human	Forward	Reverse
ALDH2	GATTTGGACAAGGCCAATTACCTG	AAGGGTGACTGGGCTCCAAAC
HMGCR	TCTGGCAGTCAGTGGGAACTATT	CCTCGTCCTTCGATCCAATTT
HMGCS	GATGTGGGAATTGTTGCCCTT	ATTGTCTCTGTTCCAACTTCCAG
β-Actin	GATCATTGCTCCTCCTGAGC	ACTCCTGCTTGCTGATCCAC
ABCG5	ACCCAAAGCAAGGAACGGGAA	CAGCGTTCAGCATGCCTGTGT
PCSK9	CACCCTAGAAGGTTTCCGCA	TCACTCCTCCAGGCTCAGAC
Idol	GAGAAACCGGATCTCCCAGC	CTGGGGAACACAAGAGGTGG
LDLR	CAGTCTGGAGGATGACGTGG	ACTGTCCGAAGCCTGTTCTG
mouse	Forward	Reverse
ALDH-Geno-Forward(5'-3')	CGGGAATTGAACTTGGTAGCCAG	GCGTAAGGCATGCGCCATCAC
ALDH-Seq-Forward(5'-3')	CCTGAGCCGAATGCTTTTAAG	CTCACGCTCCTTACTGGAC
LDLR Forward(5'-3')	ACACAGCCTAGAGGGGTGAA	AGGTGAATTTGGGCGAGTGG
PCSK9 Forward(5'-3')	GCTTCTGCTCCAGAGGTCATC	TGTGAGGTCCCACTCTGTGA
L32 Forward(5'-3')	TTAAGCGAAACTGGCGGAAAC	TTGTTGCTCCCATAACCGATG

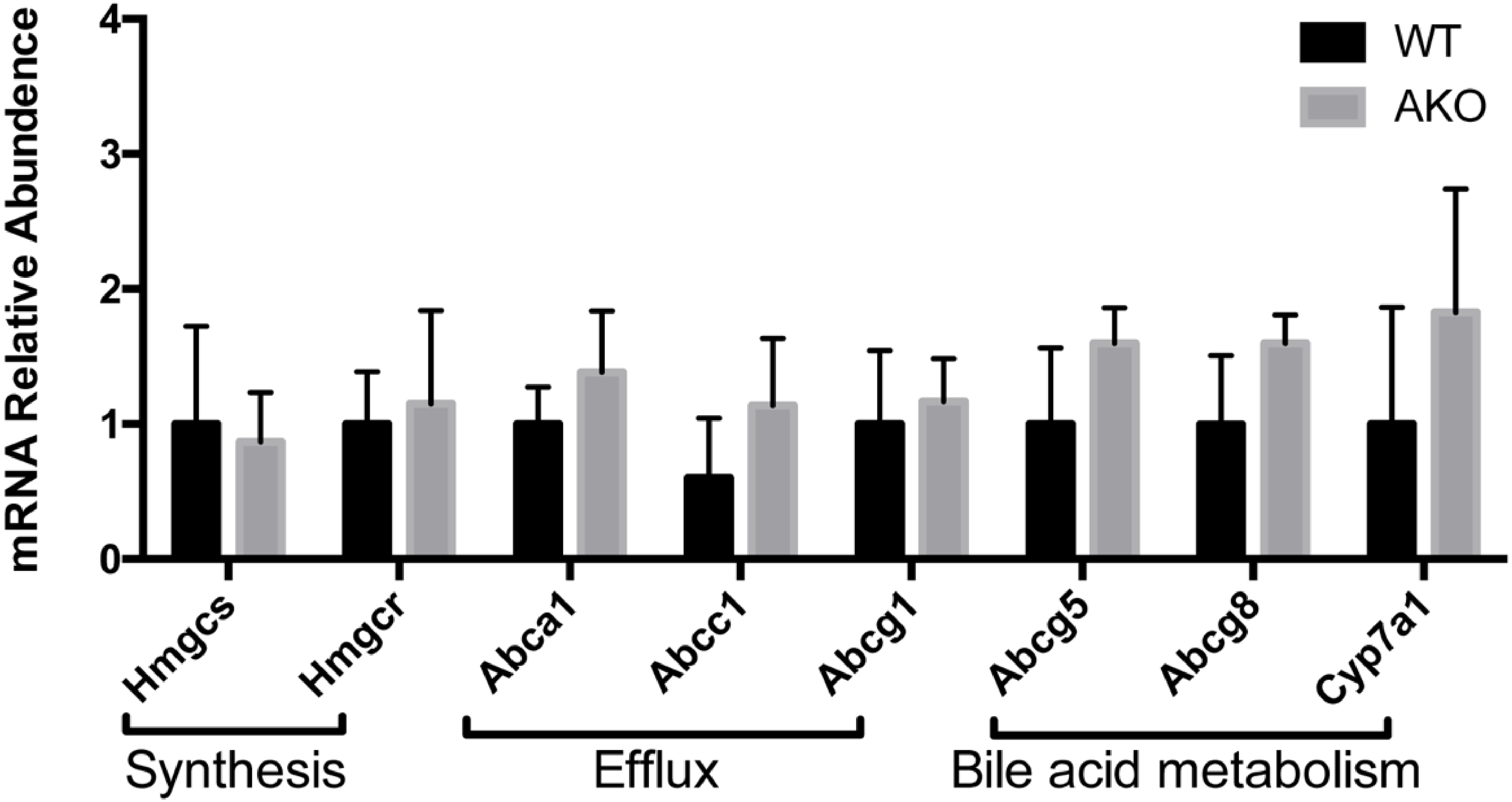
Supplemental table 1. Primer sequence for human and mouse

Location	Protein			
Mitochondrial	40S ribosomal protein S3	DNA-directed RNA polymerase, mitochondrial	Dimethyladenosine transferase 1, mitochondrial	ATP-dependent Clp protease proteolytic subunit, mitochondrial
	Ubiquitin-40S ribosomal protein S27a	NADH dehydrogenase [ubiquinone] iron-sulfur protein 5	Probable dimethyladenosine transferase	39S ribosomal protein L14, mitochondrial
	ATPase family AAA domain-containing protein 3B	Mitochondrial proton/calcium exchanger protein	Calmodulin 1 (Phosphorylase kinase, delta), isoform CRA_a	Leucine-rich repeat-containing protein 59
	ADP/ATP translocase 2	Argininosuccinate synthase	Succinate dehydrogenase [ubiquinone] flavoprotein subunit, mitochondrial	Protein/nucleic acid deglycase DJ-1
	ATP synthase subunit alpha, mitochondrial	Phosphate carrier protein, mitochondrial	Voltage-dependent anion-selective channel protein 3	Pantothenate kinase 2, mitochondrial
	Serine/threonine-protein phosphatase PGAM5, mitochondrial	Pyruvate carboxylase, Mitochondrial	NADH dehydrogenase (Ubiquinone) 1 alpha subcomplex, 10, 42kDa variant	cDNA FLJ59258, highly similar to Grave disease carrier protein
	60 kDa heat shock protein, mitochondrial	cDNA FLJ60124, highly similar to Mitochondrial dicarboxylate carrier	tRNA dimethylallyltransferase	Vesicle amine transport protein 1 homolog (T californica), isoform CRA_a
	28S ribosomal protein S34, mitochondrial	Lysine--tRNA ligase	Voltage-dependent anion channel 2, isoform CRA_a	
Endoplasmic Reticulum	Insulin-induced gene protein	Aminoacyl tRNA synthase complex-interacting multifunctional protein 1	Protein disulfide-isomerase	40S ribosomal protein S3
	p180/ribosome receptor	Golgin subfamily B member 1	Prolyl 4-hydroxylase subunit alpha-1	60S ribosomal protein L10
	tRNA-splicing ligase RtcB homolog	Double-stranded RNA-binding protein Staufen homolog 2	60S ribosomal protein L36a	60S ribosomal protein L10
	40S ribosomal protein S3a	Calreticulin	Signal recognition particle 14 kDa protein	Dystonin
	Heterogeneous nuclear ribonucleoprotein R	Unconventional myosin-Id	60S ribosomal protein L27a	40S ribosomal protein S8
	60S ribosomal protein L5	Vesicle-associated membrane protein-associated protein B/C	60S ribosomal protein L36a-like	60S ribosomal protein L21
	60S acidic ribosomal protein P0	Argininosuccinate synthase	Signal recognition particle subunit SRP68	60S ribosomal protein L18

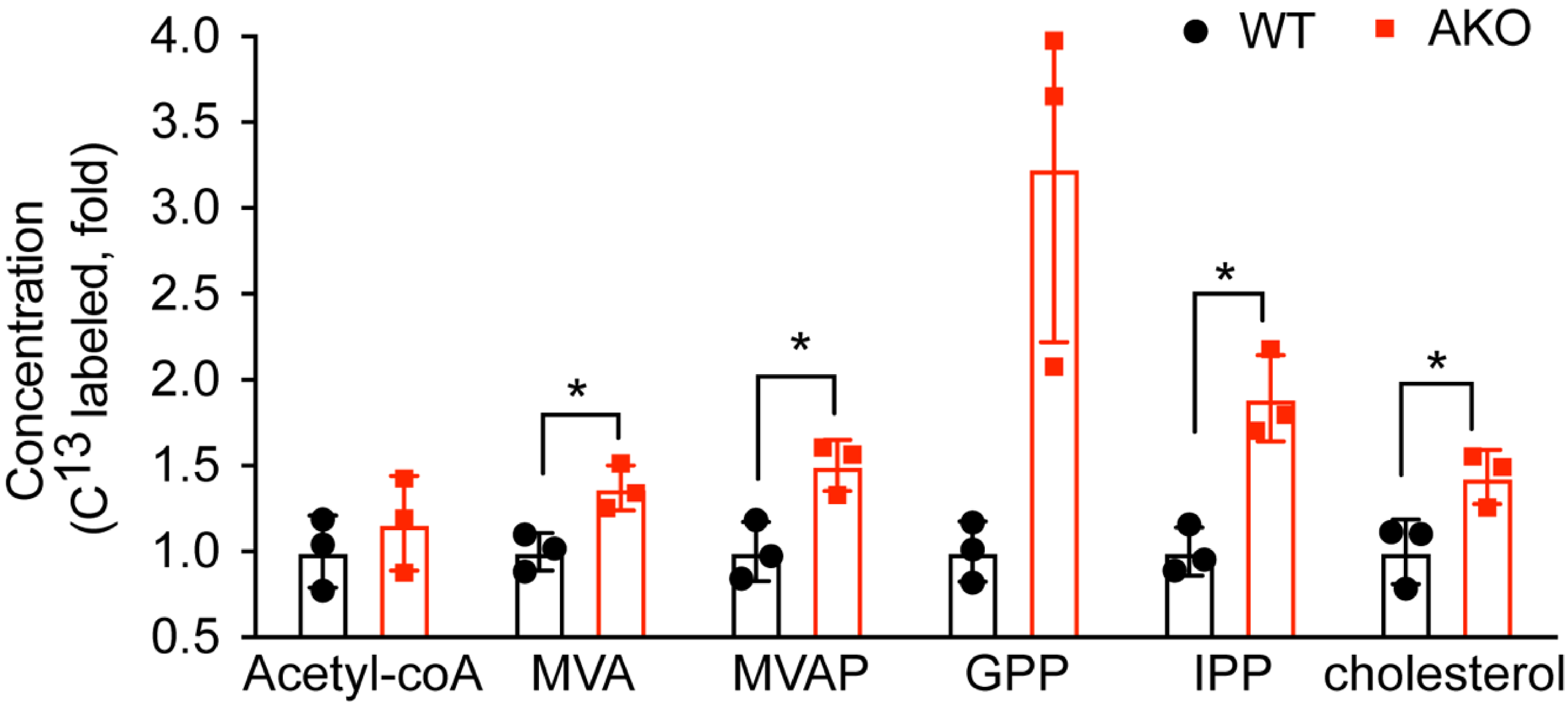
Location	Protein			
Endoplasmic Reticulum	Endoplasmic reticulum chaperone BiP	Fibrinogen alpha chain	40S ribosomal protein S26	Leucine-rich repeat-containing protein 59
	Protein-glutamine gamma-glutamyltransferase 2	Dolichyl-diphosphooligosaccharide--protein glycosyltransferase subunit 1	Sphingosine-1-phosphate lyase 1	Signal recognition particle 9 kDa protein
	DnaJ homolog subfamily A member 1	Signal recognition particle 19 kDa protein	Endoplasmin	NACHT, LRR and PYD domains-containing protein 3
	Huntingtin	Alpha-2-HS-glycoprotein	Aspartyl/asparaginyl beta-hydroxylase	Protein/nucleic acid deglycase DJ-1
	Translocon-associated protein subunit alpha	Myeloid-derived growth factor	40S ribosomal protein S28	Perilipin-2
	Ubiquitin-40S ribosomal protein S27a	Transitional endoplasmic reticulum ATPase	Membrane protein MLC1	Vesicle transport through interaction with t-SNAREs homolog 1A
	Protein SEC13 homolog	40S ribosomal protein S29	Metadherin, isoform CRA_a	Calcium load-activated calcium channel
	Epididymis secretory protein Li 310	Ribosome biogenesis regulatory protein homolog	Very-long-chain (3R)-3-hydroxyacyl-CoA dehydratase	Emerin
	Ribosomal protein S23, isoform CRA_a	Pyridoxal-dependent decarboxylase domain-containing protein 1	Protein jagunal homolog 1	Cytoplasmic protein
	60S ribosomal protein L27	U3 small nucleolar RNA-associated protein 15 homolog	60S ribosomal protein L4	

Supplemental table 2. Mitochondrial and ER proteins binding to ALDH2 in 7702 cells.

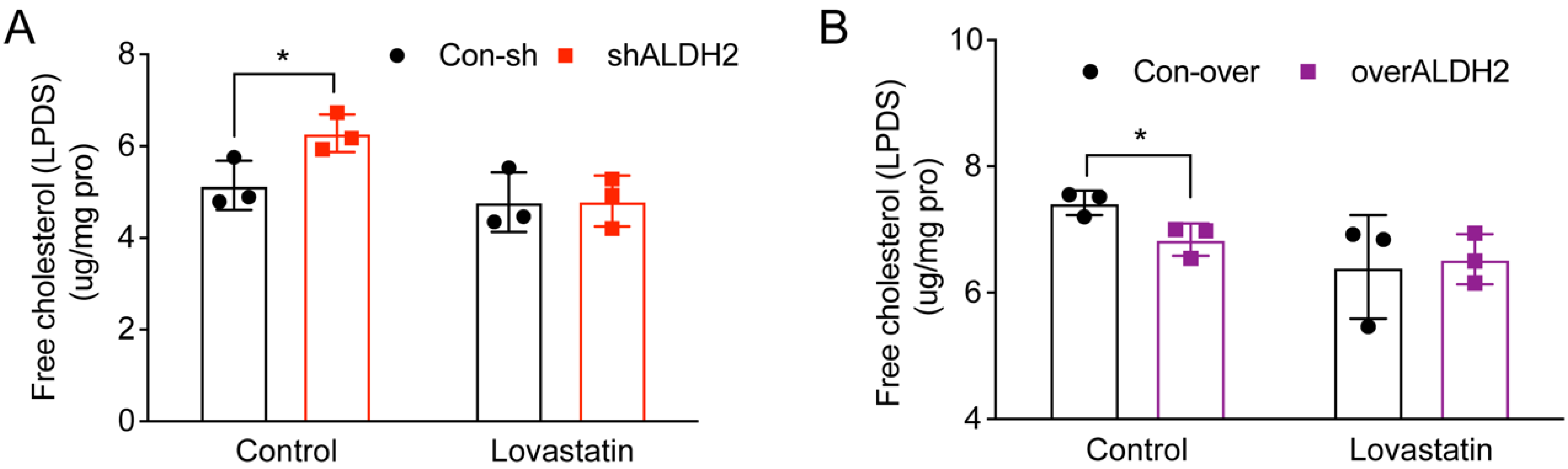
A



Supplemental Figure 1. mRNA levels of cholesterol metabolism-related genes in WT (wild type) and AKO (ALDH2 knockout) mouse liver (chow diet, n=5).

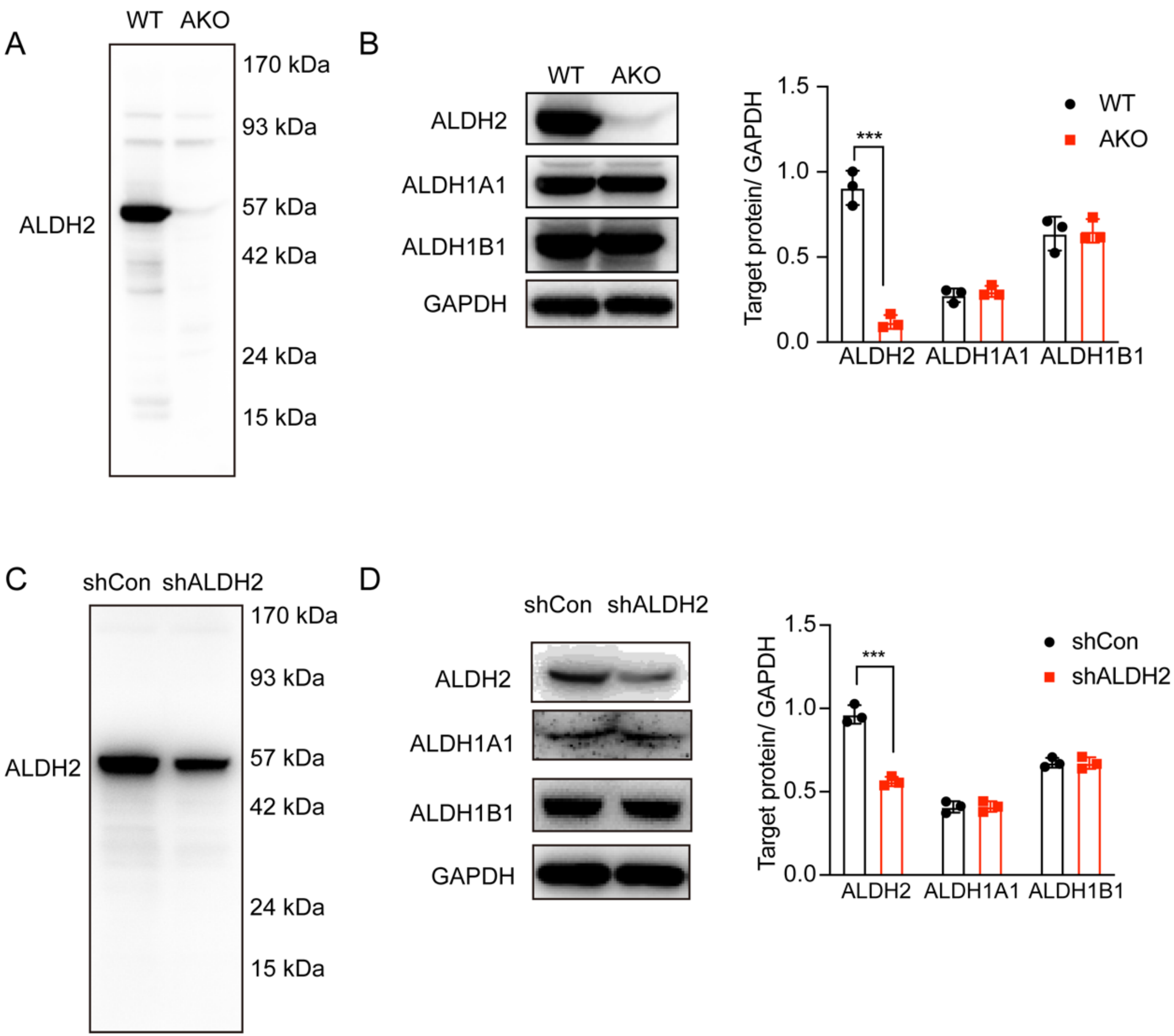


Supplemental Figure 2. LC-MS analysis for intermediates in *de novo* cholesterol synthesis by using ^{13}C-acetate to incorporate into cholesterol in WT and AKO hepatocytes (n=3). Statistical comparisons were made using a 2-tailed Student's t test. All data are mean ± SD. *P < 0.05.



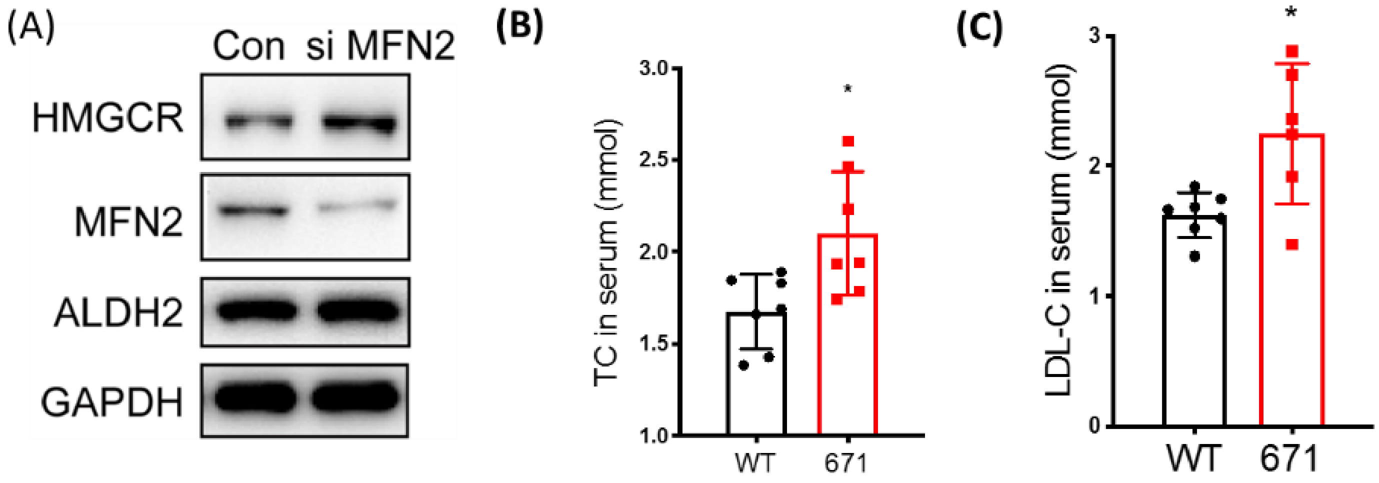
Supplemental Figure 3. Lovastatin treatment inhibits the free cholesterol levels due to knocking down ALDH2 or overexpressing ALDH2.

(A) GC-MS analysis of free cholesterol levels in 7702 cells knockdown (A) or overexpressing ALDH2 (B) and Lovastatin treatment (n=3). Statistical comparisons were made using a 2-tailed Student's t test. All data are mean ± SD. *$P < 0.05$.

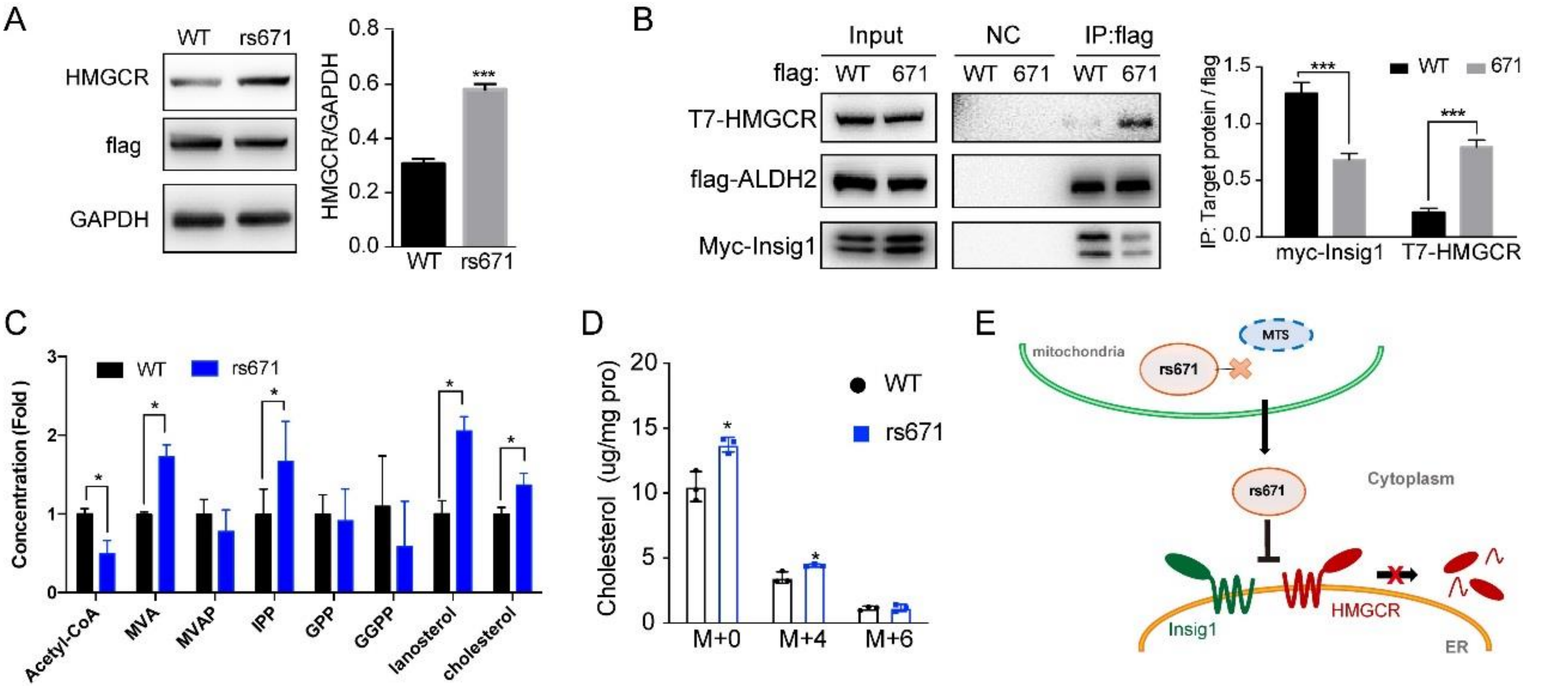


Supplemental Figure 4. Specificity of ALDH2 antibody against other ALDH isoforms.

(A) ALDH2 expression (whole gel) in WT and ALDH2 KO mouse liver. (B) ALDH2, ALDH1A1 and ALDH1B1 expression in WT and ALDH2 KO mouse liver (n=3). (C) ALDH2 expression (whole gel) in WT and shALDH2 7702 cells. (D) ALDH2, ALDH1A1 and ALDH1B1 expression in WT and shALDH2 7702 cells (n=3).

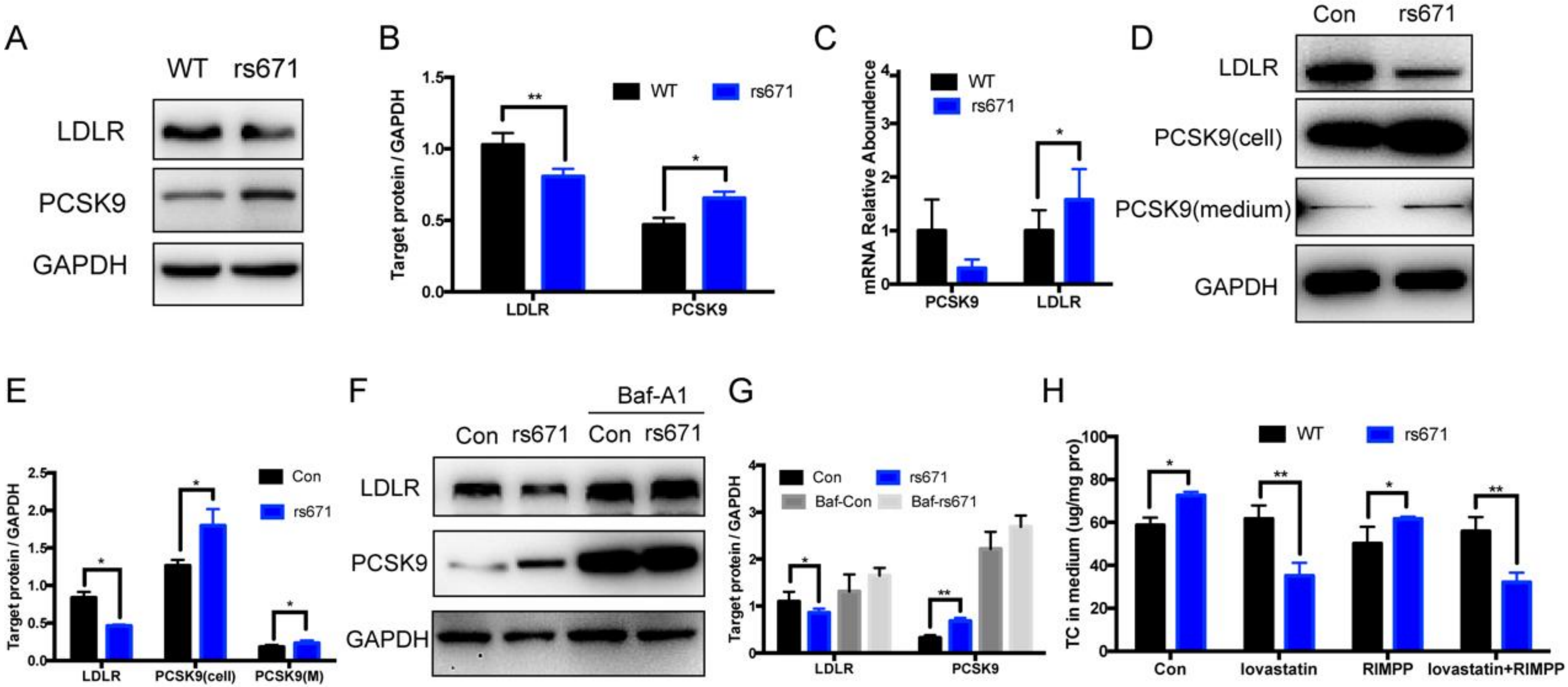


Supplemental Figure 5. A, Western blots of HMGCR in WT hepatocytes treated with si control or siMFN2 RNA. B, Total cholesterol in female rs671 mice; C, LDL-C in female rs671 mice



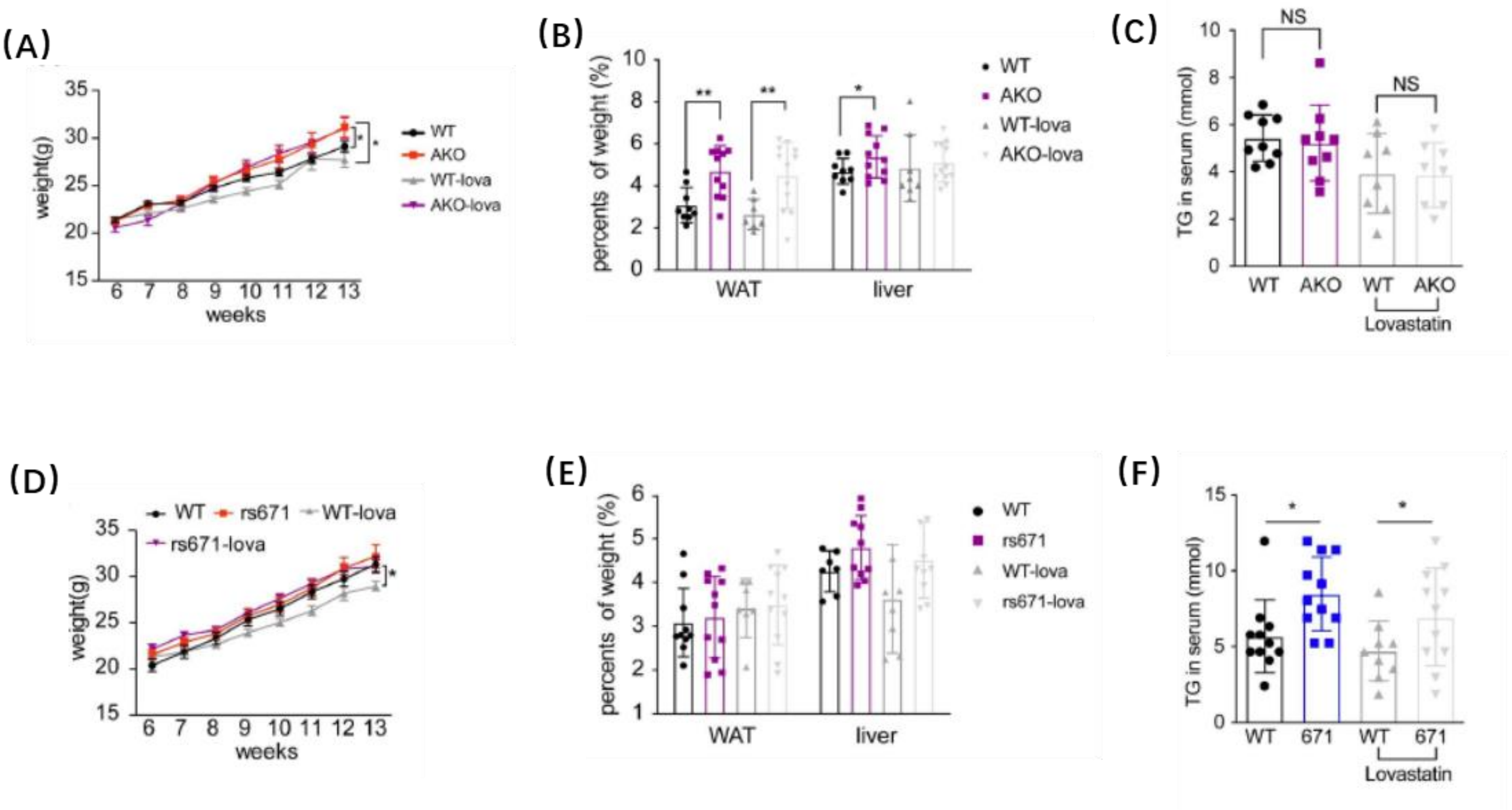
Supplemental Figure 6. ALDH2 rs671 (ALDH2*2) mutant stabilizes HMGCR and increases cholesterol synthesis through decreasing the binding of Insig1 and HMGCR.

(A) Representative image (left) and quantification (right, n=3) of HMGCR expression in 7702 cells transfected with wild type ALDH2 (WT) or ALDH2*2 (671) plasmid. (B) Immunoprecipitation of wild type ALDH2 (WT) or ALDH2*2 (671), HMGCR and Insig1 (left is representative image, right is quantification, n=3). (C) Ubiquitination of HMGCR in 7702 cells transfected with wild type ALDH2 (WT) or ALDH2*2 (671) plasmid. (D) Levels of major intermediates in de novo cholesterol synthesis in WT and rs671 7702 cells (n=3). (E) ^{13}C-acetate incorporation into cholesterol in wild type and ALDH2 knockout hepatocytes (n=3). (F) Schematic summary of ALDH2*2 (671) decreasing HMGCR degradation. Statistical comparisons were made using a 2-tailed Student's t test or ANOVA. All data are mean ± SD. *P < 0.05, **P < 0.01, ***P < 0.001. WT: wild type; AKO: ALDH2 Knockout.



Supplemental Figure 7. ALDH2 rs671 mutant increases PCSK9 expression.

(A-B) LDLR and PCSK9 expressions in WT and ALDH2 rs671 mutant mouse liver (B, quantification, n=3). (C) mRNA levels of PCSK9 and LDLR in 13-week old mouse liver (western diet, n=10). (D-E) LDLR and PCSK9 expression in WT and ALDH2 rs671 mutant hepatocytes (E, quantification, n=3). (F-G) LDLR and PCSK9 expressions in hepatocytes treated with or without Baf-A1 (G, quantification, n=3). (H) GC-MS analysis of total cholesterol in hepatocytes treated with RIMPP or lovastatin (n=3). Statistical comparisons were made using a 2-tailed Student's t test or ANOVA. All data are mean ± SD. $*P < 0.05$, $**P < 0.01$.



Supplemental Figure 8. Tissue weights, percentages of WAT and liver of weight in wild type (WT), ALDH2 knockout (AKO), and rs671 knock-in mice with or without Lovastatin treatment. (A) WT, AKO mouse weights during the course of experiments. (B) Percentage of WAT and liver weight/body weights in WT and AKO mice. (C) Serum TG levels in WT and AKO mice. (D) WT, rs671 mouse weights during the course of experiments. (B) Percentage of WAT and liver weight/body weights in WT and rs671 mice. (C) Serum TG levels in WT and rs671 mice. Statistical comparisons were made using a 2-tailed Student's t test or ANOVA. All data are mean ± SD. *P < 0.05, **P < 0.01.