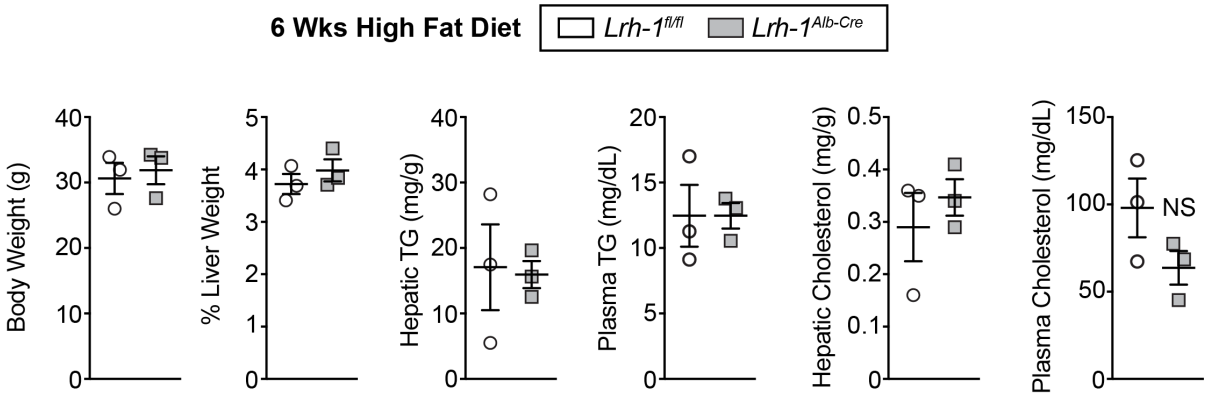
Supplemental Fig. 1 Liver weights and hepatic/plasma metabolites in *Lrh-1*$^{AAV\text{-}GFP}$ and *Lrh-1*$^{AAV\text{-}Cre}$ mice on standard diet (SD).

(A) Percent liver weight of *Lrh-1*$^{AAV8\text{-}GFP}$ and *Lrh-1*$^{AAV8\text{-}Cre}$ adult male mice fed standard diet 8 wks post infection **(B)** Measurement of hepatic cholesterol levels and plasma ALT levels on fasted *Lrh-1*$^{AAV8\text{-}GFP}$ and *Lrh-1*$^{AAV8\text{-}Cre}$. **(C)** GTT (i.p.) assay on fasted *Lrh-1*$^{AAV8\text{-}GFP}$ and *Lrh-1*$^{AAV8\text{-}Cre}$. **(D)** Measurements of plasma lipids from fasted *Lrh-1*$^{AAV\text{-}GFP}$ and *Lrh-1*$^{AAV8\text{-}Cre}$ mice; n = 5 and n = 6 per group, respectively. Error bars represent ± SEM. Unpaired Student's *t* test for panels (a, d).



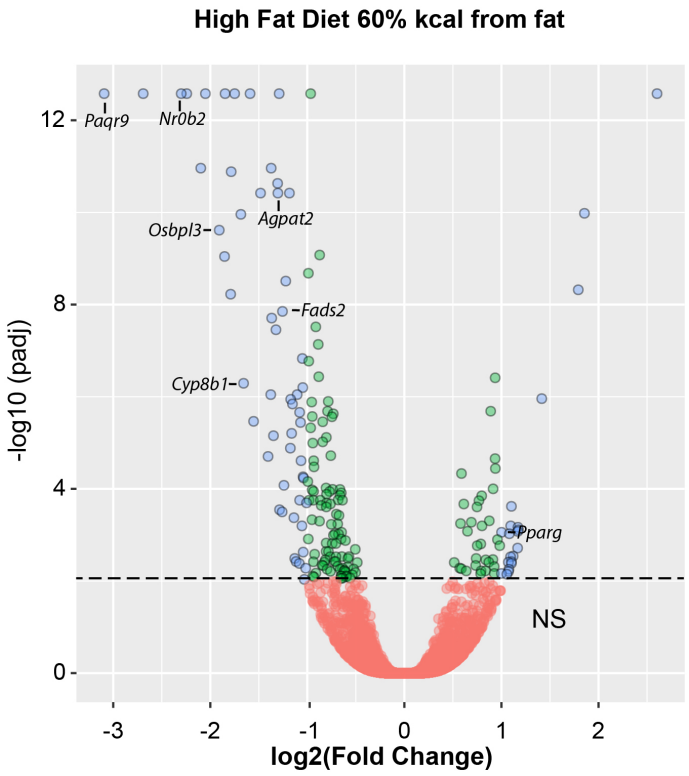
Supplemental Fig. 2 *Lrh-1*$^{AAV\text{-}Cre}$ mice exhibit normal rate of VLDL secretion on SD and improved plasma cholesterol profile on HFD.

(A) Measurement of hepatic bile acids from livers of *Lrh-1*$^{AAV8\text{-}GFP}$ and *Lrh-1*$^{AAV8\text{-}Cre}$ male mice fed HFD for 6 wks and 8 wks post infection; n = 2, n = 3, respectively. **(B)** ITT on fasted *Lrh-1*$^{AAV8\text{-}GFP}$ and *Lrh-1*$^{AAV8\text{-}Cre}$ mice fed HFD, n = 8 and n = 6 per group. **(C)** Assessment of VLDL secretion in *Lrh-1*$^{AAV8\text{-}GFP}$ and *Lrh-1*$^{AAV8\text{-}Cre}$ fed SD with n = 2 and n = 4 per group, respectively. **(D)** Hepatic/plasma cholesterol, lipoprotein profile and qPCR analysis of *Pcsk9* expression in livers from *Lrh-1*$^{AAV8\text{-}GFP}$ and *Lrh-1*$^{AAV8\text{-}Cre}$ mice fed HFD. For plasma cholesterol n = 14 and n = 9 per group, for lipoprotein profile n = 5 per group, and for *Pcsk9* expression, n = 5 and n = 4 per group. Error bars represent ± SEM. Unpaired Student's *t* test for panel (D).



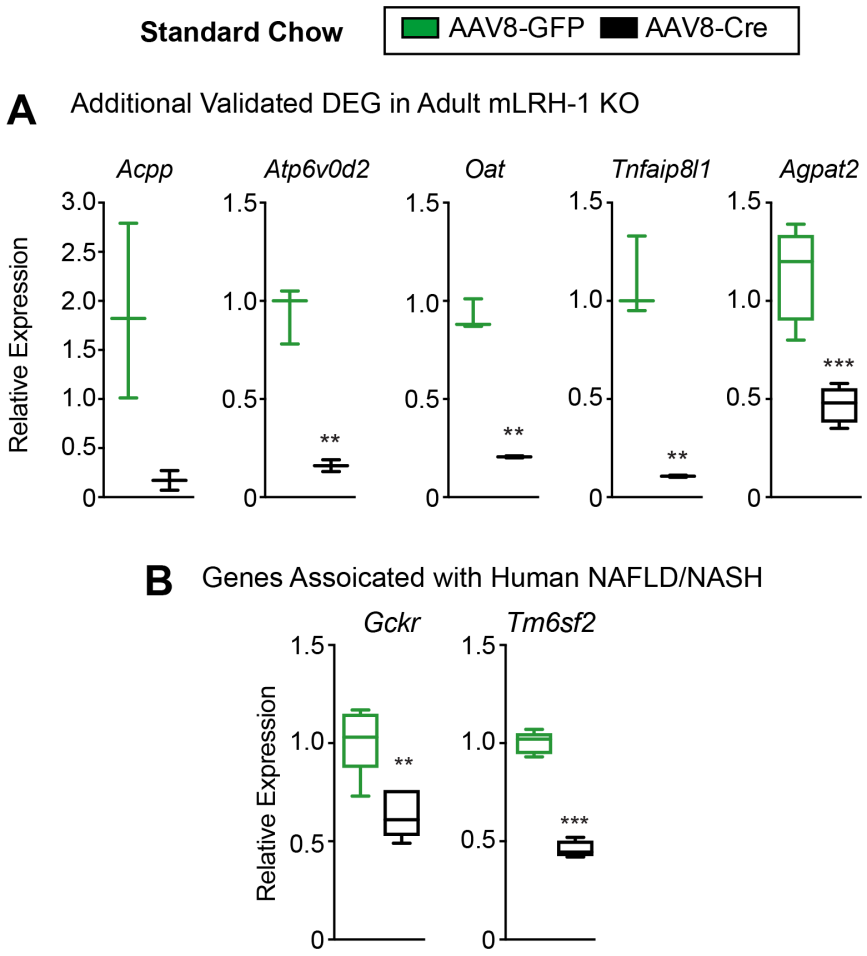
Supplemental Fig. 3 *Lrh-1*$^{AAV\text{-}Alb\text{-}Cre}$ mice fed high fat diet do not exhibit significant changes in lipid metabolism.

Body weight and measurement of plasma/hepatic metabolites of adult male *Lrh-1*$^{fl/fl}$ and *Lrh-1*$^{AAV\text{-}Alb\text{-}Cre}$ mice fed high fat diet. Number of mice per group was n = 3. Error bars represent ± SEM.



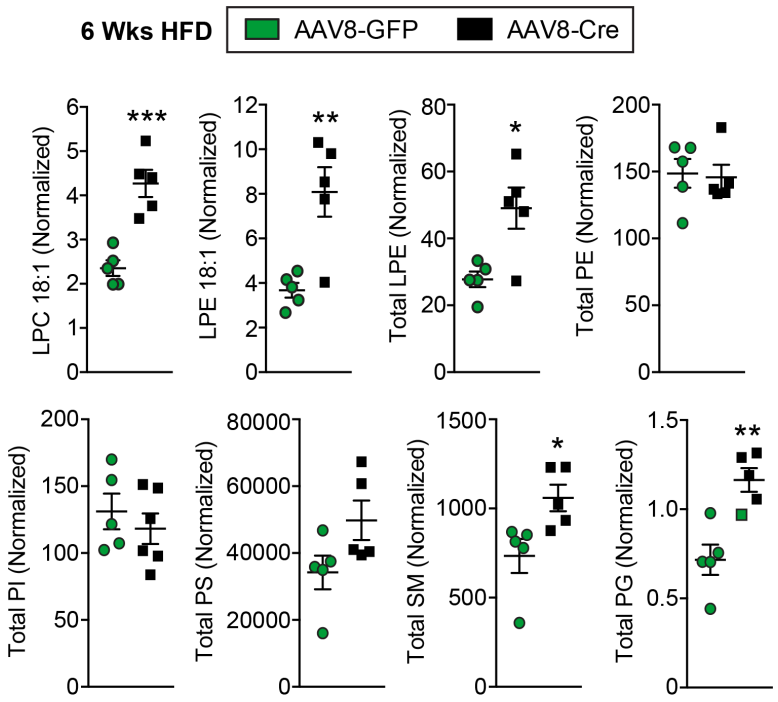
Supplemental Fig 4. Volcano plot of DEGs in *Lrh-1*$^{AAV8\text{-}Cre}$ fed HFD.

Volcano plot of DEGs between *Lrh-1*$^{AAV8\text{-}GFP}$ and *Lrh-1*$^{AAV8\text{-}Cre}$ mice fed HFD with DEGs of an adjusted p-value <0.01 shown above the black dashed line (green circles), and those with <0.01 and absolute log2 fold change > 1.0 highlighted (blue circles).



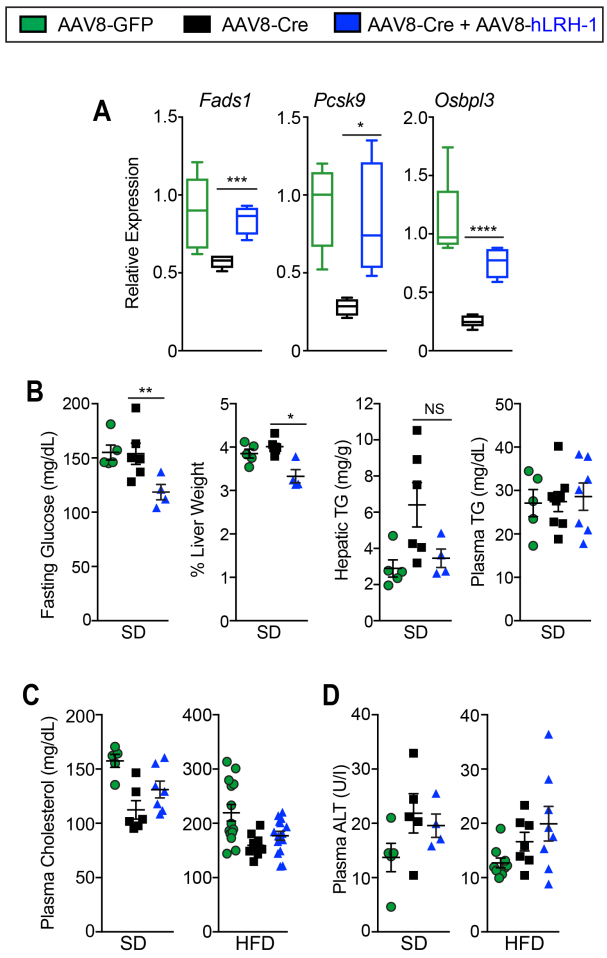
Supplemental Fig. 5 Validation of DEGs in *Lrh-1*$^{AAV8\text{-}Cre}$ mice fed standard diet and expression of known risk alleles for NAFLD.

(A) Relative gene expression by qPCR of *Accp*, *Atp6v0d2*, *Oat*, *Tnfaipa8l1* and *Agpat2* in livers from *Lrh-1*$^{AAV8\text{-}GFP}$ or *Lrh-1*$^{AAV8\text{-}Cre}$ male mice fed standard diet 8 wks post infection, n = 2, n = 3 per group. **(B)** qPCR analysis of known NAFLD risk alleles *Gckr* and *Tm6sf2* in livers from *Lrh-1*$^{AAV8\text{-}GFP}$ or *Lrh-1*$^{AAV8\text{-}Cre}$ mice fed standard diet, n = 5 per group. Error bars represent ± SEM. Unpaired Student's *t* test for panels A and B.



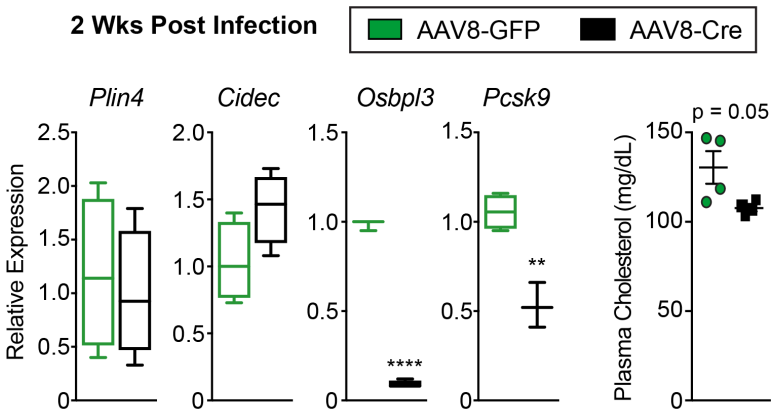
Supplemental Fig. 6 Additional Measurements for Total Phospholipids.

Normalized values for total oleic acid lysophosphatidylcholine (LPC 18:1), oleic acid lysophosphatidylethanolamine (LPE 18:1), lysophosphatidylethanolamine (LPE), phosphatidylethanolamine (PE), phosphoinositol (PI), phosphatidylserine (PS), sphingomyelin (SM) and phosphatidylglycerol (PG) in control and *Lrh-1*$^{AAV\text{-}Cre+hLrh1}$ male mice fed HFD for 6 wks and 8 wks post infection; n = 5 per group. Error bars represent ± SEM. Unpaired Student's *t* test for all bar graphs.



Supplemental Fig. 7 Additional genes complemented by hLRH-1, hepatic/plasma cholesterol and plasma ALT measurements in *Lrh-1*$^{AAV\text{-}Cre+hLrh1}$.

(A) qPCR analysis of *Fads, Pcsk9 and Osbpl3* on livers of *Lrh-1*$^{AAV\text{-}Cre+hLrh1}$ male mice fed standard diet, 8 wks post infection. **(B)** Measurement of fasting glucose, % liver weight and TG values for mice as indicated maintained on standard diet with number per group shown in individual bars. **(C)** Measurement of fasting plasma cholesterol and plasma ALT in *Lrh-1*$^{AAV\text{-}Cre+hLrh1}$ fed standard diet (SD) or HFD. Number per group is indicated on bar graphs. Error bars represent ± SEM. Unpaired Student's *t* test for panel A.



Supplemental Fig. 8. Changes in hepatic gene expression and plasma cholesterol after acute ablation of LRH-1 in livers (*Lrh-1*$^{AAV\text{-}Cre}$) compared to control mice.

Hepatic expression of *Cidec*, *Plin4*, *Osbpl3* and *Pcsk9* transcripts and fasting plasma cholesterol measurements in control and *Lrh-1*$^{AAV\text{-}Cre}$ mice fed standard diet 2 wks post-infection. Number per group n = 4. Error bars represent ± SEM. Unpaired Student's *t* test.

Supplementary Table S1 – Forward and Reverse Primers Used in Study

Name	Forward 5'-3'	Reverse 5'-3'
36b4	GGCACCGAGGCAACAGTT	TCATCCAGCAGGTGTTTGACA
Acacb	GGAACTGCTGCTTCCCATCTA	GCAGTCTTCCATTCCAGCACA
Acpp	AGCCACCAAGGACGGAATT	TGAAACCCGCTTGACATCAG
Acta2	AGATCATTGCCCCTCCAGAAC	CCAGCTTCGTCGTATTCCTGT
Acpp	AGCCACCAAGGACGGAATT	TGAAACCCGCTTGACATCAG
Agpat2	GAGGGTACACGCAACGACAAT	AGAAAGACGAGTACACCACGG
Atp6v0d2	GAGCTGTACTTCAATGTGGACCAT	TGAGTTAGGAGGCTGGCTTTG
Cidec	AGGACTTTATTGGCTGCCTG	CATGCTGAAGAGGGTCCAG
Col1a1	CTTCACCTACAGCACCCTTGT	GGGAGGTCTTGGTGGTTTTGT
Elovl5	TGGGAAGGCAAATACAACTT	GTCCATGAATTCGATGAGTT
Fads1	GTTCACCAATCAGCCTTCAAC	TCATACTTGATGCCGTACTTGG
Fads2	TGAAGTCTCTCTGCGCCAAG	CAGCTCCCCAGACTTCTTCAG
Fasn	CCAAGACTGACTCGGCTACTG	TCAGGTTCAGTTGGTTCTGCT
Gckr	CCATCTCCTGCCATGTCCAG	CCTTAGCCTCAGTGATGGAGC
Gnmt	CCCACATGGTAACCCTGGAC	GAAAGACGCCAAACAGTGTGG
Hamp2	CTTTGCACGGGGAAGAAAGC	TGCAGATGGGGAAGTTGATGT
mLrh-1	CCATTACGGTCTCCTCACGTG	AGGGACATCGTTTTCTCTGCG
hLRH-1	CAGAGAAAGCGTTGTCCTTACTG	TTATTCCTTCCTCCACGCATT
Oat	TGGCGGTTTATACCCTGTGT	CCTGGTTTAATGGTCAGCATTATC
Osbpl3	GGATCGAGAAGCTGCAGAGAG	GGTAGGTCCCATTGCTTACCC
Paqr9	CGAGAACCCCACACTCTTCG	CTCAGGGATCTTGCTCACGTT
Pcsk9	AATCAAGGAGCATGGGATCTC	ACATTGCATCCAGTCAGGG
Pctp	CGTCCTGGCTCATTAACTGGG	TGTGGTAGTTCTGACACGCTT
Pcyt1a	TGGAAGAGAAGAGCATCGACC	CATCCGACCTTTTCCCTCCTT
Pemt	GGCAATTGTCTACGTGGTTGC	TGCAACCTGGTAGCTTTCTGT
Plcxd2	CATTCTGCCATAGTTTTCAAGTGA	CCCCCGATATTAGAGAACATGTG
Plin4	AAACAGCAACAGACCCCTC	AACTTCCCATGTCCTTGTCTC
Scd1	AGAACTTACAAGGCACGGCT	TTCTGAGAACTTGTGGTGGGC
Shp	ACGATCCTCTTCAACCCAGAT	GTACCAGGGCTCCAAGACTTC
Slc27a2	TCTTTCAACACATCGCGGAGT	TGCGGTGTTTAAAAGTCCCAG
Slc27a5	TACTGGAGAGGTGGAGTGTGT	CAACCTTACCCTCACACCCTG
Srebp1c	GGTTTCCAACATGACCTGAGC	ATACCCTCCTCATAGCAGGCC
Tff3	AGTATCCCAAATGTGCCCTGG	CATCAGCAGCAGGAGTCCAG
Tm6sf2	ATTCTACTGCCTGGCTGCCTA	ACCCATGTGTGAGAACTGCG