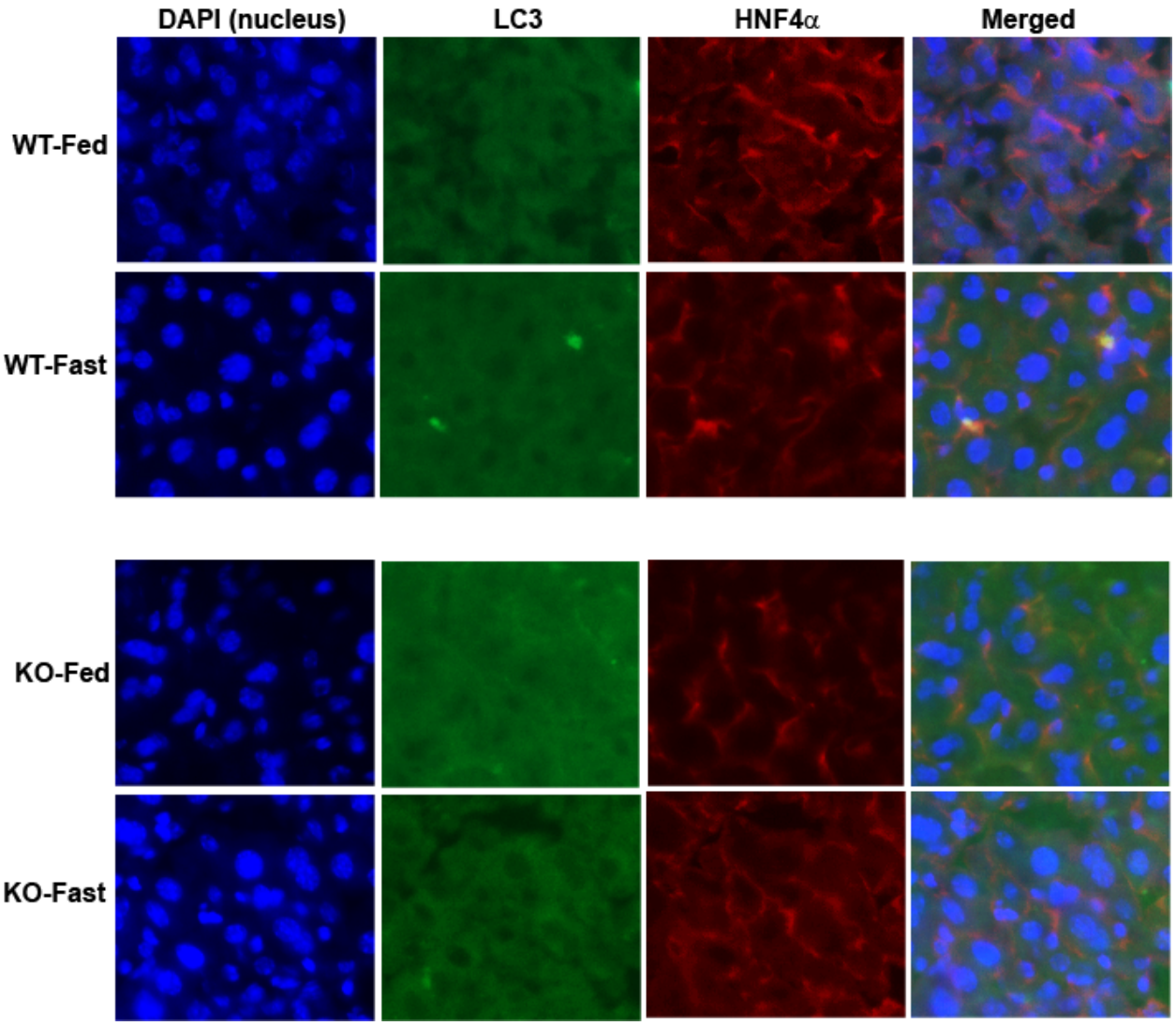
S-Table 1. Primer information for qPCR used in this study

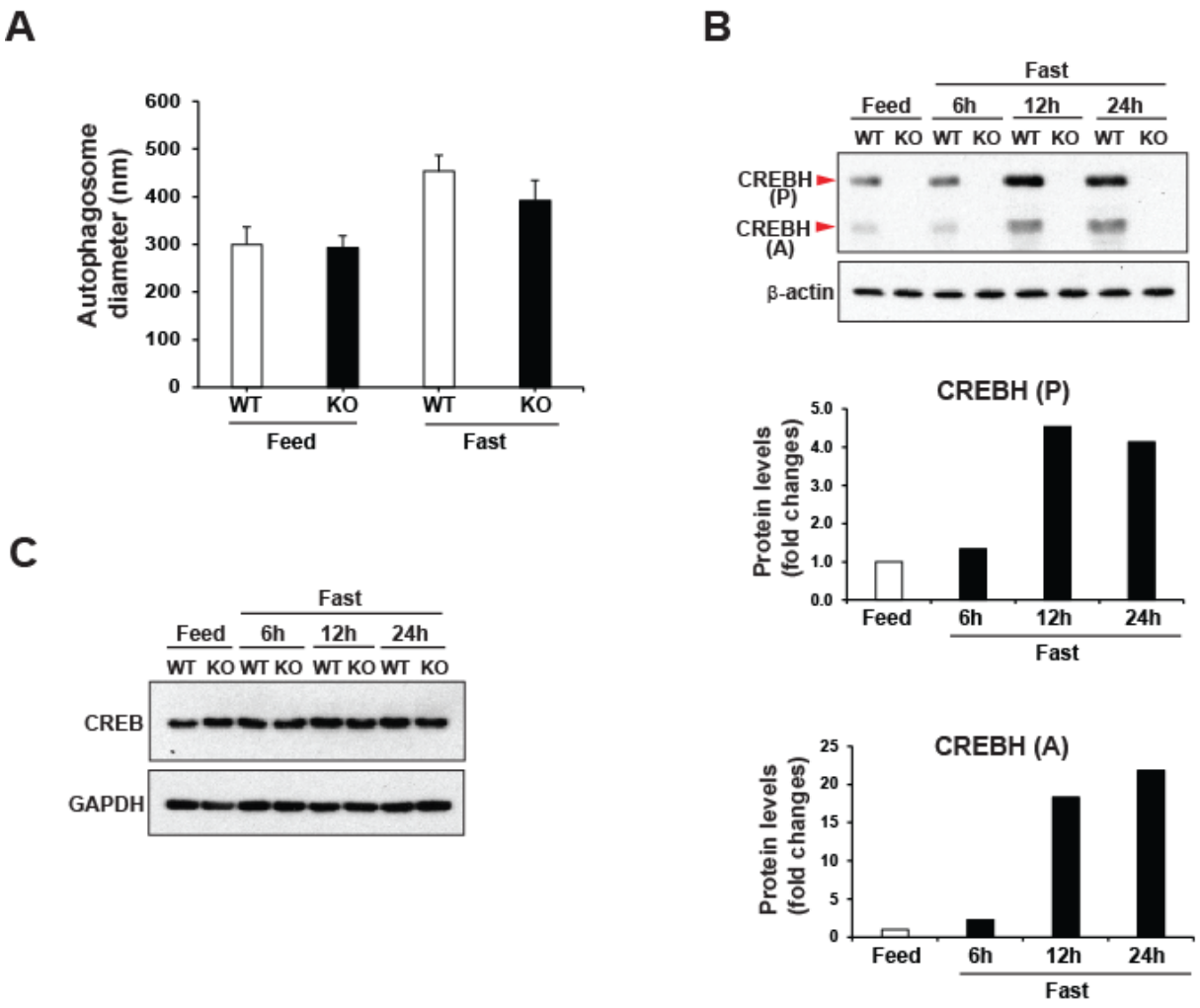
Gene name	Forward	Reverse
Atg7	CAGAAGAAGTTGAACGAGTA	CAGAGTCACCATTGTAGTAAT
Atg2b	CCTCCACTCTCAGAATCA	GTCCATCACACGAACATAA
Ulk1	ACCATTGTCTACCAGTGT	AGTGTCTTGTTCTTCTCATAA
LC3b	GCTCTTTGTTGGTGTGTA	TCTTCTGTTGCTGTTGTC
TFEB	CAGAAGCGAGAGCTAACAGAT	TGTGATTGTCTTTCTTCTGCCG
CTSA	GCTACCTCAGAGCATCGGAC	GTTAAGCCAAAGCACCACGG
GBA	GCCTCCCAGAAGAAGACACC	ATATCCCCTGGCTGACCCTT
GLA	TTGGGGTCAGAGCATTGGAC	AGTCATAACCTGCATCCCGC
PSAP	CAGCAGATGGTCTGGAGCAA	CAAGTTCCCAGCTTCGGTGA
CLCN7	CGAGATGCCTATCCACGCTT	CCCGGAAGAGCTTGAACACT
MCOLN1	TGGGCCAATGGATCAGCTTT	GTTCTTGTAACTGGCGCTGC
ATP6V1H	AAGACCAGCAGGTTCGCTAC	TGCAGAAAGGCTGAACAGGT
β-actin	GATCTGGCACCACACCTTCT	GGGGTGTTGAAGGTCTCAAA

S-Table 2. Primer information for ChIP assay and EMSA used in this study

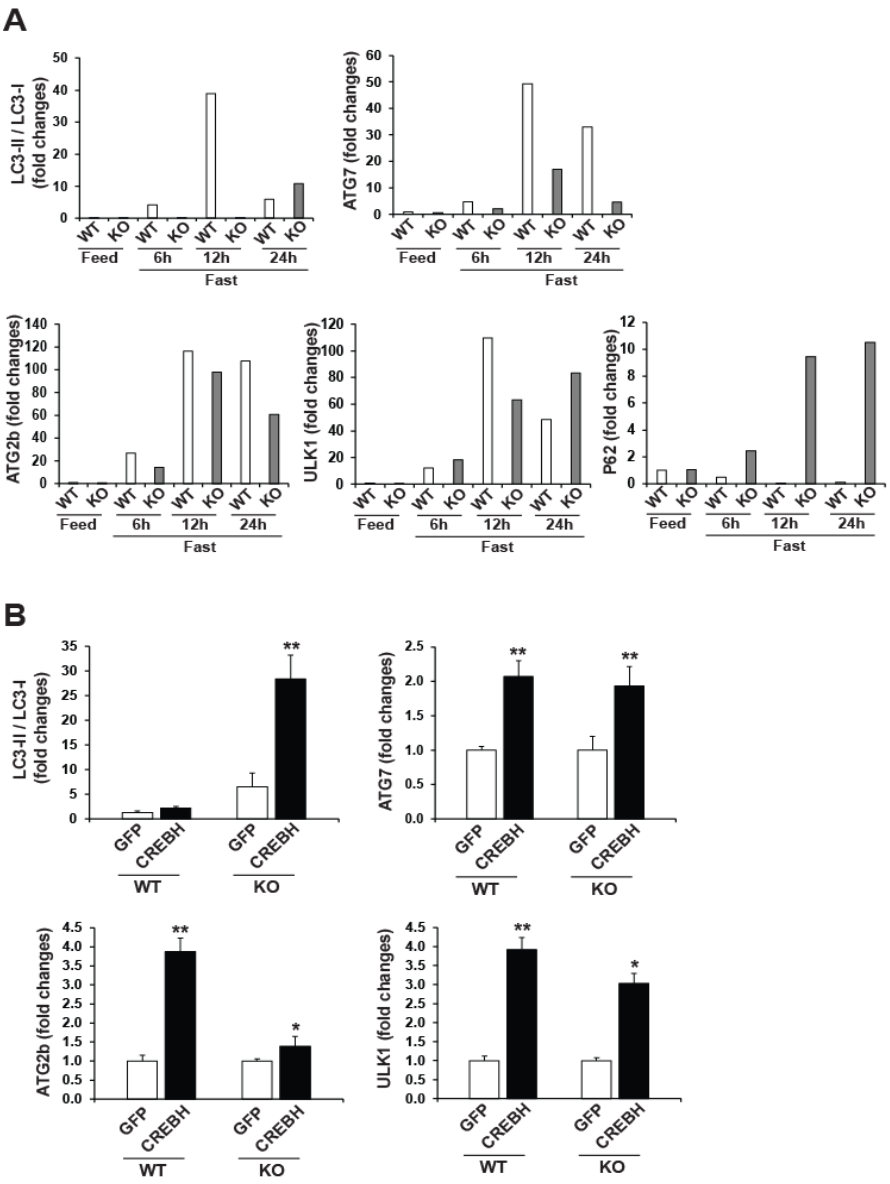
Chromatin Immunoprecipitation assay		
Gene name	**Forward**	**Reverse**
Atg7	ACTGGGGCAAAGCAAAGGTA	CCATGGCTGTTCTGGACCTG
Atg2b-Low	AGTCCGGGCGTCCGCCGGCG	GCGAGACCCAGACTCCCGGC
Ulk1	CCAATTCCAGGACCCGACAC	GCAGACTGGGCATGTCAATG
LC3b	AGTTAACAGATGCTCGCCCA	TGTGTGTGTCTCAGTCCGC
TFEB-PPRE	TATCCTAGTGCCGGGAGCTT	TTTGGTTTCATCCGGCAGGT
TFEB-CRE	ATCTAGTTGTAGAACAGCCA	ATCCCACCCCCATAATTGCC
Electrophoretic mobility shift assay		
LC3b-Foward	Biotin-ACCCTCGGTGACGCGCCGCGGTCACCTGACTCGGCTGCG	
LC3b-Reverse	Biotin-CGCAGCCGAGTCAGGTGACTCGCGGCGCGTCACCGAGGGT	



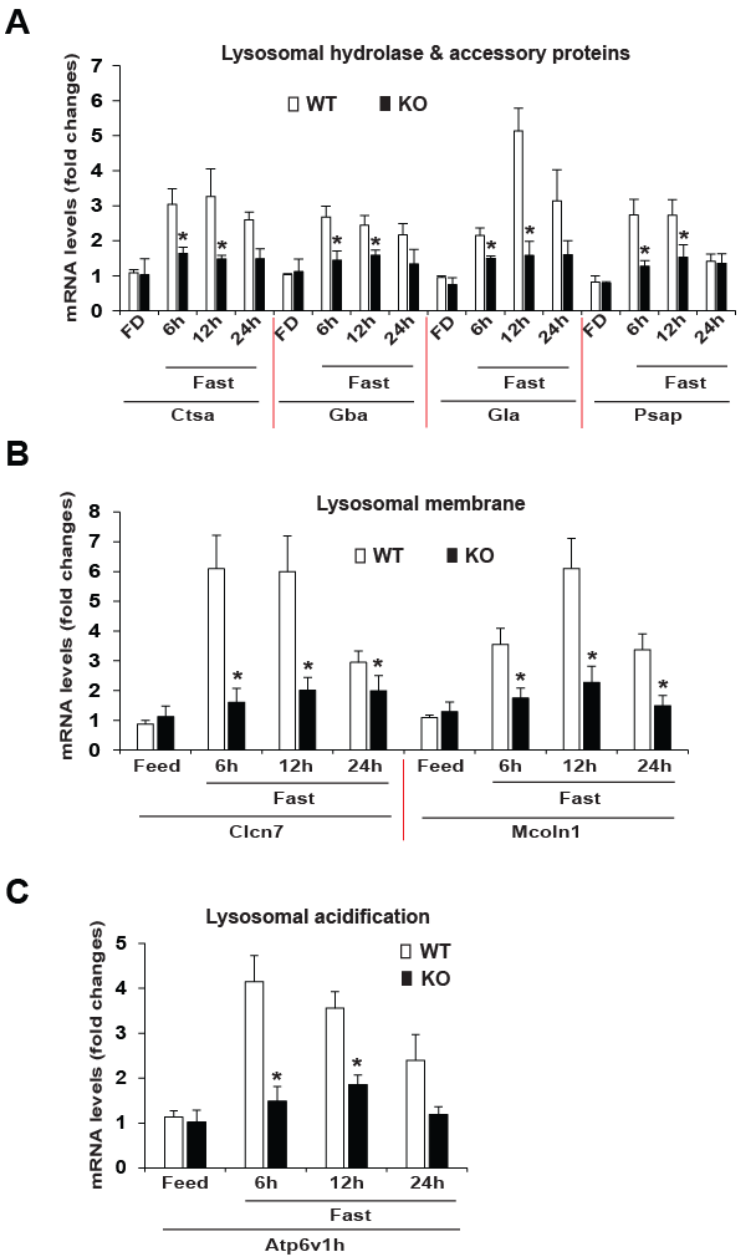
S-Figure 1. Co-immunofluorescence staining of LC3b and HNF4α in the liver tissue sections of WT and CREBH-KO mice. Co-immunofluorescence staining of LC3b and the hepatocyte-specific marker HNF4α proteins in the frozen liver tissue sections from WT and CREBH-KO mice fed normal chow or fasted for 24 h. Liver tissue sections were stained with DAPI for nuclei (Blue). LC3b protein was visualized by green fluorescence, and HNF4α protein was visualized by red fluorescence. Magnification: 40 ×.



S-Figure 2. Autophagosome size analysis as well as levels of CREBH and CREB in the livers of WT and CREBH-KO mice. (A) The sizes of autophagosomes in the liver tissues of WT and CREBH-KO mice under feeding or after fasting. Transmission electron micrograph (TEM) analysis was performed with liver tissue sections from CREBH-KO and WT mice under feeding or after 14h-fasting. The size of autophagosome in the liver of WT or CREBH-KO mice was quantified with the TEM images using ImageJ software. The autophagosome diameter, which reflect the size of autophagosome, was calculated based on the scale bar of the TEM image. Data represent mean ± SD (n= 12 for WT-fed; 15 for KO-fed, WT-fast, and KO-fast). The change in hepatic autophagosome size between WT and CREBH KO mice was not statistically significant. **(B)** Expression and cleavage of CREBH protein were increased in mouse livers in response to fasting. WT and CREBH-KO mice were subjected to feeding or fasting for 6, 12, or 24 h before they were euthanized for liver tissue collection. Western blot analysis was performed with liver protein lysates pooled from three mice per group per genotype to determine the levels of the CREBH precursor [CREBH (P)] and its cleaved form [CREBH (A)]. Quantification of the CREBH precursor and cleaved proteins in the livers of WT mice in the fed state or after fasting for 6, 12, and 24 h. The CREBH protein signals, determined by Western blotting densitometry, were normalized to the β-actin signal. The fold changes in the levels of the precursor or activated form of CREBH were determined by comparing the protein signals to those obtained under the fed condition (defined as 1). **(C)** Western blot analysis of CREB with WT and CREBH-KO mouse liver protein lysates pooled from three mice per group per genotype. Levels of GAPDH were determined as a loading control.



S-Figure 3. CREBH regulate expression of the key autophagy genes in the liver in response to fasting. (**A**) Quantification of LC3b, ATG7, ATG2b, P62, and Ulk1 proteins in the liver tissues of WT mice in the fed state or after fasting for 6, 12, and 24 h. Protein signals, determined by Western blotting densitometry in Figure 3, were normalized to the β-actin signal. The fold changes in protein levels or ratios were determined by comparing the protein signals to those obtained under the fed condition (defined as 1). CREBH (P), CREBH precursor; CREBH (A), activated/cleaved form of CREBH; LC3-I, a cytosolic form of LC3b; LC3-II, lipid-modified LC3b form. (**B**) Quantification of LC3b, ATG7, ATG2b, P62, and Ulk1 proteins in the livers of CREBH-KO and WT mice infected with Ad overexpressing GFP or activated CREBH. Protein signals, determined by Western blotting densitometry in Figure 3, were normalized to the β-actin signal. The fold changes in protein levels or ratios were determined by comparing the protein signals to those obtained under the fed condition (defined as 1).



S-Figure 4. CREBH regulate expression of the genes involved in lysosomal biogenesis in the liver upon fasting. (**A-C**) Expression levels of the genes encoding functions in lysosomal hydrolase and accessory (A), lysosomal membrane (B), and lysosomal acidification (C) in the livers of CREBH-KO and WT mice under the feeding condition or after fasting for 6, 12, or 24 h. mRNA expression levels were determined by qPCR. Fold changes in mRNA levels were determined by comparison to the mRNA levels in one of the WT control mice under the feeding condition. Data represent mean ± SEM (n=3). * P≤0.05. FD, feeding condition.

S-Figure 5. CRE- or PPRE- binding sites in the promoter regions of human and mouse autophagy genes

Human *LC3b* gene promoter

CAAACGCGCCAAGGCTGGGCGTCGAGTGACCGCGGGCGGAGGTCACCAGCGGCCACTCCCCGGAAG
CCACCCACGGACCACGCGCGCCCCTGCACGCAGAGGGGGCCAGGGCTCCACGGGCGAGCGGCGACCC
TGCCTCCCGGAGACGGCGCGGCCTGCCCTGCGCGCCTCAGCCCCGGGTGCCGGCGTCTCGGGCAGCAC
CACCAAGTCTCTCTGGAGGGGAAAGGATGGTCGGATTTGCCCCATGTCCCTTCCTCTGACCCCTCCCT
CAAGAGTGCCCCGGGACACCCCGCCTGTGGCTCAGCCTCCCCCGCCCCGCGCTGCCATCTCCTCAGGG
CCGGGCAGCAGGCTCCCGAGCGCCCACAGACCCGGGGTGCGGCCCAGCCCACAACCGTCACCTCAGG
GGCCTCAGGCGCCCAGCGGTGCTGGGCGGGGCTGGGGCACGACCGGGAGCATGCGCAGAGCGCGCGT
TTCGCCCATCGCGCACGCGCACACACCTGCTCCGCCCCCACGCTGCGTGCCGCTGC

Red highlighted: CRE-binding sequences; ⇨ Transcription start site.
Blue highlighted: PPRE-binding sequences

Mouse *Lc3b* gene promoter

GTGGACAGGTTGCACAGCAGGTGTTGTCATCGTCTTTCTTTTTGCTGGAACAAGTCATTGATCTTGCCC
TCCAGTGACAAGTCTGTAGTCCCTAAGACCTTGCTTCTCTGCGACTTCGACTTAGGAGTAAATACTTGT
GTTTATGGCAGCACAAGACTCGCAGGGGAAGAGCCACAAGATCAGATCCAACCATAAGTCTCCAGAC
GTCTCCATAATCGTAAAGATTTAGCAGGCAAGACCACAAGTCAGTTTTTTGTTATTTGGGTTGGGGGTG
GGTAGAATTTAGGAATATCAAATACAGCGGCAAGGGAGCGGACATTGGACCACCAGCTGGATCATGC
AAGGACTGAAGGGGAATGAAGTCTGCAGGACTCCATCGCACCAGCGCCCGACCCACGCAGACAGTTA
ACAGATGCTCGCCCAGCGTGTGCCCAGCGAGCGCCCAGCGTAGCTGGCGTAGGCGCAGGCGCATGGC
TTGGGCGCGTGCACGCTTGCGCGCCGCCGAGCTTCCGCCACGCCTGTCATGGCCGCTCTCGGGGGCCG
CCCTCGGCCCCACCCTCGGTGACGCGCCGCGAGTCACCTGACTCGGCTGCGGACTGAGACACACACA
AGGGAAGTGATCGTCGCCGG

Red highlighted: CRE-binding sequences; ⇨ Transcription start site.
Blue highlighted: PPRE-binding sequences

Human *ATG7* gene promoter

CTTACAGGCCAGACAGAGAAACTTGTCATAATCACTGCTGTATCTTCAACACCTGTCCAG
TGCTTTGCACGCGCAGAGTATCTAACCAAGTATTGAGAGGGAATAACTTTATCTCACTGA
CAGTGAGGAGCTGTGAGAATGATGGCTGGTTTAGAAAAGGCAGGTTGGTCACTGTCGACG
TTCACTGGCCTTTTCCTACTAAAATTCTCATCTCCTGGCTCTCCACACCTGCCACCCTGA
TGGCCCCTGTGCTGCGTTTGATGCCGCCTCTCCTGGAGAATGACCATGGTGATCATTCCT
GTCATCCTCTGAAATCAAAAGAGAGAACGTGGGCACTTTCTTAAAAGCCTGAAGGGAATG
TAGACATTCCGACATCTGGTAAGGGAGACGCTCTCCATCGCTTCCCCCGGGGGCGTCACC
GCCCCCTGATGCCCCGCCCTCCTCACGACTCAAGTTCCTCCCACTTAGCTCGGTGCTAGG
GGCGCATTTCCCCGGCATGCCTCTGCTCCTTTGCGCACGCGCGCCGCTTCCCAGTGGCAA

Red highlighted: CRE-binding sequences; ⇨ Transcription start site.
Blue highlighted: PPRE-binding sequence

Mouse *Atg7* gene promoter

CTATTTGGTTTTCCAGAGTTATATTTGGTTGGGCAAGCTTTTAATCCCAGCACTGGGGCAAAGCAAAG
GTAAGCAATCTCTGTTAGTTCGAGACTAGACTGATGTATATGTATACAGGTCCAGAACAGCCATGGCT
ATGTAGTGTAAATATGAGGAGGACATAACCTTTTTAAAAAATTAAAAATGCAAAAAATAAAAATAAA
CCTTGTTTCAACACCTGTACTCTAGTACTACCCTGGGAATTTGATAATAATTCCGTAGTACTGTAGAGT
CACTCGCAGGAAAATGTAATCTTCCTTACAAGCCCCAAAAGGCTTACAGGTTAGCCTTAGTTTCGCCCC

GTTATTGCGGTTGTGGGGACCCTTTTTGTCGTTGTTGTTGTAGTGCGCATGCGCGCCGCGCTTCCGCGTT
TGTGTGGGCTGGGTTA**TTACGTCA**TTGGGCCGCGCGGCGCGGCTGGGGCTGTGGTTGCC

Red highlighted: CRE-binding sequences; ⇨ Transcription start site.
Blue highlighted: PPRE-binding sequence

Human *ATG2b* gene promoter

GGTTTTGGATGATTAGGCTGTGTGTGCGTGTGAGAATGATCACATGTGGCGTCATGCTTTGTACAGAGC
CTCAGACCACTGGGCCTCGTCCAGTGAGAGTCCTCTCTGGCGACATCACACGCGGAGCAGCCAGGGGC
CACCTTAGATCTCAGATCTCTCAGAGCAATACTTTTCTGAACTGCCACTGTGCCTGGGTGGTTGGGTTG
GTGTCATGCTTCTGACTAGAGTAGATCGCGCATGTCCACCAGTGATACGTTGAGTCCTTACAGTTCCCC
CCATGGAGTCCCATAAGCAGCTCCATCGAGATCTGTCAGCAAGTTGCAGGACCCCACAATGTTCTGAC
ATGTTAAGACCCCCTTACATGACGAGTAGAGAGGCAGCTGAGGCCACAACCGTGTCTTCCTCTTGAAT
GGAGCTAACTCGGAACCCCCCGTTTTCTCTTCCTTTCTGCCCACCACTGAACATTGCCTTTTAGATAACT
CAGTGTTTCTTCTAGATGTCATAGCAATAGACTTTCACTTTCATGAAGTTTGGGTACGATTTGGATTCT

Red highlighted: CRE-binding sequences; ⇨ Transcription start site.

Mouse *Atg2b* gene promoter

CTTCCCGCAGGACAGCCCAGGCCGGACCCGCCCCCAGACCTC**CGTCA**GCCCTAGTCCGGGCGTCCGCC
GGCGCCCACCACCCGGCCGCGACTGCAAGGTGAGGCTCCACGGGCCGCAGCCAGCCCGCCCACTCAC
CTGCGAGCGCCCATGTTCTTATCCTTCCTCGTCCGGAGACTGCTGGGGCCAGGATCGCGGCGCCGGCA
CCACCCCCGCCCCCGCAGTTGCCCCGCCCATTCACCACCGGAAATGCTCCGGCCCGGACTCTTTTTTTC
CTGCCTTTCCAGCCCCGCAATA**TGGCGTCA**CTTCCGCTTGTGCCCGTTGCTCGAGT**ACGTG**CCGGGAG
TCTGGGTCTCGCGAACGCGCCAGGAGCGTGTCTCAGGCGGGAGCGCGCCCAGTAGATGACAA

Red highlighted: CRE-binding sequences; ⇨ Transcription start site.

Human *ULK1* gene promoter

CCCGC**TCCCGGGTCA**GCGCGCCTCTCTGCTCTCACCGGGGCGGGTCCGCTGTTGGCCCCACCC**ACTCC**
CAGCCCACCCGGCCCCGCAATGGCCCGGCCCAGCCGCCCACCCCCCCGACAGGCCCCGCCCCATTCCC
CGCCCCCCGTCTGTTCGTCACGCCCGGCTCTGCCGCTCCGTCCCGCAGGCCCCGCCCTCATCCCGCCTC
CCGGCCCGGCGCGCAGTGCGGCTGCGCGGGCGTCTCAGGCTCTGAGGCCCGGGCGCCGCGGCTCTTTT
GTTTCTCCGTTGGGGCCGAGCCCGGGCCCTAGTTGGAGCCGGAGTCGGAGTCGGAGTCGGATCCGGAT
TCGGATTAG

Red highlighted: CRE-binding sequences; ⇨ Transcription start site.

Mouse *Ulk1* gene promoter

TT**TCACGTG**TTTATATACACATACACGGTGCATATAAACTCCTGTAGGCACACACATATACATATGAA
TAAAA**TAAGTAA**ATAAAAAAATGCCTAGGGCAGAGCGAGGAGCCACAACCAGAGATAGTTCTGACAT
TGACATGCCCAGTCTGCTCCGGCCCCGCCCATATCCTGCCTCCTCTCTCAAGTCCCGCCCAGTATAGG
GCTGGGCCCATCGAGGCACCACCCAAACTAAGCCCCGCCCCGCCTTACCACCCACGAGGTGTCCTTCA
TTTCGGACCCGCCCATTGCCAGCTCCGCCGCCTAAGAGGTCTTGAGCTCCGCAGGCCCCGCCCATCGTC
CCGCCTCCACGCA GTGCGGCTGC GCCGGCACCT CGGGCTCCGA GGCCCGGGTG CCGCGGATCT

Red highlighted: CRE-binding sequences; ⇨ Transcription start site.

Human *TFEB* gene promoter

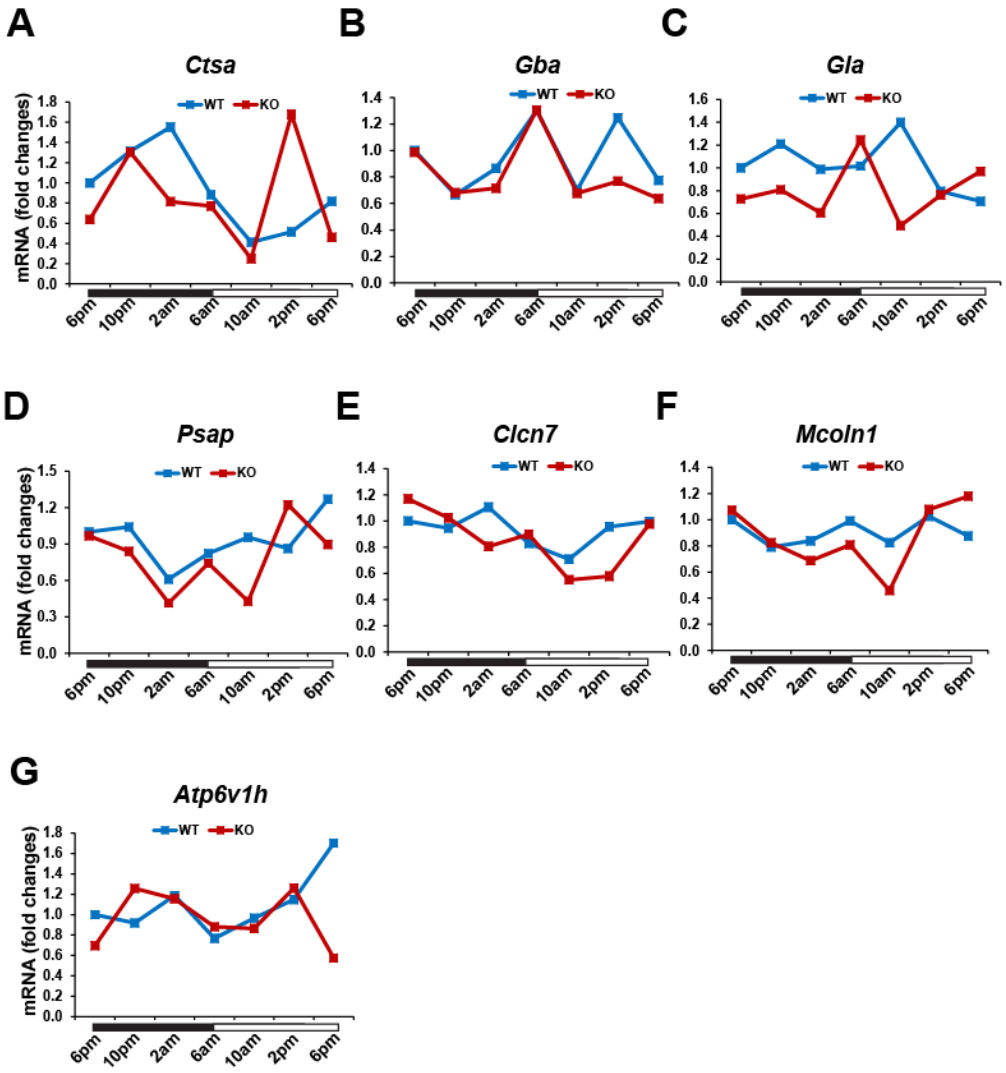
AAAGGAGCTCAGACAATTTCTGTCCAGCAACATGACAGCAAGCTCAGGCTGGTGAGTGGGGCGTGAA
TGTAGAACCCATAATGGGGGCAGGCTGCCTTCTACTGTCATATTTTACCCTGTACTTCGCC**AGGCTCA**C
CCTGGCCTTGCCCCTTATGATCGGGGCAGGGATAAAAGTGCAAGATCCAGGTCCTGCACCCTACTCCT
GAAGAGAAGCTGGAATTTGCCACAGGTGGAGTTTCTATGGCAACAGCACACCATCTGGGACCCTCTCT
TTCTTAGACTGCCGAAGGACAGCAGGCTGCAGGTTCCTTTTAGGGGGTCGGACAGGCTACAGAATGTA
TCTTAGTGAGCCCTGGTTTGGGACTTTACTGAGAGGAGAAAGAAAACTTGCAAAGAGGTAGAAGTGG
ATCAGAGGTTCCAAGTCAAGAGCCTCTTTCCCTGTAAGCCTTAGTCCGCTGAGCGGGACTACAAATCC
CGGCGTGCCTAGCGGG**ACTGCA**GCGAGAACTACGCTTCCCAGTGGGCGCCGAGGCAGAGCGGCTGGG
AGGCGGGGCGGGTGCTGCCCTCTGCCGGGCGCGGAGGGAAGTGTGCGTGGAGTGACAGCCGGAGCCC
GGGCGCCGGGCGCGGGGCTCGGTGACAGCGGAGGCGGCGGCCCGGGCGGGACGCAGGGAGCGGCCG
CGGCGCCGACGGCGGCGGGAAGGGC

Blue highlighted: PPRE-binding sequences; ⇨ Transcription start site.

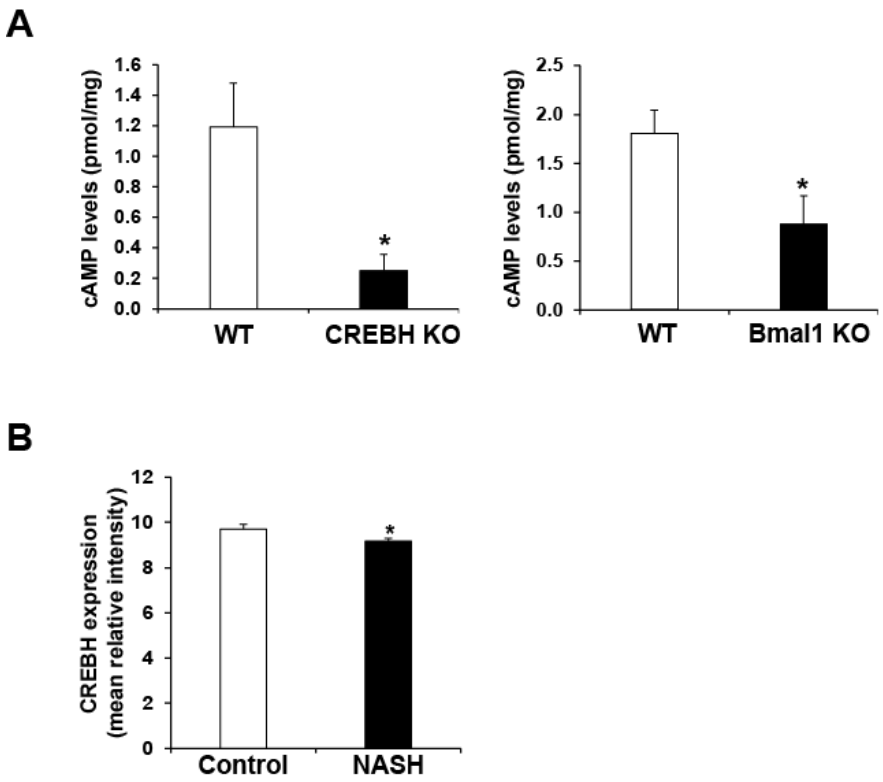
Mouse *Tfeb* gene promoter

CAAAAGGGCAAGATCCAGGTCATGTACCACATTCCAAACGGAGACAGGATATATCGTGAAAACGGTG
GGCTCTTGAGCAAGGGGTTTTACATCTCACACCTGCCGGATGAAACCAAAAATCATGCTCCGAGCACA
GTGGCCGTTTCCCACCAAGCAGGCAAACTTGCAGAGGGCATTCATTCCATCAGGAGTGTCTGAATAAC
AAAACAAGTCAGGAGCAGCGGCACACACCTCCGACTCCAGCCTTCTCGGAAAGCAGAGGCAGGCAGA
GCTCCGTCAGATTGAGGGCACTATAATCAACTAATGAATTCTAGGCCACCTAGGGTTATTGGGTGAGT
CTGTCTCCACGAAACAAAGCAATGGTAGAAATAACAATAAACCAGGTTTTAAGGCCAAGCACGTGAA
CCCTGGAGTGTGGCTTCAGTGCTCTCGCAGACCTTGCCTGGAAATCTCACAAAGAGAACTGTTTTGATT
ACAGTTAAGATATCCTGCCTTCCTTTATAACCTTTTACCTAATCAATTACAACAGTATCTGCTACACCC
CAGGAAACGTCCCGTGATCTTATTTAAATTTCTCAGCAGTCCTTTGAAGATAACTGCGGGACTCACCAA
AACAAAAGTATGGCTATGGTAAAAGACAAGGAGAAGCCCTTGCCCAGTTTCTCTGTCAACTTCCTCTT
AATCCCTATGTTTTAAATCTACATTAAAAAAAAATATATATATATATATATATTTAAACCAGGCAGT
AGTGGTATACACTTTTAATCCCAGCACTTGAAAGACAGGTGGATCTCTGTGAATTTAAGGCCAGACTG
TCTACAA**ATCTAGTTGTAGAACAGCCA**AGGCTACACAAAGAAACCTTGTGTTAAAGAACCATAT**ATAT**
ATACTTACATAAATATATATTTAATAAACTAAAGCTCTGCTTCATACTGGCTTGTCCTGAAATTATAAT
AATAATTAATAGAGCAACAACAATAATATT**GTTATTATTATTTA**ACAAAGAAATCAAAACTCTCCCC
AAGTGGAAGTTGCTAAGGGATAGGGTAACTTCTCAGGTTGCTGCTGGCAATTATGGGGGTGGGATCCT
ATATTTTTCTCACACTTTCTGGAAAAGCAGCTTGCTATTGGTCCTTTTTCAAGATGTAATAATATAGAA
AGTCATATCACGGGCCATCATTGACAGAAAAAAAGAGAGAAAATCTGAAAAGGGAATAAAGACAAA
CGTTTAAAGTTAAAAATTGACTCCTAGGGAACAGCGGCCAGTAGAATGGAACTGCAAATCCCGGCGA
GCCCTTCGCGGCTGCGACGGGGACTACATTTCCCAGCGGGCACAGCGGTAGGCCTATGGGGGGCGGTC
CGCAGGAGGGGCGGGTGCTGCCCTCTGCCGGGCGCAGCGGGAAGTGGGCGCGGAGTGACAGCCGGA

Red highlighted: CRE-binding sequences; ⇨ Transcription start site.
Blue highlighted: PPRE-binding sequence



S-Figure 6. CREBH regulates rhythmic expression of the genes involved in lysosomal biogenesis in the liver of mice under the circadian clock. (**A-F**) Rhythmic expression levels of *CTSA*, *GBA*, *GLA*, *PSAP*, *CLCN7*, *MCOLN1*, and *ATP6V1H* genes in the livers of CREBH-KO and WT control mice across a 24-h circadian cycle. mRNA expression levels were determined by qPCR. Fold changes in mRNA levels were determined by comparison to levels in one of the WT control mice at the starting circadian time point. Each expression point represents a pooled liver sample from 3 to 5 mice per group per time point. Black and white bars represent circadian dark-light phases.



S-Figure 7. Analyses of cAMP levels in CREBH-KO or Bmal1-LKO mouse livers and *CREBH* mRNA levels in human NASH patient livers. (A) Levels of cAMP in liver tissues of CREBH-KO, Baml1-LKO, and WT control mice determined by Cyclic AMP Competitive ELISA Kit (Invitrogen). Each bar donates mean ± SEM (n=4). Total protein concentrations (mg/mL) of homogenized liver tissues were determined for normalizing the sample loading. The mean value was expressed as pmol cAMP per mg of total liver proteins. **(B)** Expression levels of *CREBH* mRNA in livers of human patients with non-alcoholic steatohepatitis (NASH) and control individuals without NASH. The analysis presented in this graph was conducted on the dataset obtained from the National Center for Biotechnology Information (NCBI) Gene Expression Omnibus (GEO) Database (www.ncbi.nlm.nih.gov/geo/). The dataset we analyzed was deposited by Ahrens et al (*Cell Metab* 2013, 18: 296-302). Liver samples were obtained from patients undergoing liver biopsy for suspected NAFLD or intraoperatively for assessment of liver histology. Control samples were obtained from individuals excluded for liver malignancy and with liver histology that demonstrated absence of both cirrhosis and malignancy {Ahrens, 2013 #73}. Expression levels of mRNAs were based on the mRNA microarray data obtained from the GSE48452 dataset from the GEO database. The mean relative intensity for *CREBH* mRNA is shown. Each bar donates mean ± SEM (n=12 for Control and 14 for NASH). * $P \leqslant 0.05$.