Supplementary Figure Legends

Figure S1: Weight gain with age in mice fed different diets. (**A**) Age-dependent changes in body weights of male *Clk*$^{\Delta 19/\Delta 19}$ and *Clk*$^{wt/wt}$ mice in Figure S2A-H fed a chow diet. **P*<0.05, ***P*<0.01, multiple t-tests. (**B**) Three-month-old male *Clk*$^{\Delta 19/\Delta 19}$ and *Clk*$^{wt/wt}$ mice (Figure S2I-M) were fed a cholate-containing high fat diet *ad libitum* for 2 months, and body weights were measured. (**C**) Three-month-old mice were fed a Western diet. Body weights were measured at the end of 2 months (Figure S2N-T). Mean ± SD, **P*<0.05, ***P*<0.01, ****P*<0.001, unpaired Welch's two-tailed test. The data are representative of 2 experiments.

Figure S2: Effect of age and different diets on hepatic lipid accumulation in *Clk*$^{\Delta 19/\Delta 19}$ mice and their wild-type (*Clk*$^{wt/wt}$) siblings.

(**A-H**) Male *Clk*$^{\Delta 19/\Delta 19}$ and *Clk*$^{wt/wt}$ siblings (n=6/group) were fed a chow diet *ad libitum*. Plasma and liver samples were collected at 1 month (1 m), 3-4 months (3 m), 6-8 months (6 m), and 10-12 months (10 m). (**A**) Oil Red O staining showed increased hepatic lipid accumulation in *Clk*$^{\Delta 19/\Delta 19}$ mice at all ages. The data are representative of 2 independent experiments. (**B**) Hepatic triglyceride concentrations were significantly higher in older *Clk*$^{\Delta 19/\Delta 19}$ mice compared with same age *Clk*$^{wt/wt}$. (**C**) Plasma ALT levels were significantly higher in 10-month-old *Clk*$^{\Delta 19/\Delta 19}$ mice. (**D**) Mice (n=3 mice/group/time point) were IP injected with [^{3}H]OA (1 μCi/0.5 ml PBS). Livers were collected after 2 hours to measure radioactivity. (**E-H**) Changes in hepatic expressions of genes in (**E**) lipid metabolism, (**F**) ER stress, (**G**) inflammation, and (**H**) cancer were quantified in triplicate via quantitative real-time PCR. *Gapdh* mRNA levels were used as controls. The gene/Gapdh ratio in 3-month-old WT mice was normalized to 1. Mean ± SD, **P*<0.05, ***P*<0.01, ****P*<0.001, *****P*<0.0001, two-way ANOVA, Sidak's multiple comparison test.

(**I-M**) Male *Clk*$^{\Delta 19/\Delta 19}$ and *Clk*$^{wt/wt}$ littermates were weaned to a chow diet. At 3 to 4 months of age, mice were fed a cholate-containing high fat diet for 2 months and were used for tissue collection (n=6/group) or injected with [^{3}H]OA to study hepatic uptake (n=3-4/group). (**I**) Top, a representative picture of livers. *Clk*$^{\Delta 19/\Delta 19}$ livers were larger and weighed more. Below, *Clk*$^{\Delta 19/\Delta 19}$ liver sections stained with Oil Red O showed more lipid staining. The data are representative of 2 experiments. (**J**) Liver–to–body weight ratio and (**K**) hepatic triglyceride, cholesterol, free fatty acids, and TBARS levels were higher in *Clk*$^{\Delta 19/\Delta 19}$ mice. (**L**) Plasma ALT and AST levels increased in *Clk*$^{\Delta 19/\Delta 19}$ mice. (**M**) *Clk*$^{\Delta 19/\Delta 19}$ livers accumulated more [^{3}H]OA 2 hours after IP injections (n=3-4/group). Mean ± SD, ***P*<0.05, ****P*<0.001, unpaired Welch's two-tailed test.

(**N-T**) Male *Clk*$^{\Delta 19/\Delta 19}$ and *Clk*$^{wt/wt}$ littermates at 3-4 months of age were fed a Western diet for 2 months and were used for tissue collections (n=6 /group) or hepatic [^{3}H]OA uptake studies (n=3/group). (**N**) Liver triglyceride and (**O**) TBARS levels were higher in *Clk*$^{\Delta 19/\Delta 19}$ littermates. (**P**) Plasma ALT levels were higher in *Clk*$^{\Delta 19/\Delta 19}$ mice. (**Q**) Oil Red O staining revealed more lipid accumulation in *Clk*$^{\Delta 19/\Delta 19}$ mice. The data are representative of 2 experiments. (**R**) Hepatic fatty acid uptake after 2 hours of IP-injected [^{3}H]OA was higher in *Clk*$^{\Delta 19/\Delta 19}$ livers (n=3/group). (**S**) Fatty acid uptake and oxidation in liver slices. Mean ± SD,; ***P*<0.05, ****P*<0.001, unpaired

Welch's two-tailed test. (**T**) Expression analysis of candidate genes in lipid metabolism, inflammation, ER stress, and cancer in the livers of $Clk^{\Delta 19/\Delta 19}$ and $Clk^{wt/wt}$ mice. Mean ± SD, **P*<0.05, ***P*<0.01, ****P*<0.001, *****P*<0.0001, multiple comparison t-test.

Figure S3: Knockdown of CLOCK and SREBP1c has no effect on the expression of CD36 in Huh7 and primary mouse hepatocytes. (A, C) Huh7 cells and (**B, D, E**) mouse primary hepatocytes were treated with different siRNA in triplicate for 72 hours and were used to measure indicated mRNA levels. siClk, siSrebp1c, siGata4, siShp, and siUsf2 had no effect on CD36 expression in these liver cells.

Figure S4: Effect of knockdown of different genes and overexpression of CLOCK on CD36 expression in Huh7 cells. (**A**) Huh7 cells were transfected in triplicate with indicated siRNA and used to measure *CD36* and *GAPDH* mRNA levels after 72 hours. (**B**) Huh7 cells were transfected with indicated siRNA. After 72 hours, they were incubated with 0.5 μCi of [^{3}H]OA. After 1 hour, cells were washed, collected, and quantified in a scintillation counter. Mean ± SD; **P*<0.05, ***P*<0.01, ****P*<0.001, *****P*<0.0001, One-way ANOVA multiple comparisons.

(**C-D**) Huh7 cells were transfected with a control plasmid (Ctrl) or a plasmid expressing human CLOCK cDNA. After 72 hours, cells were used (C) to measure uptake of [^{3}H]OA. Mean ± SD, **P*<0.05, ***P*<0.01, ****P*<0.001, *****P*<0.0001, Unpaired Welch's two-tailed test. (D) In a separate experiment cells were used to measure mRNA levels. Mean ± SD, **P*<0.05, ***P*<0.01, ****P*<0.001, *****P*<0.0001, multiple t-tests.

Figure S5: Location of E-boxes and HIF1α binding sites in the promoters of different genes. (**A**) Human PHD2 (gene *EGLN1*) promoter contains one E-box. Human PHD3 (gene *EGLN3*) contains three E-boxes. In ChIP, we assessed the binding of CLOCK and BMAL1 to the −756 to −762 and −295 to −539 E-box for *EGLN1* and *EGLN3*, respectively. (**B**) Mouse *Egln1* and *Egln3* genes contain one and two E-boxes, respectively. (**C**) The human *CD36* gene contains three potential HIF1α binding sites. We assessed the binding of HIF1α to sites 1 and 2 of CD36 in ChIP assays. We identified one HIF1α binding site in the mouse *Cd36* promoter and studied the binding of HIF1α to this site in our ChIP assays.

Figure S6: Circadian expression of *Egln* and *Hif1α*: Livers were collected from *ad libitum* chow-fed mice (n=6/time point), and mRNA levels were measured by qPCR. Mean ± SD, **P*<0.05, ***P*<0.01, ****P*<0.001, *****P*<0.0001, Two-way ANOVA, Sidak's multiple comparison test.

Figure S7: Effect of $CoCl_2$ and DMOG on fatty acid uptake and oxidation in Huh7 cells: Huh7 cells were transfected with siCTRL or siCLK. After 72 hours, cells were treated with $CoCl_2$ (200 μM) or DMOG (200 μM) for 12 hours. Cells were used to measure fatty acid uptake of [^{3}H]OA for 1 hour and fatty acid oxidation of [^{14}C]OA for 2 hours. Mean ± SD, **P*<0.05, ***P*<0.01, ****P*<0.001, *****P*<0.0001, unpaired Welch's two-tailed test.

Figure S8: **LPS induced hepatosteatosis in female $Clk^{\Delta 19/\Delta 19}$ mice.**

Female (3-month-old) *Clk*$^{\Delta 19/\Delta 19}$ and *Clk*$^{wt/wt}$ mice on a chow diet were fed a Western diet for 2 months and IP injected with LPS (0.25 mg/kg/injection, three times on alternate days). Mice then continued on a Western diet for an additional 2 months, and livers and plasma were collected for analyses at the end (n=6/group). Another set of animals was used to study uptake of IP-injected [^{3}H]OA by the liver (n=6/group).

(**A**) Livers from LPS-injected *Clk*$^{\Delta 19/\Delta 19}$ mice showed accumulation of lipids after Oil Red O staining. The data are representative of 2 experiments.

(**B-C**) *Clk*$^{\Delta 19/\Delta 19}$ livers had higher levels of triglyceride, and cholesterol, and TBARS. Mean ± SD; ***P<0.0001, Welch's two-tailed t-test.

(**D**) Plasma ALT levels were higher in LPS-injected *Clk*$^{\Delta 19/\Delta 19}$ mice. In contrast, plasma β–HB concentration were lower in LPS-injected *Clk*$^{\Delta 19/\Delta 19}$ mice. Mean ± SD; ***P<0.001, Unpaired Welch's two-tailed t test.

(**E**) Hepatic fatty acid uptake and oxidation. LPS-injected *Clk*$^{\Delta 19/\Delta 19}$ mice took up more [^{3}H]OA and oxidized less [^{14}C] PA. **P<0.01, Welch's two-tailed t-test.

(**F-J**) Quantification of different indicated mRNA in the livers of LPS-injected *Clk*$^{\Delta 19/\Delta 19}$ and *Clk*$^{wt/wt}$ mice. Mean ± SD, *P<0.05, **P<0.01, ***P<0.001, ****P<0.0001, multiple t-tests.

(K) Proteins were quantified by Western blotting using specific antibodies. The data are representative of 2 experiments.

Figure S9: Changes in hepatic mRNA levels in *Clk*$^{\Delta 19/\Delta 19}$*Apoe*$^{-/-}$ and *Apoe*$^{-/-}$. Male (10-month-old) *Clk*$^{\Delta 19/\Delta 19}$*Apoe*$^{-/-}$ and *Apoe*$^{-/-}$ mice fed a chow diet were IP injected with LPS (0.25mg/kg) three times on alternate days. Two months after the last injection, livers were collected to measure mRNA levels (n= 6/group). Mean ± SD, *P<0.05, **P<0.01, ***P<0.001, ****P<0.0001, multiple t-tests.

Figure S10: Effect of oxLDL and $CoCl_2$ on CD36 and HIF1α expression in J774 cells.

(**A-C**) J774 cells were transfected with siCtrl, siClk or siHIF1α. After 72 hours, cells were treated with oxLDL (100 μg/ml) or $CoCl_2$ (200 μM) for 12 hours. Cells were used to measure mRNA and protein levels of CD36 and β-actin by real-time PCR and immunoblotting. Mean ± SD, *P<0.05, **P<0.01, ***P<0.001, ****P<0.0001, two-way ANOVA multiple comparisons. The data are representative of 2 experiments.

(**D**) J774 cells were first transfected with adenoviruses expressing shCtrl or shCLOCK. After 72 hours, confluent monolayers were incubated in serum free media for 18 hours and then incubated with media containing 50% serum and oxLDL (100 μg/ml) for 2 h. Media was changed to serum free media containing oxLDL (100 μg/ml). At different indicated times, temporal expression of different proteins was studied. The data are representative of 2 experiments.

Figure S11: Different stages and markers in the development of liver diseases.

Data are summarized to show changes in gene expression, biochemical determinations, and disease phenotype to illustrate changes in the development of liver diseases in *Clk*$^{\Delta 19/\Delta 19}$ and *Clk*$^{\Delta 19/\Delta 19}$ Apoe$^{-/-}$ mice. For comparison, data from 3-month-old chow-fed *Clk*$^{\Delta 19/\Delta 19}$ mice were normalized to 1, and changes with increasing age and other interventions in these mice were plotted as fold changes. Similarly, data from chow-fed 3-month-old *Apoe*$^{-/-}$ mice were normalized to 1, and other changes in *Clk*$^{\Delta 19/\Delta 19}$*Apoe*$^{-/-}$ mice were reported as fold changes from this number. WT, *Clk*$^{wt/wt}$ mice; Δ19, *Clk*$^{\Delta 19/\Delta 19}$ mice; e$^{-/-}$, *Apoe*$^{-/-}$ mice; Δ19/e$^{-/-}$, *Clk*$^{\Delta 19/\Delta 19}$*Apoe*$^{-/-}$ mice; CD, chow diet; WD, western diet.

(**A**) Older *Clk*$^{\Delta 19/\Delta 19}$ mice develop steatosis on a chow diet. The Western diet induces steatohepatitis in older *Clk*$^{\Delta 19/\Delta 19}$ mice. Additional insults, such as LPS or $CoCl_2$, enhanced steatohepatitis, and the disease advanced to cirrhosis in these mice. In contrast, *Clk*$^{\Delta 19/\Delta 19}$*Apoe*$^{-/-}$ mice developed steatohepatitis at young age and cirrhosis at an older age on a chow diet. These mice were more susceptible to additional insults, such as LPS and $CoCl_2$, and developed cirrhosis and hepatocellular carcinoma. Livers in advanced cirrhosis and carcinoma in LPS- and $CoCl_2$-injected mice did not show steatosis.

(**B**) Liver triglycerides increased with age or with a Western diet in *Clk*$^{\Delta 19/\Delta 19}$ mice. Hepatic triglyceride levels tended to be lower in cirrhosis and cancer after LPS and $CoCl_2$ injections.

(**C**) Fatty acid uptake by the *Clk*$^{\Delta 19/\Delta 19}$ liver increased in all conditions. In *Clk*$^{\Delta 19/\Delta 19}$*Apoe*$^{-/-}$ mice, liver fatty acid uptake increased until the liver was steatotic. Cirrhotic and cancerous livers took up fewer fatty acids.

(**D**) Liver TBARS levels increased throughout the progression of liver diseases in both mouse models. This might be an indicator of the severity of liver diseases in mice.

(**E**) Plasma ALT levels increased with the progression of liver diseases. Plasma ALT also could serve as a marker of disease severity.

(**F**) *Cpt1* mRNA levels consistently decreased in all *Clk*$^{\Delta 19/\Delta 19}$ mice compared with controls. All the other genes (*Mttp*, *Cd36*, *Dgat2*, *Fas*) showed a biphasic response. Expression of these genes increased in early stages of liver diseases (steatosis and hepatosteatosis) but decreased with advanced-disease stages (cirrhosis and carcinoma).

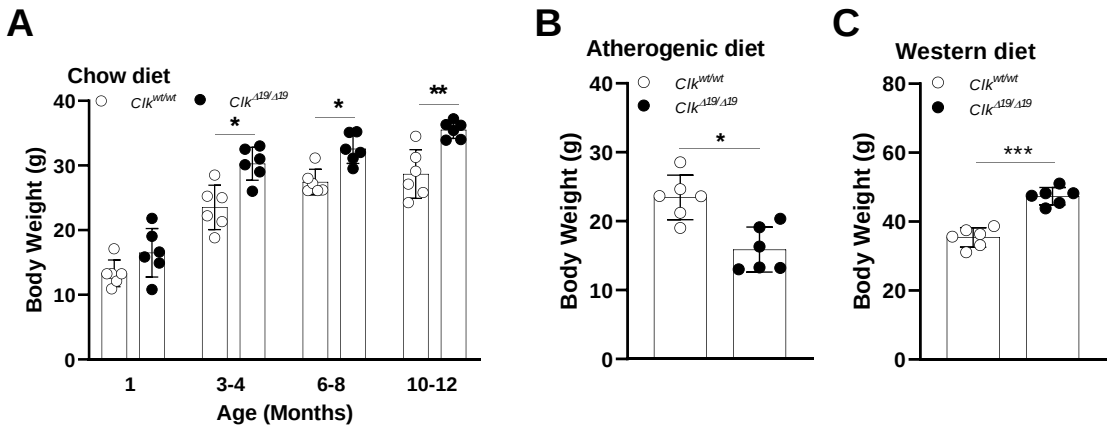
(**G**) Inflammatory response, measured as increases in select genes, was modest in *Clk*$^{\Delta 19/\Delta 19}$ mice compared with *Clk*$^{\Delta 19/\Delta 19}$*Apoe*$^{-/-}$ mice. In *Clk*$^{\Delta 19/\Delta 19}$*Apoe*$^{-/-}$ mice, TNFα, Tlr4, and Il-6 increased by greater than 10-fold compared with *Apoe*$^{-/-}$ mice after LPS and $CoCl_2$ injections.

(**H**) *Clk*$^{\Delta 19/\Delta 19}$ mice did not show significant increase in cancer genes. However, injection of LPS or $CoCl_2$ in *Clk*$^{\Delta 19/\Delta 19}$*Apoe*$^{-/-}$ mice significantly increased the expression of Mdm2, C-myc, and Bcl-2 but reduced Trp53. These changes were associated with advanced cirrhosis and carcinomas. Thus, *Clk*$^{\Delta 19/\Delta 19}$ mice in the absence of apoE are highly susceptible to inflammatory and carcinogenic response, and this response is associated with cirrhotic liver and hepatocellular carcinoma.

(**I**) All *Clk*$^{\Delta 19/\Delta 19}$ mice had reduced mRNA levels of all three PHD proteins. Further reductions in these mRNA levels were associated with the onset of steatosis, steatohepatitis, and cirrhosis. Thus, normal PHD proteins most likely protect against development of liver diseases.

Figure S12: Changes in the expression of ER stress response genes in different mouse models. The ER stress in *Clk*$^{\Delta 19/\Delta 19}$ mice is characterized by increases in *Ire1α* and its downstream target *Xbp-1*.

Figure S1


A
Chow diet
Clk^wt/wt
Clk^Δ19/Δ19
Body Weight (g)
0
10
20
30
40
*
*
**
1
3-4
6-8
10-12
Age (Months)
B
Atherogenic diet
Clk^wt/wt
Clk^Δ19/Δ19
Body Weight (g)
0
10
20
30
40
*
C
Western diet
Clk^wt/wt
Clk^Δ19/Δ19
Body Weight (g)
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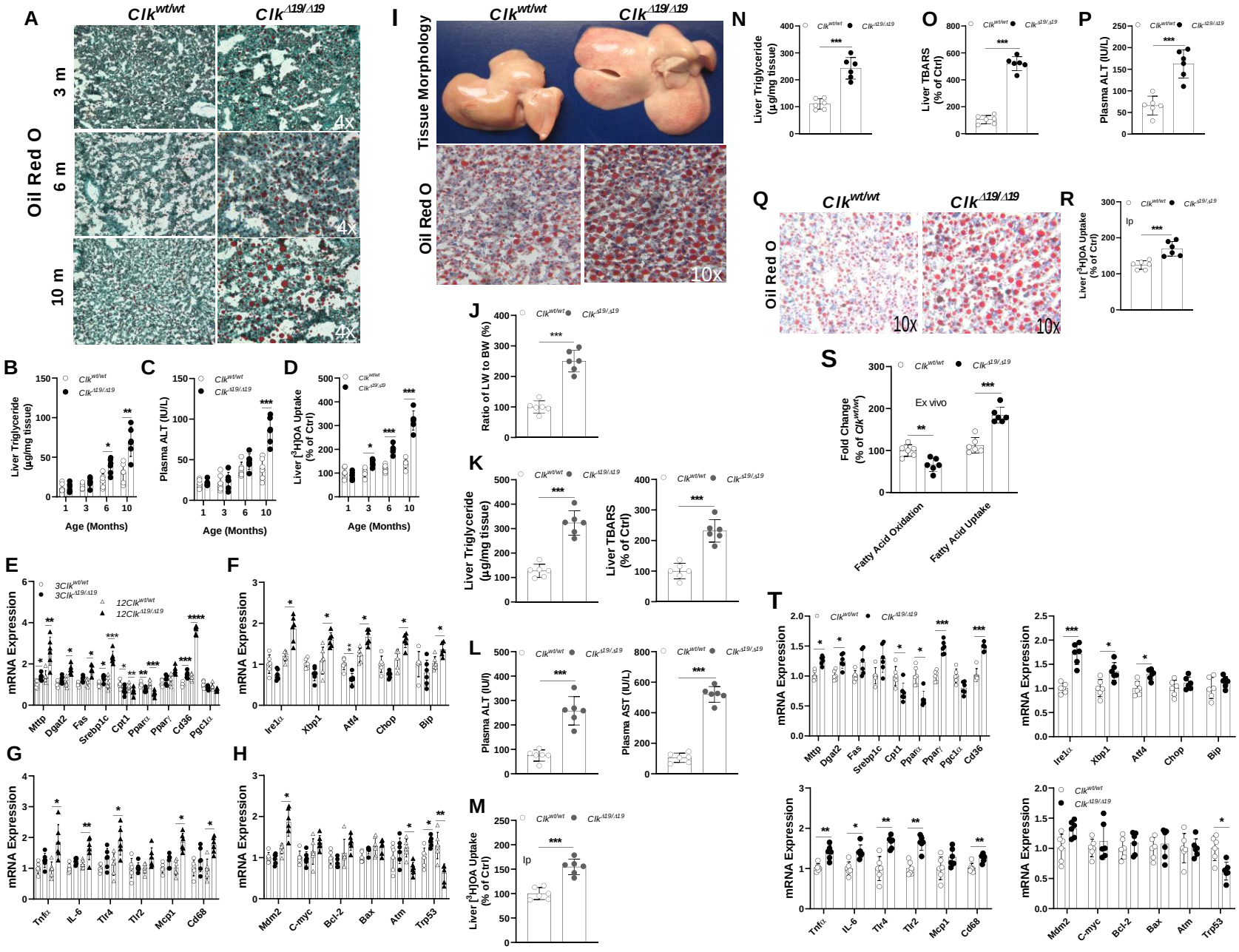
20
40
60
80
***

Figure S2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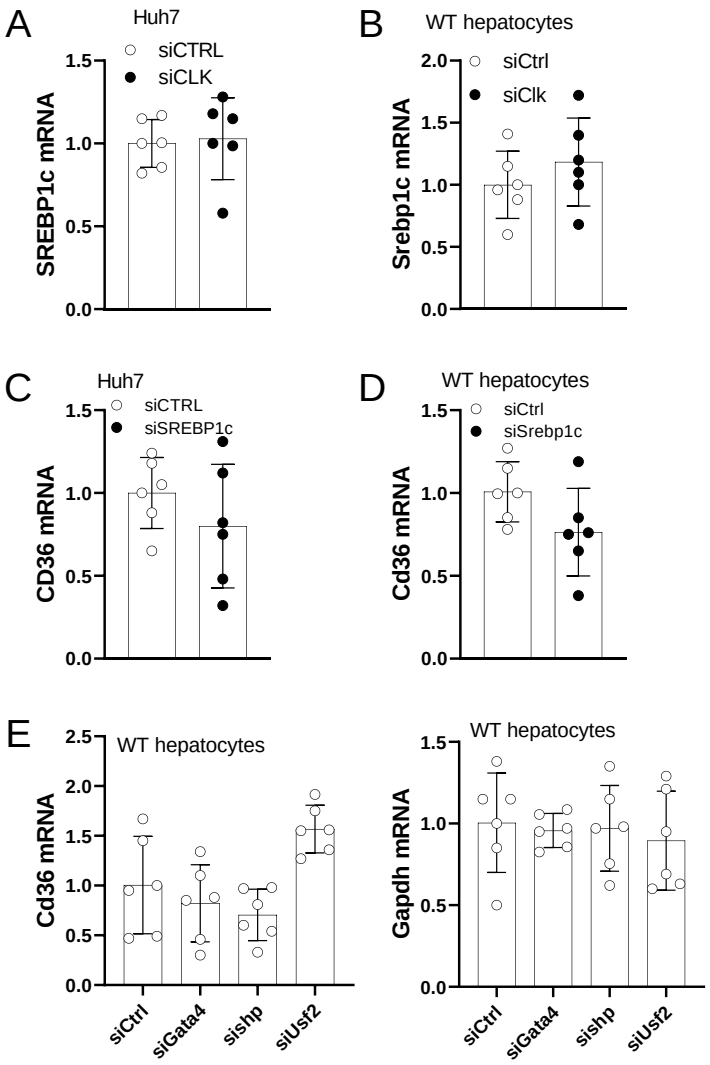
Clk^wt/wt
Clk^Δ19/Δ19
Oil Red O
3 m
6 m
10 m
4x
B
Liver Triglyceride (μg/mg tissue)
Age (Months)
C
Plasma ALT (IU/L)
D
Liver [3H]OA Uptake (% of Ctrl)
E
mRNA Expression
3Clk^wt/wt
3Clk^Δ19/Δ19
12Clk^wt/wt
12Clk^Δ19/Δ19
Mttp
Dgat2
Fas
Srebp1c
Cpt1
Pparα
Pparγ
Cd36
Pgc1α
F
Ire1α
Xbp1
Atf4
Chop
Bip
G
Tnfα
IL-6
Tlr4
Tlr2
Mcp1
Cd68
H
Mdm2
C-myc
Bcl-2
Bax
Atm
Trp53
I
Tissue Morphology
Oil Red O
10x
J
Ratio of LW to BW (%)
K
Liver Triglyceride (μg/mg tissue)
Liver TBARS (% of Ctrl)
L
Plasma ALT (IU/l)
Plasma AST (IU/L)
M
Ip
N
Liver Triglyceride (μg/mg tissue)
O
Liver TBARS (% of Ctrl)
P
Plasma ALT (IU/L)
Q
R
S
Ex vivo
Fold Change (% of Clk^wt/wt)
Fatty Acid Oxidation
Fatty Acid Uptake
T

Figure S3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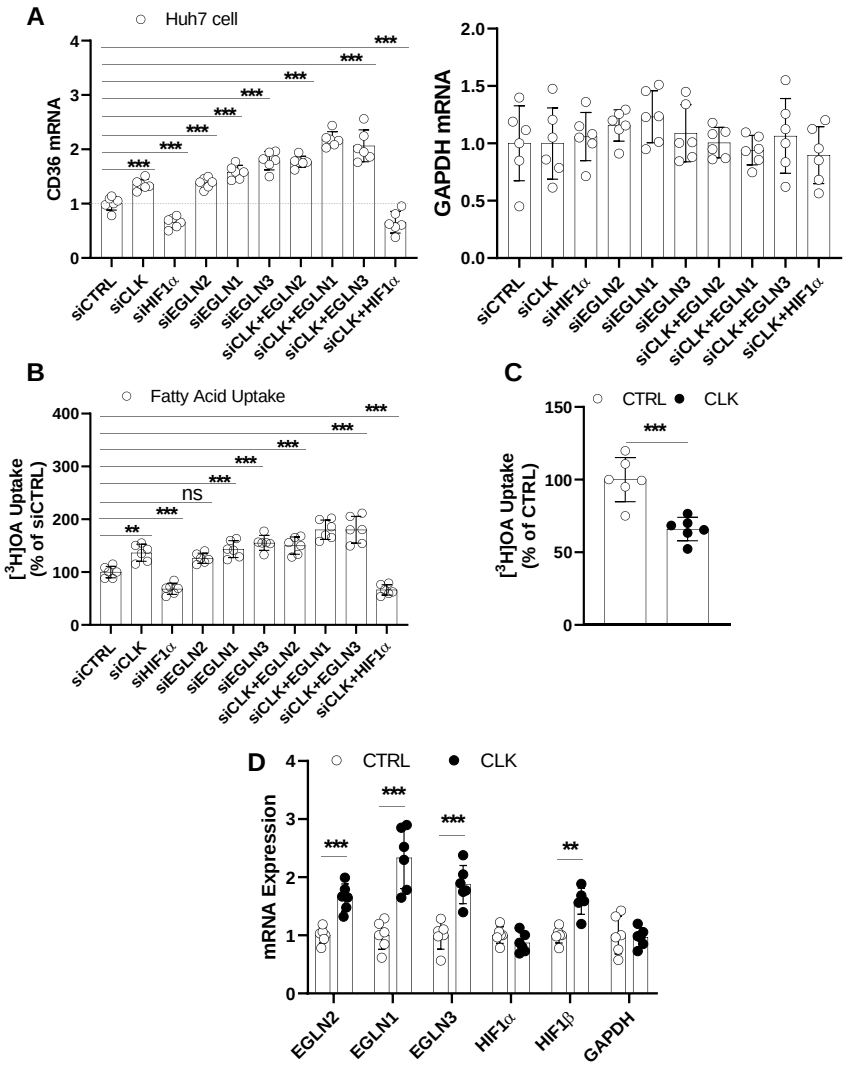
Huh7
siCTRL
siCLK
SREBP1c mRNA
1.5
1.0
0.5
0.0
B
WT hepatocytes
siCtrl
siClk
Srebp1c mRNA
2.0
1.5
1.0
0.5
0.0
C
Huh7
siCTRL
siSREBP1c
CD36 mRNA
1.5
1.0
0.5
0.0
D
WT hepatocytes
siCtrl
siSrebp1c
Cd36 mRNA
1.5
1.0
0.5
0.0
E
WT hepatocytes
Cd36 mRNA
2.5
2.0
1.5
1.0
0.5
0.0
siCtrl
siGata4
sishp
siUsf2
WT hepatocytes
Gapdh mRNA
1.5
1.0
0.5
0.0
siCtrl
siGata4
sishp
siUsf2

Figure S4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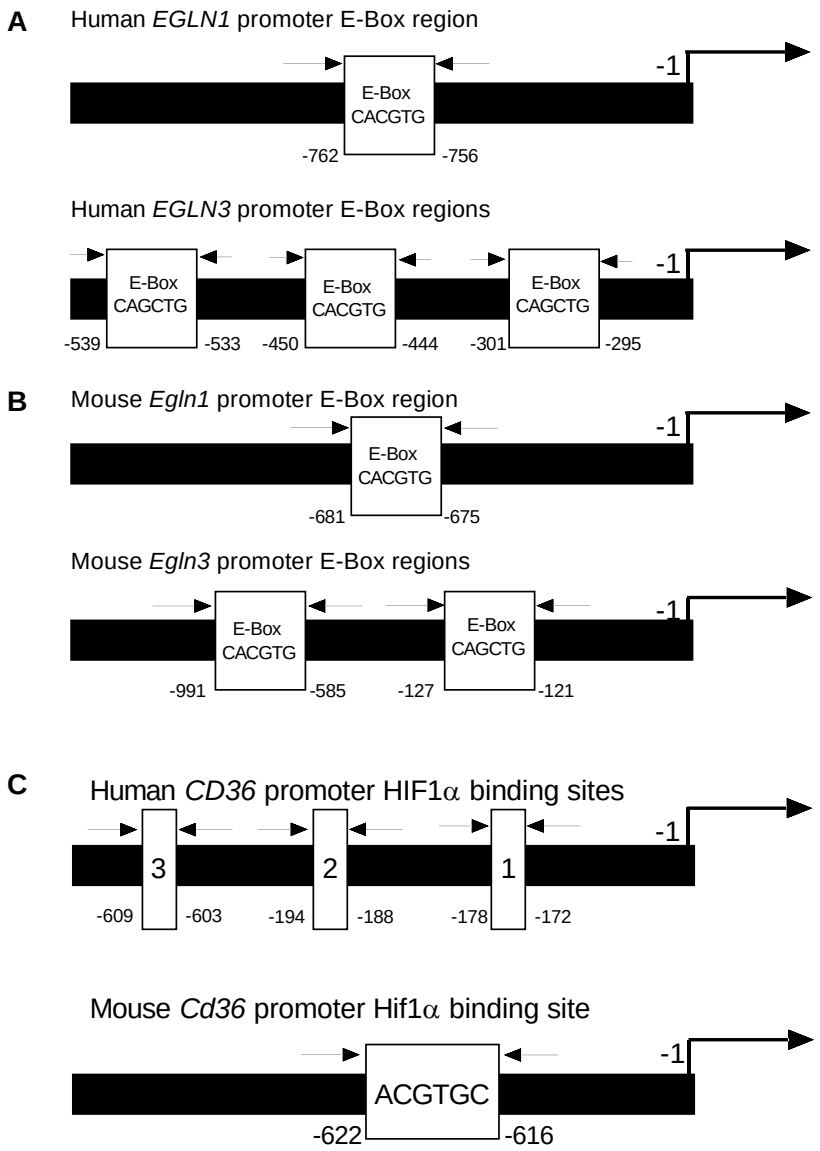
Huh7 cell
CD36 mRNA
GAPDH mRNA
siCTRL
siCLK
siHIF1α
siEGLN2
siEGLN1
siEGLN3
siCLK+EGLN2
siCLK+EGLN1
siCLK+EGLN3
siCLK+HIF1α
B
Fatty Acid Uptake
[3H]OA Uptake (% of siCTRL)
ns
C
CTRL
CLK
[3H]OA Uptake (% of CTRL)
D
mRNA Expression
EGLN2
EGLN1
EGLN3
HIF1α
HIF1β
GAPDH

Figure S5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Human EGLN1 promoter E-Box region
E-Box
CACGTG
-1
-762
-756
Human EGLN3 promoter E-Box regions
E-Box
CAGCTG
E-Box
CACGTG
E-Box
CAGCTG
-1
-539
-533
-450
-444
-301
-295
B
Mouse Egln1 promoter E-Box region
E-Box
CACGTG
-1
-681
-675
Mouse Egln3 promoter E-Box regions
E-Box
CACGTG
E-Box
CAGCTG
-1
-991
-585
-127
-121
C
Human CD36 promoter HIF1α binding sites
3
2
1
-1
-609
-603
-194
-188
-178
-172
Mouse Cd36 promoter Hif1α binding site
ACGTGC
-1
-622
-616

Figure S6

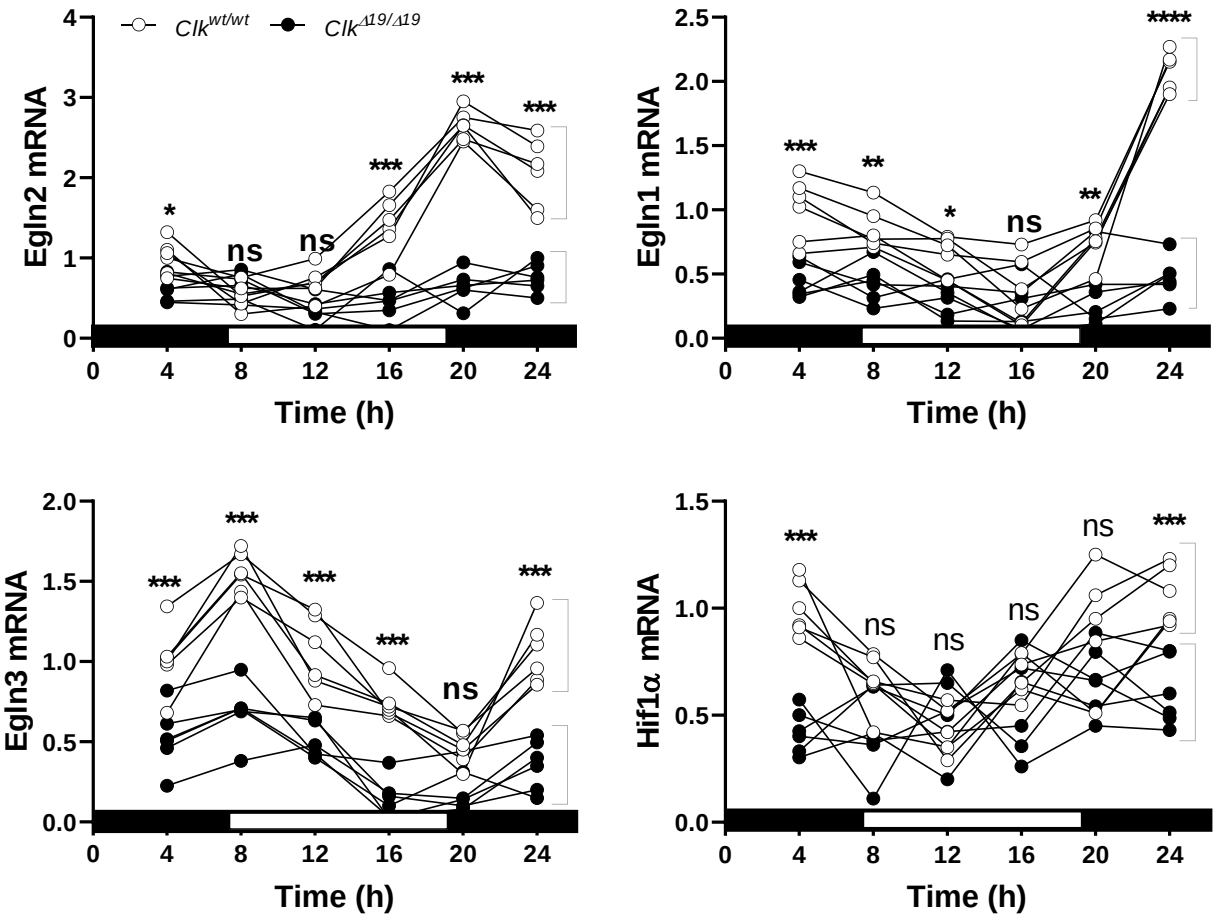

Clk^wt/wt
Clk^Δ19/Δ19
Egln2 mRNA
Egln1 mRNA
Egln3 mRNA
Hif1α mRNA
Time (h)
ns
*
**
***
****

Figure S7

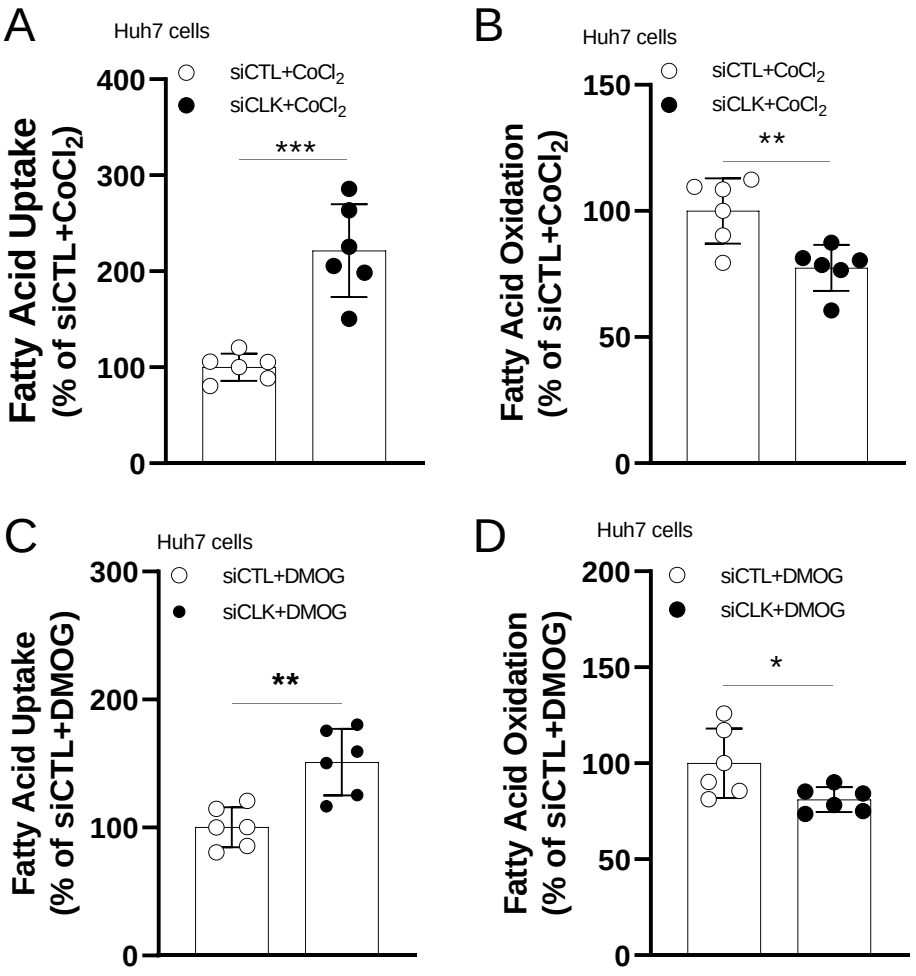
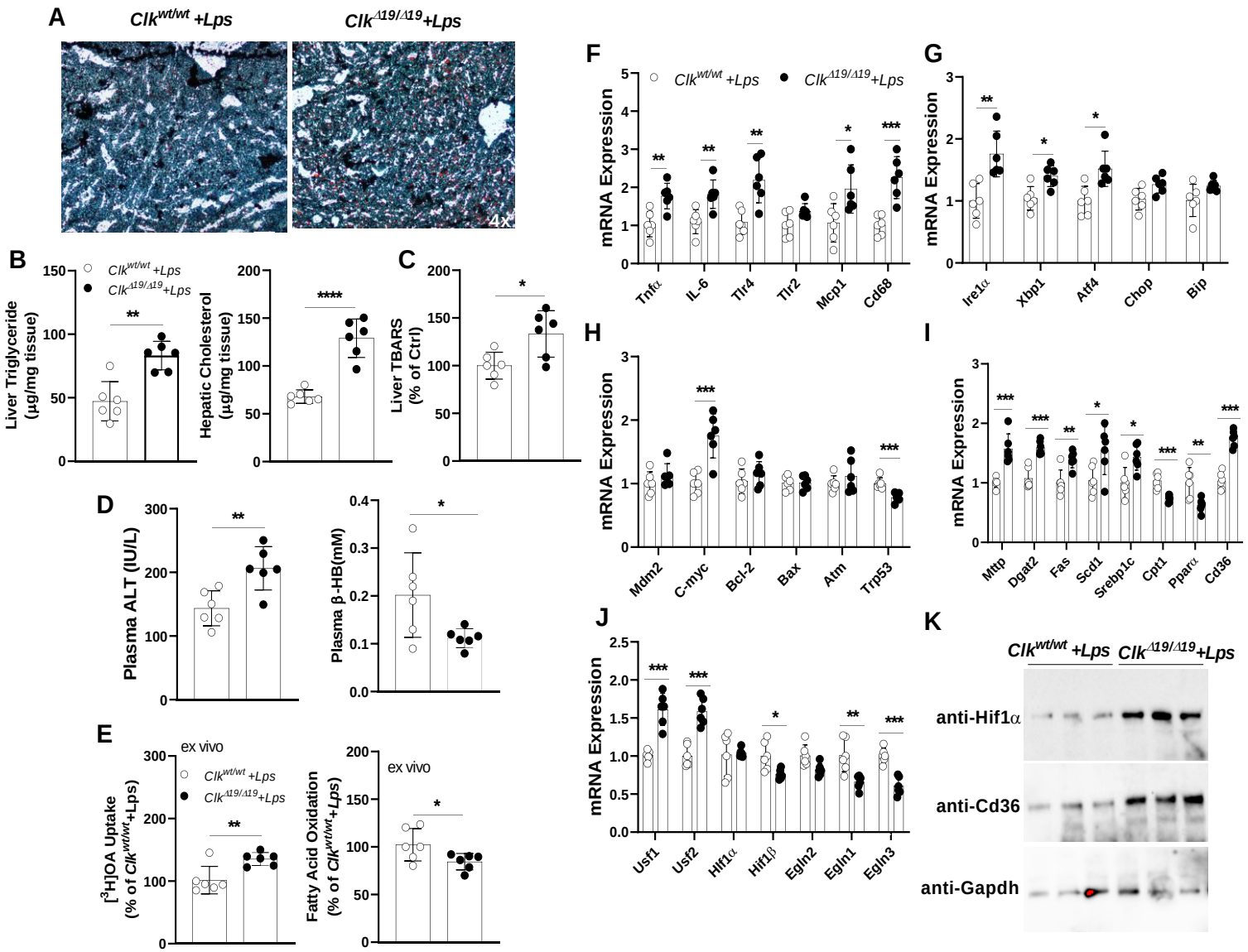

A
Huh7 cells
siCTL+CoCl2
siCLK+CoCl2
***
Fatty Acid Uptake
(% of siCTL+CoCl2)
400
300
200
100
0
B
Huh7 cells
siCTL+CoCl2
siCLK+CoCl2
**
Fatty Acid Oxidation
(% of siCTL+CoCl2)
150
100
50
0
C
Huh7 cells
siCTL+DMOG
siCLK+DMOG
**
Fatty Acid Uptake
(% of siCTL+DMOG)
300
200
100
0
D
Huh7 cells
siCTL+DMOG
siCLK+DMOG
*
Fatty Acid Oxidation
(% of siCTL+DMOG)
200
150
100
50
0

Figure S8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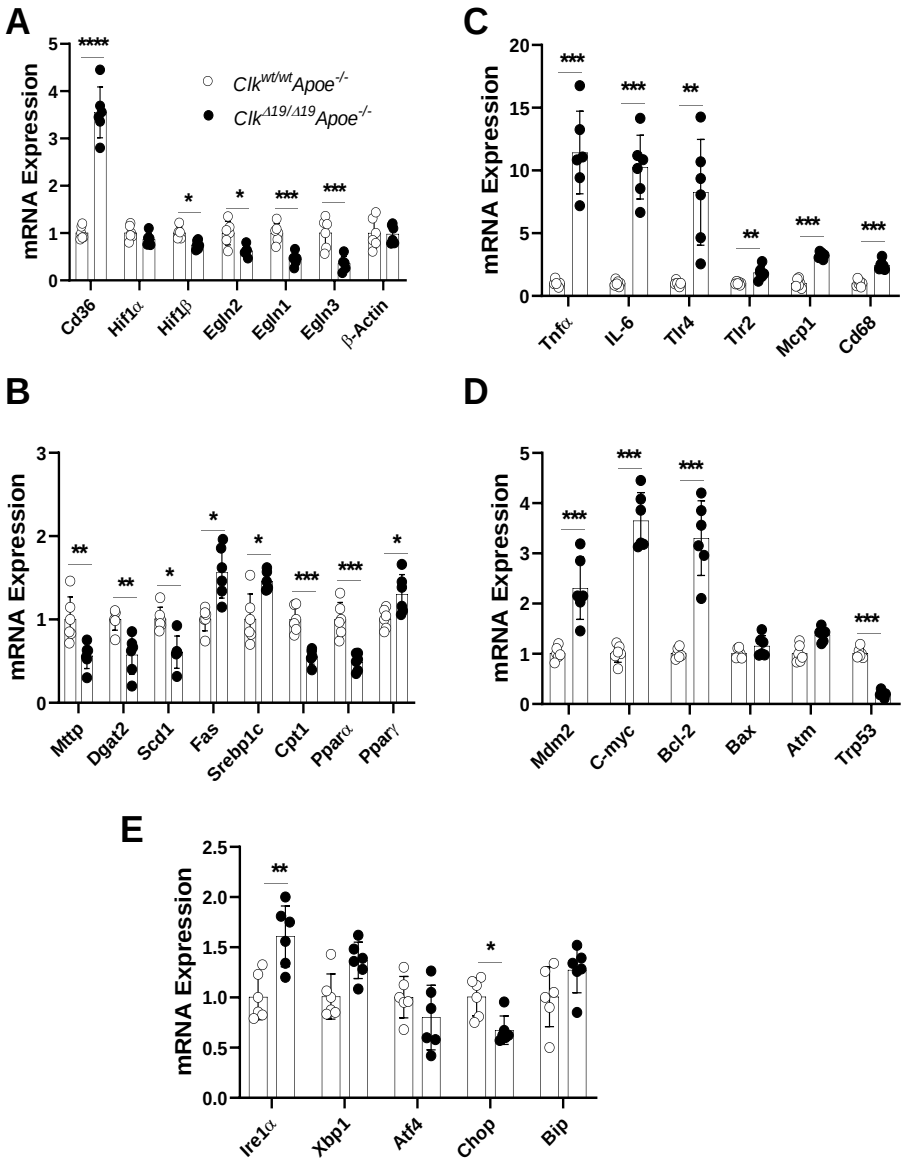
Clk^wt/wt +Lps
Clk^Δ19/Δ19+Lps
4x
B
Clk^wt/wt +Lps
Clk^Δ19/Δ19+Lps
Liver Triglyceride (μg/mg tissue)
Hepatic Cholesterol (μg/mg tissue)
C
Liver TBARS (% of Ctrl)
D
Plasma ALT (IU/L)
Plasma β-HB(mM)
E
ex vivo
[3H]OA Uptake (% of Clk^wt/wt+Lps)
Fatty Acid Oxidation (% of Clk^wt/wt+Lps)
F
mRNA Expression
Tnfα
IL-6
Tlr4
Tlr2
Mcp1
Cd68
G
Ire1α
Xbp1
Atf4
Chop
Bip
H
Mdm2
C-myc
Bcl-2
Bax
Atm
Trp53
I
Mttp
Dgat2
Fas
Scd1
Srebp1c
Cpt1
Pparα
Cd36
J
Usf1
Usf2
Hif1α
Hif1β
Egln2
Egln1
Egln3
K
anti-Hif1α
anti-Cd36
anti-Gapdh

Figure S9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mRNA Expression
Clk^wt/wt Apoe^-/-
Clk^Δ19/Δ19 Apoe^-/-
Cd36
Hif1α
Hif1β
Egln2
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Egln3
β-Actin
B
mRNA Expression
Mttp
Dgat2
Scd1
Fas
Srebp1c
Cpt1
Pparα
Pparγ
C
mRNA Expression
Tnfα
IL-6
Tlr4
Tlr2
Mcp1
Cd68
D
mRNA Expression
Mdm2
C-myc
Bcl-2
Bax
Atm
Trp53
E
mRNA Expression
Ire1α
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Chop
Bip

Figure S10

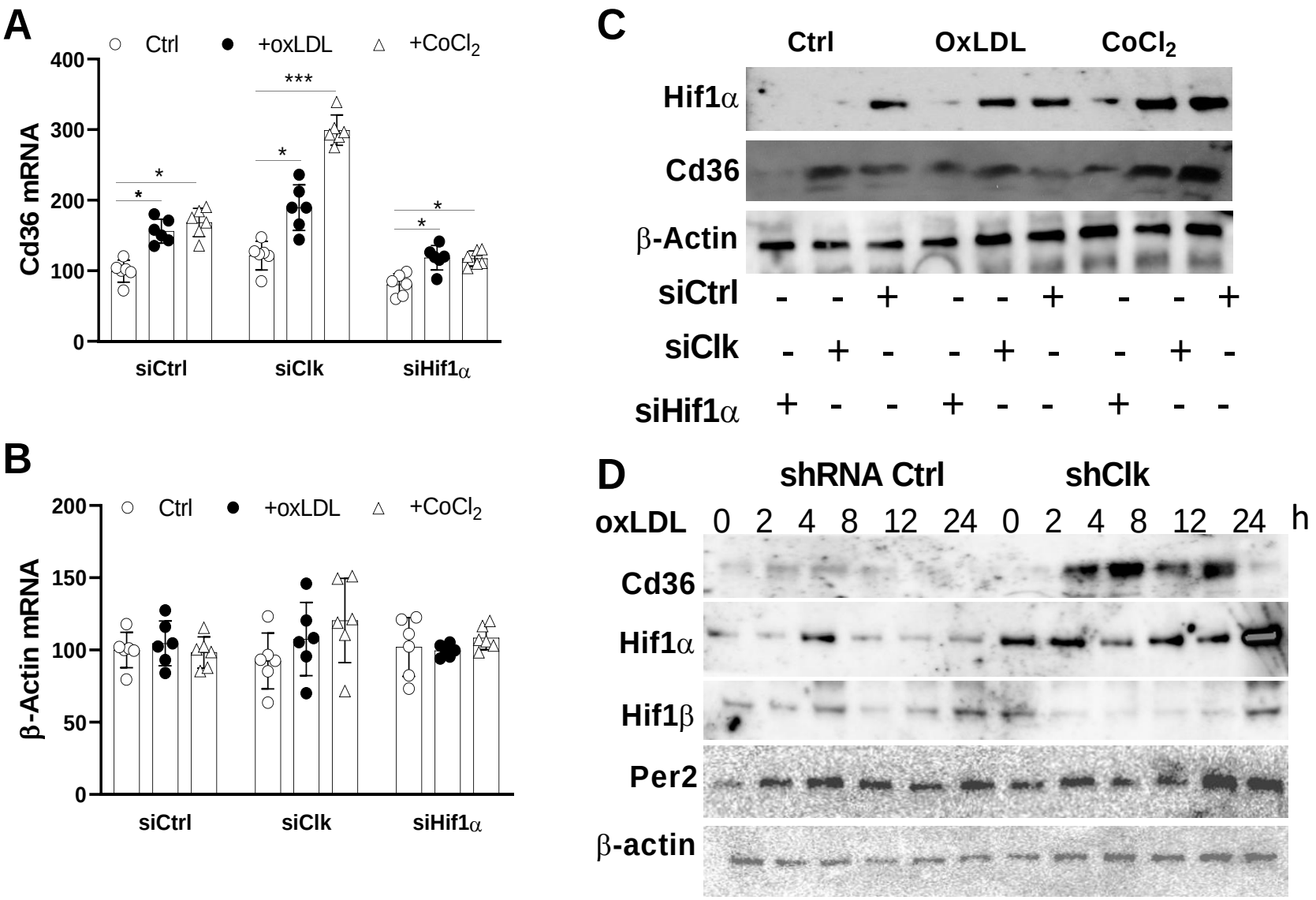
A
Ctrl
+oxLDL
+CoCl2
Cd36 mRNA
400
300
200
100
0
***
*
*
*
*
siCtrl
siClk
siHif1α
B
Ctrl
+oxLDL
+CoCl2
β-Actin mRNA
200
150
100
50
0
siCtrl
siClk
siHif1α
C
Ctrl
OxLDL
CoCl2
Hif1α
Cd36
β-Actin
siCtrl - - + - - + - - +
siClk - + - - + - - + -
siHif1α + - - + - - + - -
D
shRNA Ctrl
shClk
oxLDL 0 2 4 8 12 24 0 2 4 8 12 24 h
Cd36
Hif1α
Hif1β
Per2
β-actin

Figure S11

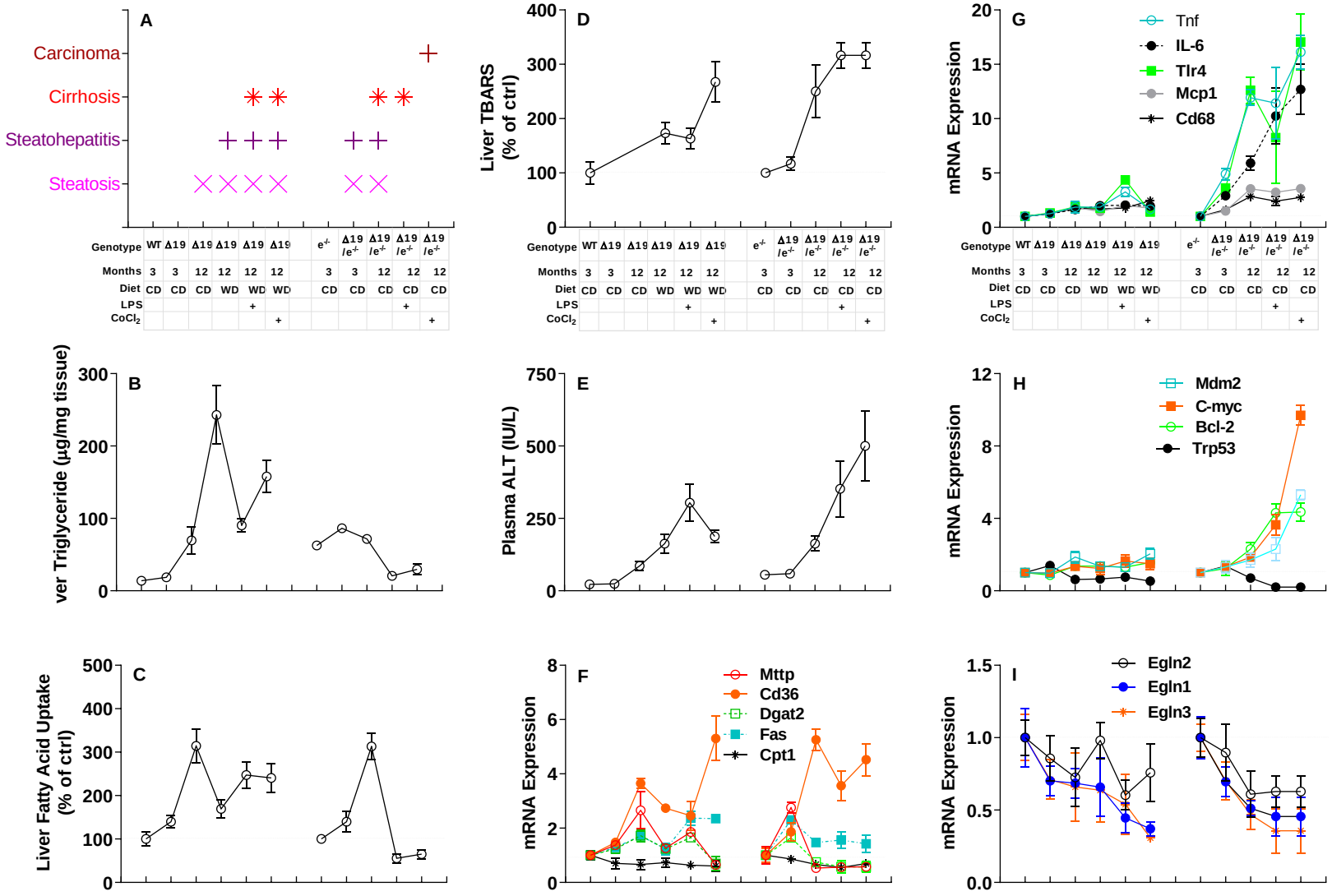
A
Carcinoma
Cirrhosis
Steatohepatitis
Steatosis
Genotype WT Δ19 Δ19 Δ19 Δ19 Δ19 e-/- Δ19/e-/- Δ19/e-/- Δ19/e-/- Δ19/e-/-
Months 3 3 12 12 12 12 3 3 12 12 12
Diet CD CD CD WD WD WD CD CD CD CD CD
LPS + +
CoCl2 + +
B
ver Triglyceride (µg/mg tissue)
C
Liver Fatty Acid Uptake (% of ctrl)
D
Liver TBARS (% of ctrl)
E
Plasma ALT (IU/L)
F
mRNA Expression
Mttp
Cd36
Dgat2
Fas
Cpt1
G
mRNA Expression
Tnf
IL-6
Tlr4
Mcp1
Cd68
H
mRNA Expression
Mdm2
C-myc
Bcl-2
Trp53
I
mRNA Expression
Egln2
Egln1
Egln3

Figure S12

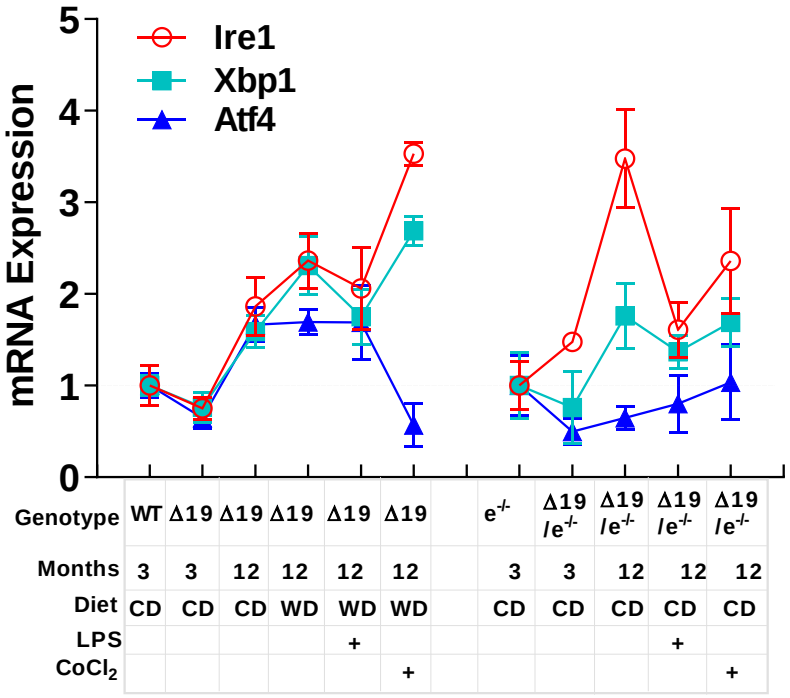
mRNA Expression
0
1
2
3
4
5
Ire1
Xbp1
Atf4
Genotype WT Δ19 Δ19 Δ19 Δ19 Δ19 e-/- Δ19/e-/- Δ19/e-/- Δ19/e-/- Δ19/e-/-
Months 3 3 12 12 12 12 3 3 12 12 12
Diet CD CD CD WD WD WD CD CD CD CD CD
LPS + +
CoCl2 + +

Table S1: Chemical, antibodies and siRNA used in the study

Name	**Cat#**	**Company**
$CoCl_2$	232696-100G	Sigma-Aldrich
LPS	L4130	Sigma-Aldrich
Collagenase type IV	LS004188	Worthington Biochemical Corporation
Poloxamer P407	16758-250G	Sigma-Aldrich
TBS-Tween 20	IBB-181	Boston Bioproducts
Alexa Fluor 488–AcLDL	L-23380	Invitrogen
[9,10-^{3}H(N)] Oleic Acid	NET289001MC	Perkin Elmer
[^{14}C] Oleic Acid	PEC317150UC	Perkin Elmer
[^{14}C] Palmitic Acid	NEC075H250UC	Perkin Elmer
Ad-m- Hif1α- shRNA	shADV-261289	VectorBiolabs
ROS detection assay kit	ab186029	Abcam
ChIP assay kit	USB 78460	Affymetrix
TBARS Assay Kit	10009055	Cayman Chemical Company
In situ cell death detection kit	12-156-792-910	Sigma-Aldrich
Triglyceride	TR22421	Fisher Scientific
Cholesterol	TR13421	Fisher Scientific
Free Fatty acids	999034691	Wako Chemicals
ALT	MAK052-1KT	Sigma-Aldrich
AST	MAK055-1KT	Sigma-Aldrich
DMOG	D3695-50MG	Sigma-Aldrich
Antibodies		
Macrophage antibody (IHC use)	ab56297	Abcam
Hif1 alpha antibody (IHC use)	EPR 16897	Abcam
Hif1 alpha antibody (WB use)	sc-35562PR	Santa Cruz Biotechnology
Hif1a antibody (CHIP)	NB100-479	Novus Biological
PHD1 antibody (WB)	Abcam113077	Abcam
Usf1 antibody (WB)	sc-36784PR	Santa Cruz Biotechnology
Usf2 antibody (WB)	sc-36785PR	Santa Cruz Biotechnology
PHD2 antibody (CHIP)	NB100-2219	Novus Biological
PHD2 antibody (WB)	4835	Cell signaling Technology
PHD3 antibody (WB, CHIP)	NB100-139	Novus Biological
Cd36 antibody (WB)	AB64014	Abcam
Clock antibody (WB)	NBP1-88615	Novus Biologicals
Clock antibody (CHIP)	ab3517	Abcam
Bmal1 antibody (CHIP)	ab93806	Abcam
Hif1 beta antibody (WB)	NB100-110	Novus Biological
Per2 antibody (WB)	NB100-125	Novus Biological
β-Actin antibody (WB)	Ab8227	Abcam
Cd36 antibody (IHC use)	AB133625	Abcam
siRNA-Mouse		
siHif1	sc-35562	Santa Cruz Biotechnology
siSrebp1c	sc-36558	Santa Cruz Biotechnology

siClock	sc-35075	Santa Cruz Biotechnology
siShp	sc-44870	Santa Cruz Biotechnology
siUsf2	sc-36785	Santa Cruz Biotechnology
siGata4	Smart pool L-040759	Dharmacon, Inc.
siPhd1	Smart pool L-051750	Dharmacon, Inc.
siPhd2	Smart pool L-040757	Dharmacon, Inc.
siPhd3	Smart pool L-040261	Dharmacon, Inc.
shCLK	Sc-401261-LAC	Santa Cruz Biotechnology
siRNA-Human		
siSREBP1	sc-36557	Santa Cruz Biotechnology
siCLOCK	sc-35074	Santa Cruz Biotechnology
siHIF1α	sc-35561	Santa Cruz Biotechnology
siPHD1	J-004277-05-0002	Dharmacon, Inc.
siPHD2	J-004276-07-0002	Dharmacon, Inc.
siPHD3	J-004274-09-0002	Dharmacon, Inc.

Table S2: Chemical composition (% weight) of different diets used in the study:

% by weight	**Chow**	**Western**	**Atherogenic**
Protein	23.9%	17.3%	20.5%
Carbohydrates	48.7%	48.5%	42.4%
Fat	5.0%	21.2%	37.1%
Cholesterol	0.02%	0.20%	1.25%
Cholic acid	-	-	0.5%
Source	LabDiet	Harland Tekland	Harland Tekland
Catalog #	5001	TD88137	TD88051

Table S3: Oligonucleotide sequences of primers used

RT-PCR primers	Forward (5′-3′)	Reverse (5′-3′)
mHifα	ctgcgtgcatgtctaatctgttc	gagcgcggaaaactcttgtt
mHif1β	cccaccattgcttctggaaa	cgtcgcttaatagccctctgtac
mUsf1	ggcttactcaagaggtgggaa	tgattgggccccttctact
mUsf2	atggaaccagaactcctcgagat	ccttctccgttcgacttcattg
mCd36	Tacctgggagttggcgagaa	gttccgatcacagcccattc
mPhd1	cgtgaggcatgttgacaatc	cgtcggtcagaccagaaaat
mPhd2	attttgccagacctgtcacc	gcaacacgtcgtcactcact
mPhd3	tgactgcaactggctggtag	gctgcttgtgggattctagc
mHif2α	ccagcactgcttcagtacca	caagtgtgaactgctggtgc
mFih-1	cccagacactggtggaagtt	aacctcagtgttccccacag

mArd-1	tggctcctcttgtctcctgt	cttgctgtccccaccactat
mVegf	caggctgctgtaacgatgaa	tttgacccttccctttcct
mFas	ggaaggctgggctctatgg	ggcgtcgaacttggagagatc
mDgat1	gttcaggtcagacagtggtttca	Tcagcatcaccacacacaccaa
mDgat2	agctgcaggtcatctcagtactaca	ctgcaggccactcctagca
mCpt1	caaacccaccaggctacagt	tccttgtaatgtgcgagctg
mPparα	tccgagggctctgtcatca	tgactgaggaagggctggaa
Mgat2	tgggatttgaacctgtagcc	tccctgtttgtcctttggtc
mPgc1α	Taggcccaggtacgacagc	gctctttgcggtattcatcc
mPgc1β	Caagctctgacgctctgaagg	Ttggggagcaggctttcac
mSrebp1c	ggagccatggattgcacatt	ggcccgggaagtcactgt
mBmal1	caaccttcccgcagctaaca	tccgcgatcattcgacctat
mClock	caccgacaaagatccctactgat	tgagacatcgctggctgtgt
mGapdh	tgtgtccgtcgtggatctga	cctgcttcaccaccttcttga
mMtp	cacacaactggcctctcattaaat	tgcccccatcaagaaacact
mL-Fabp	gtcagctgtggaaaggaagc	gatttctgacacccccttga
mScd1	ctacaagcctggcctcctgc	ggaccccagggaaaccagga
mTnfα	agcccccagtctgtatcctt	ctccctttgcagaactcagg
mIl-6	gagcccaccaagaacgatag	tccacgatttcccagagaac
mTlr4	acctggctggtttacacgtc	ctgccagagacattgcagaa
mTlr2	ctcccacttcaggctctttg	gaagtcaggaactgggtgga
mMcp1	cctgttccagttacttccctcagt	caccaacccaagcctctgttt
mCd68	cttctgctgtggaaatgcaa	agaggggctggtaggttgat
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mBip	agcctgttgctggactccta	ggaataggtggtccccaagt
mMdm2	cttcgtgagaactggcttcc	cagcacatggctctttagca
mC-myc	gcccagtgaggatatctgga	atcgcagatgaagctctggt
mBcl-2	agaaggagcaggtgcctaca	gcattttcccaccactgtct
mBax	tgcagaggatgattgctgac	gatcagctcgggcactttag
mAtm	ttacgatggcaacagcagag	tccagttctcgctgaacctt

mTrp53	gagagaccgccgtacagaag	ctgtagcatgggcatccttt
mArppo	gtccaactacttcctcaagatcatcca	acatgcggatctgctgcat
m18sRNA	agtcccttgccctttgtacaca	gatccgagggcctcactaaac
hCD36	agatgcagcctcatttccac	tgggttttcaactggagagg
hPHD1	agcccctaagtcaggctctc	agtggtagaggtggctgtgg
hPHD2	ggactggaagaagcacaagc	ggcctttacttttcccttgg
hPHD3	cgtaggaacccacacgaagt	cagatttcagagcacggtca
hHIF1α	ccacagctgaccagttatgattg	gagtaattcttcaccctggagtaggt
hHIF1β	ctgtcagatatggtacccacctgta	gccatgcgtaagatggttagct
hGAPDH	cgaccactttgtcaagctca	aggggagattcagtgtggtg
Chip assay primers	Forward (5′-3′)	Reverse (5′-3′)
mCD36	cagataacagggtggcctgg	gggctaaaaggcagaagatgg
mPHD2	ggtgtgcttgacctgagtga	aagccggaagagtgacagtg
mPHD3	gagacaccataaccgtggca	ctgaggctgagcaccagatg
hCD36	cccttctcgtgggtaatcttt	tgcccatgtgcaatctcttt
hPHD2	tgggggacatagtagggagg	gcttgtctctgtactggggg
hPHD3	ccctgggacacatcatgagg	gggaagcagcgagtagagac
Genotyping primers	Forward (5′-3′)	Reverse (5′-3′)
mClock$^{\Delta 19/\Delta 19}$	agcaccttcctttgcagttcg	tgtgctcagacagaataagta
mClock$^{wt/wt}$	ggtcaagggctacaggta	Tggggtaaaaagacctcttgcc
mApoe$^{-/-}$	actactacacaggatgcctagcc	agggtgaaagagctggacactc
mApoe$^{wt/wt}$	tgtgacttgggagctctgcagc	gcctagccgagggagagccg