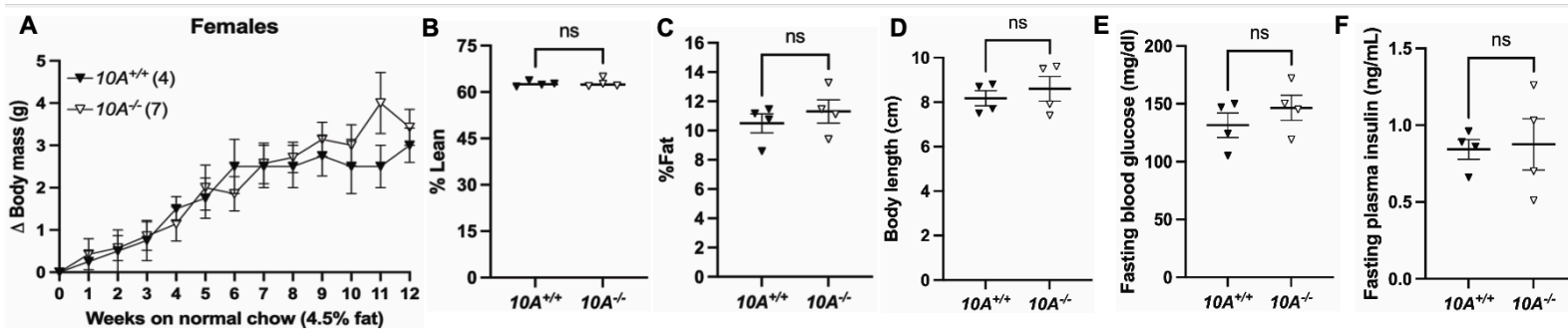
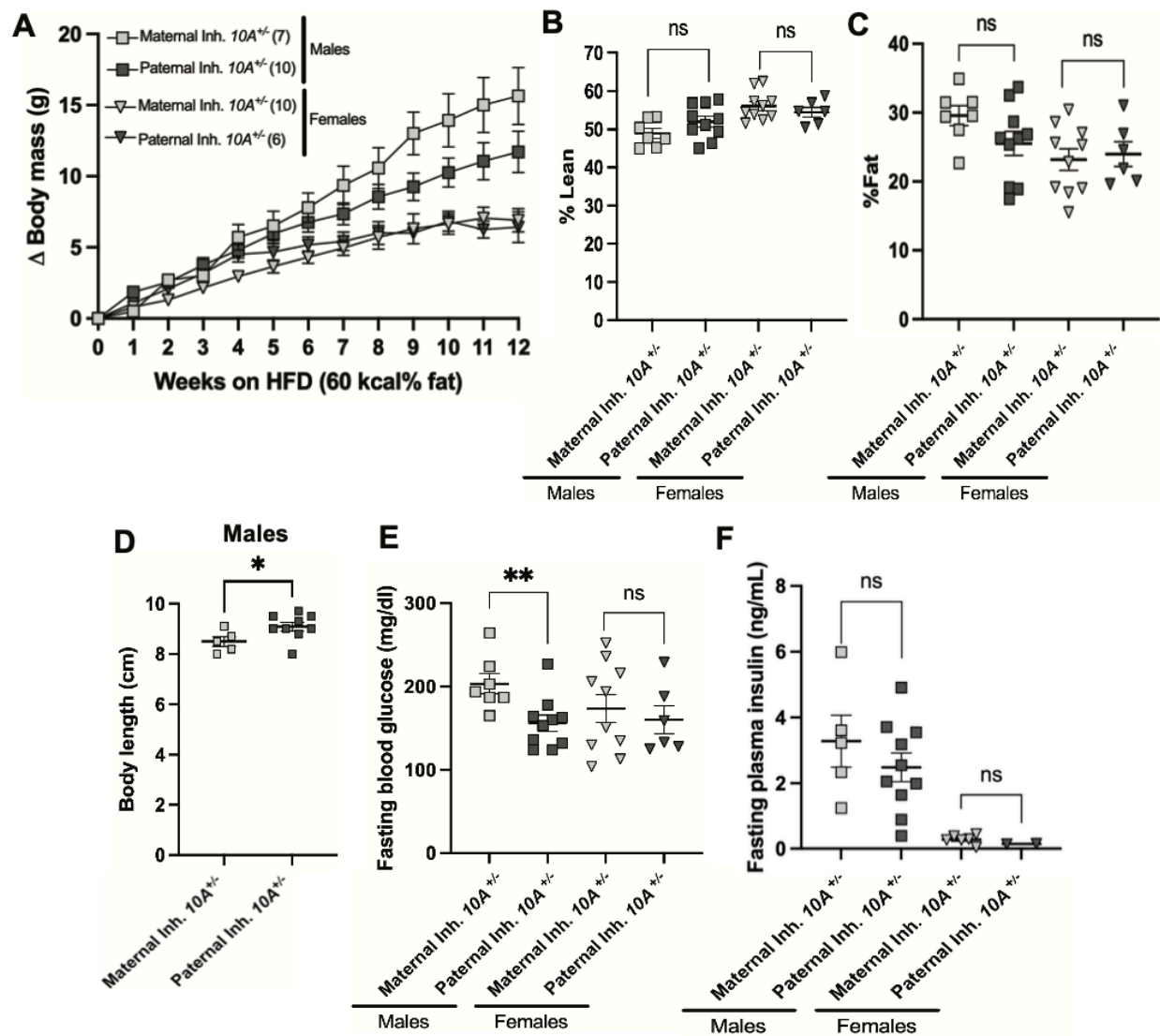


Females	10A ^{+/+} (WT) Average (n=7)			10A ^{-/-} (KO) Average (n=13)			P-value (t-test, WT vs KO)		
	Light	Dark	Full day	Light	Dark	Full day	Light	Dark	Full day
Avg. Daily Food intake (kcal/period)	6.5	9.5	14.4	5.4	9.5	13.5	0.0353 (*)	0.9515 (ns)	0.4425 (ns)
Avg. Daily Energy Expenditure (kcal/period)	17.3	28.9	46.1	17.4	28.4	45.7	0.9596 (ns)	0.5622 (ns)	0.7033 (ns)
Total Food (kcal)	47.3	57.5	57.5	43.7	54.1	54.1	0.2807 (ns)	0.4428 (ns)	0.4428 (ns)
O2 Consumption (ml/hr)	102.9	125	115.5	103.3	123.1	114.6	0.8895 (ns)	0.5891 (ns)	0.7477 (ns)
CO2 Production (ml/hr)	81	98.1	90.8	80.3	95.9	89.2	0.7319 (ns)	0.4586 (ns)	0.5217 (ns)
Energy balance (kcal)	2.1	9.3	11.5	-1.2	9.6	8.5	0.0345 (*)	0.9394 (ns)	0.4886 (ns)
Respiratory Exchange Ratio	0.8	0.8	0.8	0.8	0.8	0.8	0.0839 (ns)	0.3642 (ns)	0.1761 (ns)
Locomotor Activity (beam breaks/hr)	121.5	269.9	206.3	100.9	266.1	195.3	0.2581 (ns)	0.8697 (ns)	0.5679 (ns)
Pedestrian Locomotion (m)	146.9	880.8	1027.7	141.1	843	984	0.8056 (ns)	0.7245 (ns)	0.7222 (ns)
Distances in cage Locomotion (m)	199.6	1017.4	1217	185.5	980.6	1166	0.5972 (ns)	0.7443 (ns)	0.6956 (ns)

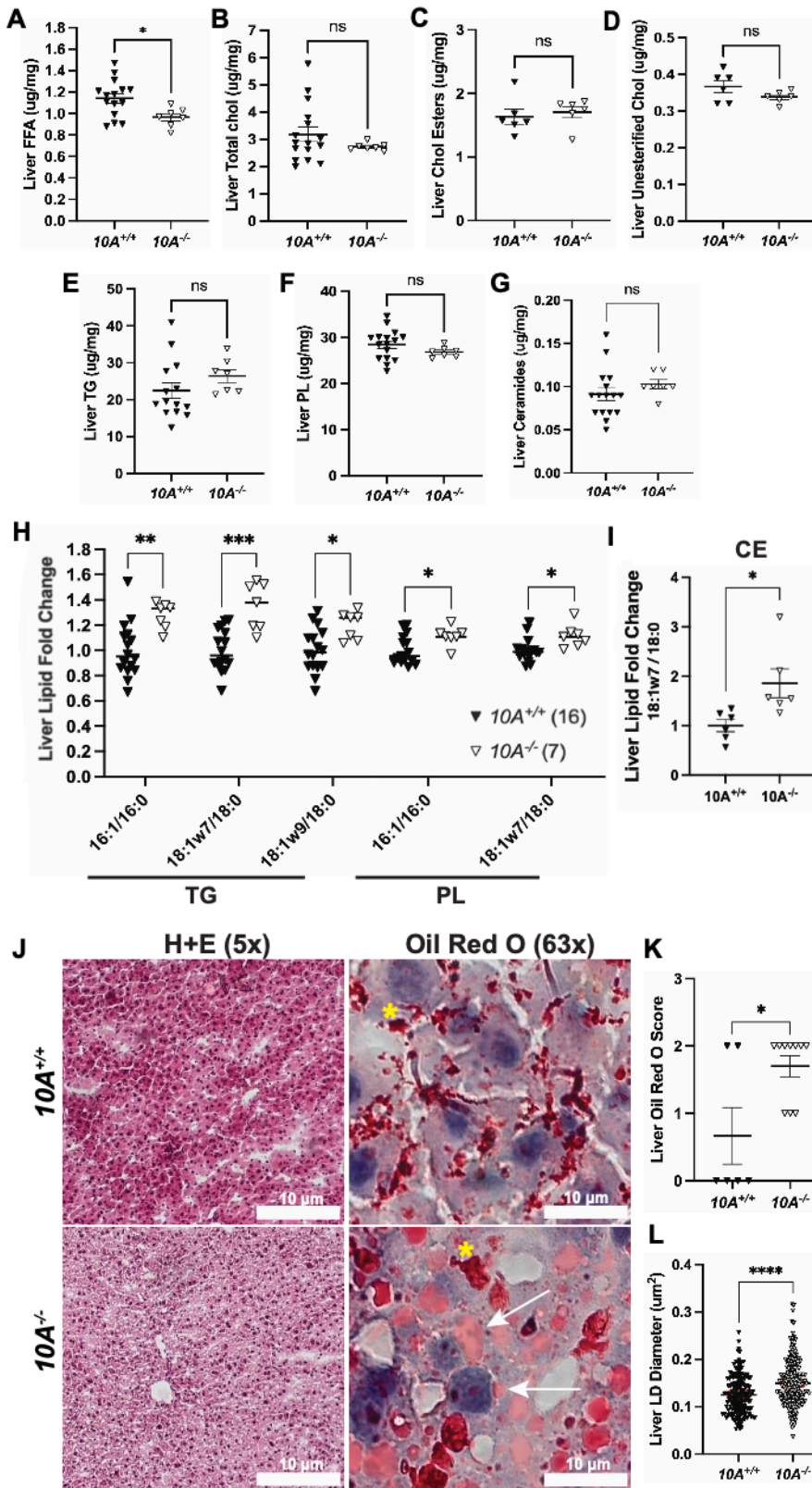
Supplemental Figure 2. *Atp10A* deletion causes alterations to average daily food intake and energy balance during the light hours in female mice fed a HFD for 13 weeks. The table indicates the parameters measured in the Sable System's Promethion Indirect Calorimetry System where the mice were single-housed. The values are an average measurement from 5 days in the system with ad libitum feeding. P value by unpaired t-test ($10A^{+/+}$ n=7, $10A^{-/-}$ n=13).



Supplemental Figure 3. *Atp10A* deletion does not affect weight gain, body composition, or glucose homeostasis in female mice after 12 weeks on normal chow. (A) Weight gain of $10A^{+/+}$ and $10A^{-/-}$ female mice over the course of 12 weeks on normal chow (4.5 kcal% fat, Ad lib feeding), P value by 2-way ANOVA with Sidak's multiple comparison. ($10A^{+/+}$ n=4, $10A^{-/-}$ n=7). (B) Lean and (C) fat body mass was normalized to total body mass to calculate % Lean and % Fat mass. P value by unpaired t-test ($10A^{+/+}$ n=4, $10A^{-/-}$ n=4). (D) Body length of mice was measured after CO₂ sacrifice. P value by unpaired t-test ($10A^{+/+}$ n=4, $10A^{-/-}$ n=4). (E) Fasting blood glucose was measured a 5 hour fast, via a glucometer. P value by unpaired t-test ($10A^{+/+}$ n=4, $10A^{-/-}$ n=4). (F) Fasting plasma insulin was measured after a 5 hour fast. P-value by unpaired t test ($10A^{+/+}$ n=4, $10A^{-/-}$ n=4).

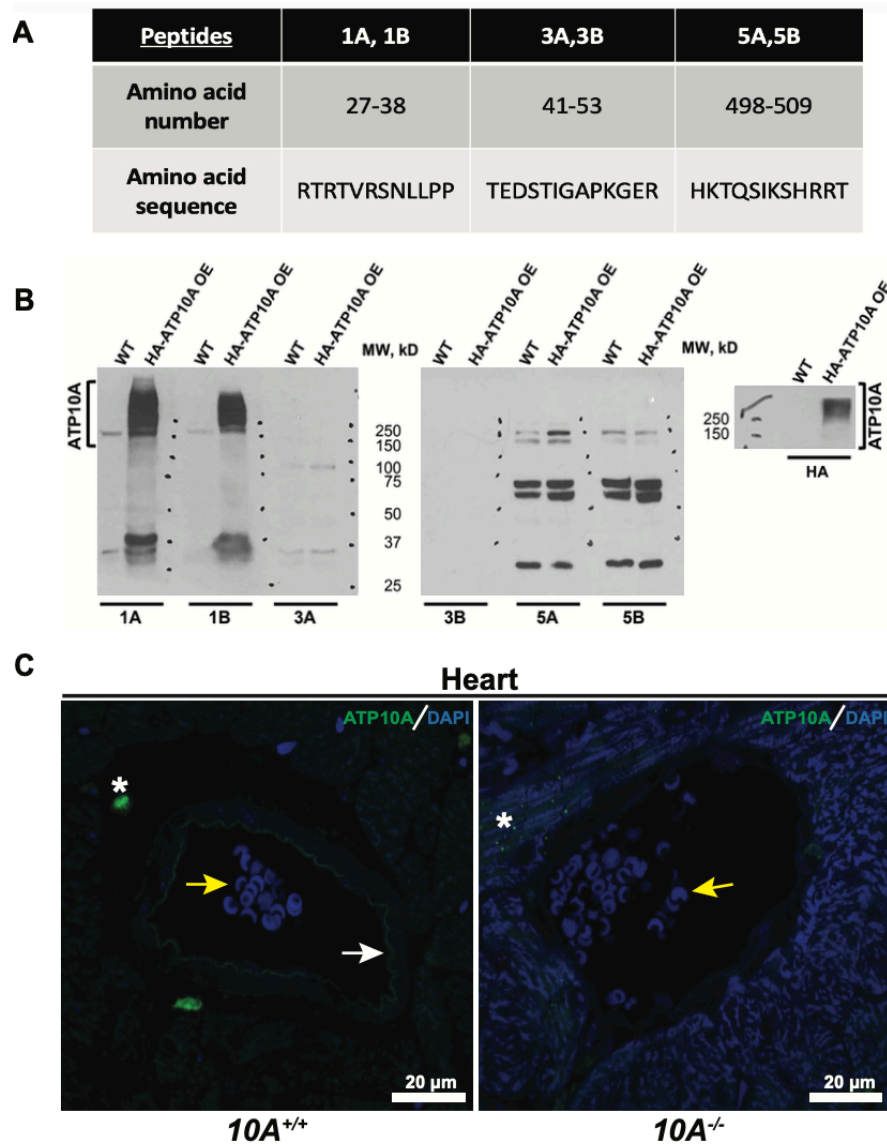


Supplemental Figure 4. Maternally inherited *Atp10A* deletion leads to smaller bodies and elevated fasting blood glucose in male mice on the 12th week on the HFD. (A) Weight gain of heterozygous males and females inheriting the KO allele maternally (Maternal Inheritance (Inh.), $10A^{+/-}$ dam X $10A^{+/+}$ sire) or paternally (Paternal Inh, $10A^{+/+}$ dam x $10A^{+/-}$ sire) over the course of 12 weeks on a HFD (60 kcal% fat, Ad lib feeding). P value by 2-way ANOVA with Sidak's multiple comparison. (**Males:** Maternal Inh. $10A^{+/-}$ n=7, Paternal Inh. $10A^{+/-}$ n=10; **Females:** Maternal Inh. $10A^{+/-}$ n=10, Paternal Inh. $10A^{+/-}$ n=6). (B) Lean and (C) fat body mass were normalized to body mass to calculate % Lean and % Fat mass. P value by unpaired t-test (**Male:** Maternal Inh. $10A^{+/-}$ n=7, Paternal Inh. $10A^{+/-}$ n=10; **Female:** Maternal Inh. $10A^{+/-}$ n=10, Paternal Inh. $10A^{+/-}$ n=6). (D) Body length of male mice was measured after CO₂ sacrifice. P value by unpaired t-test, *P=0.0490. (Maternal Inh. $10A^{+/-}$ n=4, Paternal Inh. $10A^{+/-}$ n=9). (E) Fasting blood glucose was measured after a 5 hour fast, via a glucometer. P value by unpaired t-test, **P=0.0081. (**Male:** Maternal Inh. $10A^{+/-}$ n=7, Paternal Inh. $10A^{+/-}$ n=10; **Female:** Maternal Inh. $10A^{+/-}$ n=10, Paternal Inh. $10A^{+/-}$ n=6). (F) Fasting plasma insulin was measured after a 5 hour fast, P value by unpaired t-test. (**Male:** Maternal Inh. $10A^{+/-}$ n=6, Paternal Inh. $10A^{+/-}$ n=10; **Female:** Maternal Inh. $10A^{+/-}$ n=5, Paternal Inh. $10A^{+/-}$ n=2).

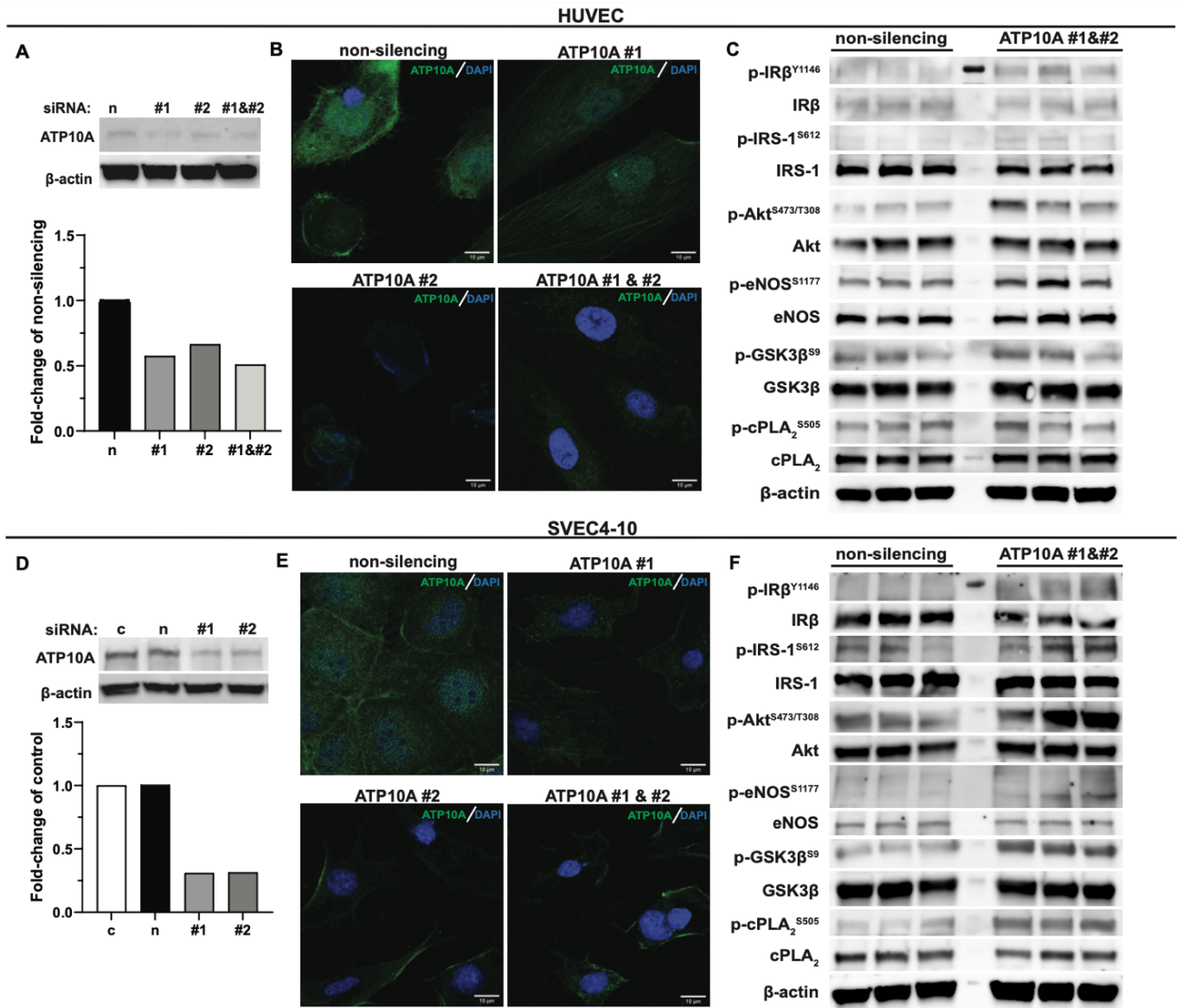


Supplemental Figure 5. *Atp10A* deletion causes changes to liver lipid metabolism in female mice after 12 weeks on a HFD.

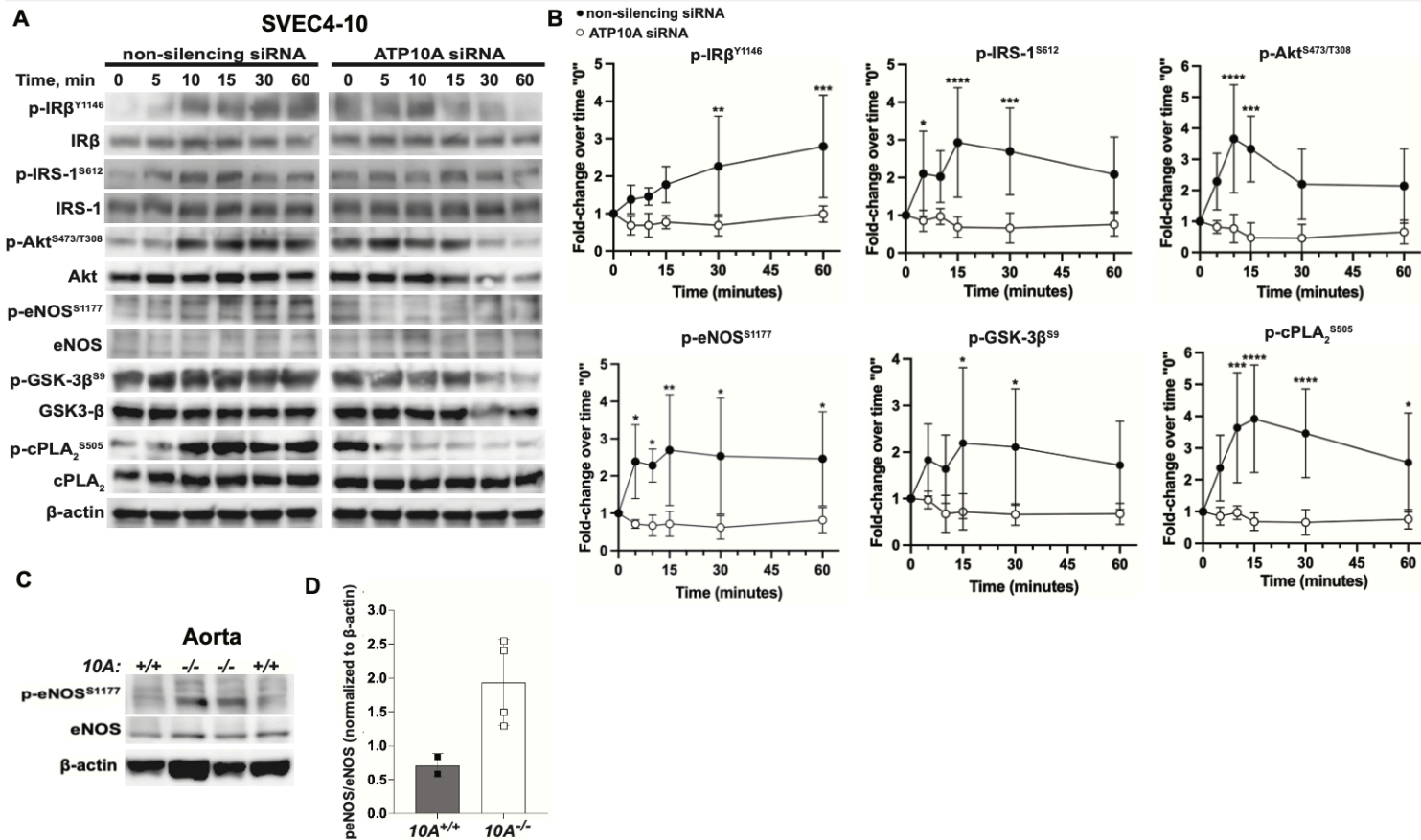
(A-G) FFAs, Total cholesterol, cholesterol esters, unesterified cholesterol, TGs, PLs, and ceramides were measured from flash frozen livers via gas chromatography. Livers were collected after a 5 hour fast or after a 5 hour fast followed by an OGTT. P value by unpaired t-test (FFA: $10A^{+/+}$ n=15, $10A^{-/-}$ n=7, *P=0.0201; Total cholesterol: $10A^{+/+}$ n=16, $10A^{-/-}$ n=7; Cholesterol esters: $10A^{+/+}$ n=6, $10A^{-/-}$ n=6; Unesterified cholesterol: $10A^{+/+}$ n=6, $10A^{-/-}$ n=6; TG: $10A^{+/+}$ n=14, $10A^{-/-}$ n=7; PLs: $10A^{+/+}$ n=16, $10A^{-/-}$ n=7; Ceramides: $10A^{+/+}$ n=16, $10A^{-/-}$ n=7). The saturation of liver (H) TG, PL, and (I) CE species was determined by gas chromatography. The fold change of unsaturated vs saturated TG, PL, CE is shown. P value by 2-way ANOVA with Sidak's multiple comparison, **TG**: **P=0.0025, ***P=0.0001, *P=0.0443, **PL**: *P=0.0297, *P=0.0281 ($10A^{+/+}$ n=16, $10A^{-/-}$ n=7). **CE**: P value by unpaired t-test, *P=0.0223 ($10A^{+/+}$ n=6, $10A^{-/-}$ n=6). (J) Representative images of livers stained with H+E (5x) or Oil Red O (63x). The coalescence of the Oil Red O stain is indicated by yellow asterisks (artifact) and the arrows point to neutral lipids stained by Oil Red O. Scale bars = 10 μ m. (K) Liver sections were scored using the Oil Red O Score described in the Materials and Methods. P value by nonparametric t-test (Mann-Whitney U), *P=0.0327 ($10A^{+/+}$ n=6, $10A^{-/-}$ n=10). (L) The diameters of Oil Red O positive lipid droplets (LD) in the liver sections with a score of 2 were measured using ImageJ. The red line in the graph represents the SEM. P value by unpaired t-test, ****P=<0.0001 ($10A^{+/+}$, 120 LDs measured, n=2, $10A^{-/-}$, 203 LDs measured, n=3).



Supplemental Figure 6. Validation of anti-ATP10A antibody: expression of ATP10A in HeLa cells with over-expressed HA-ATP10A and in mouse heart. (A) Table indicating the sequences of the ATP10A peptides that were injected into rabbits to produce antibodies. (B) Representative images of total cell lysates from HeLa cells with over-expressed HA-ATP10A (40 μ g total protein) that were subjected to Western immunoblot analyses for expression of ATP10A using antibody produced against ATP10A peptides (left and central panels) and HA (right panel). (C) Representative immunofluorescent images of hearts from female mice showing expression of ATP10A (green) and DAPI (blue). Scale bar = 20 μ m. The white asterisk indicates background staining, the white arrow indicates ATP10A positive staining, the yellow arrow indicates auto fluorescent red blood cells ($10A^{+/+}$ n=1, $10A^{-/-}$ n=1).



Supplemental Figure 7. ATP10A expression level regulates basal activity of insulin signaling pathways in endothelial cells. (A,D) Representative images and quantitative analysis of ATP10A from immunoblots of HUVEC (A) and SVEC4-10 (D) untreated (c), transfected with non-silencing siRNA or ATP10A siRNA constructs (#1, #2, #1). (B, E) Representative immunofluorescent images showing expression of ATP10A and DAPI in HUVEC (B) and SVEC4-10 (E) transfected with non-silencing siRNA (n) or ATP10A siRNA constructs (ATP10A#1, ATP10A#2, ATP10A#1). Scale bar=10 μ m. (C, F) Representative images of basal phosphorylated and total IR β , IRS-1, Akt, eNOS, GSK-3 β , and cPLA2 from immunoblots of HUVEC (C) and SVEC4-10 (F) transfected with non-silencing siRNA or ATP10A#1 siRNA constructs. Note that the lane between the non-silencing and ATP10A#1 treated lanes contains the molecular weight marker.



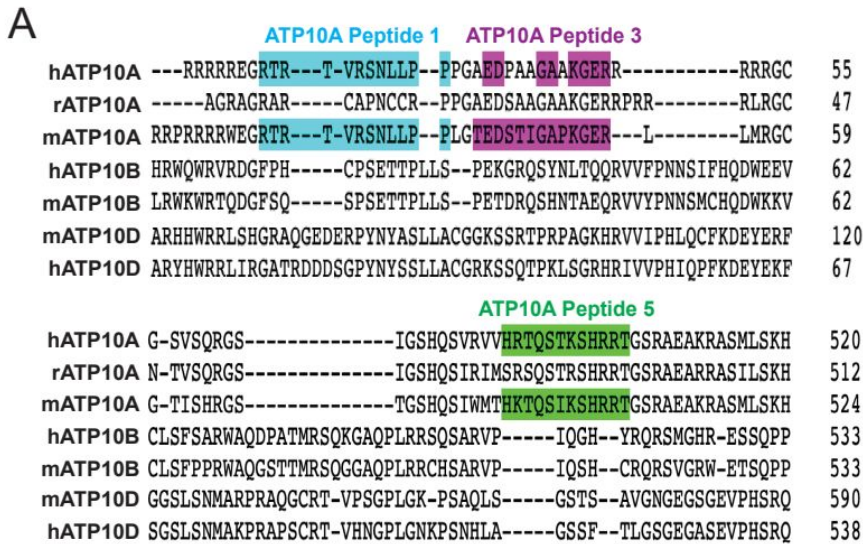
Supplemental Figure 8. ATP10A expression regulates insulin signaling pathway in SVEC4-10 cells and mouse aorta. (A)

Representative images and (B) quantitative analysis from immunoblots of phosphorylated and total IRβ, IRS-1, Akt, eNOS, GSK-3β, and cPLA₂ from SVEC4-10 cells transfected with non-silencing siRNA or ATP10A siRNA constructs and then treated with insulin for 0-60 mins. Mean measurements of 3-6 independent experiments are shown. *p≤0.05; **p≤0.005; ***p≤0.0005; ****p≤0.0001 between cells transfected with non-silencing or ATP10A siRNA by 2-way ANOVA with Sidak's multiple comparison test. (C) Representative blots and (D) quantitative analysis from immunoblots of phosphorylated and total eNOS from total aorta lysates from male mice (10A^{+/+} n=2, 10A^{-/-} n=4, statistics were not done due to low n). Mean measurements of 3-6 independent experiments are shown.

Supplemental Table 2

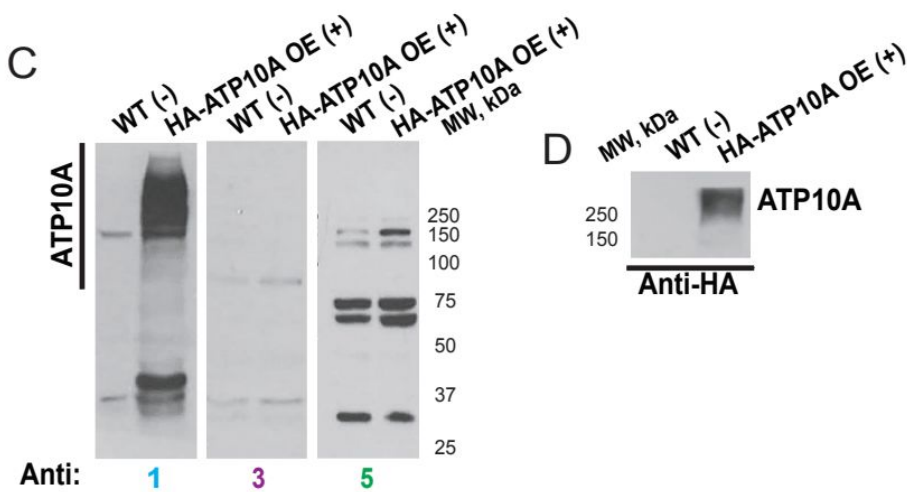
Pathway Kegg							
geneSet	description	size	overlap	expect	enrichmentRatio	pValue	FDR
g.jp/kegg-bin/show_pathway? mmu04972+13487+1878	Pancreatic secretion	103	5	0.809063	6.179985	0.0012387	0.393894
tp://www.kegg.jp/kegg-bin/show_pathway? mmu00730+	Thiamine metabolism	15	2	0.117825	16.974359	0.0059725	0.566786
g-bin/show_pathway? mmu04080+110902+11555+126	Neuroactive ligand-receptor interaction	289	7	2.270091	3.083577	0.0072516	0.566786
www.kegg.jp/kegg-bin/show_pathway? mmu05150+1351	Staphylococcus aureus infection	56	3	0.439879	6.820055	0.0095348	0.566786
www.kegg.jp/kegg-bin/show_pathway? mmu00561+2355	Glycerolipid metabolism	61	3	0.479154	6.261034	0.0120409	0.566786
tp://www.kegg.jp/kegg-bin/show_pathway? mmu04964+	Proximal tubule bicarbonate reclamation	22	2	0.17281	11.573427	0.0126837	0.566786
g.jp/kegg-bin/show_pathway? mmu04145+12845+1499	Phagosome	181	5	1.421752	3.516787	0.0135801	0.566786
p://www.kegg.jp/kegg-bin/show_pathway? mmu00790+1	Folate biosynthesis	26	2	0.20423	9.792899	0.0174897	0.566786
www.kegg.jp/kegg-bin/show_pathway? mmu04976+1384	Bile secretion	71	3	0.557704	5.379198	0.018093	0.566786
://www.kegg.jp/kegg-bin/show_pathway? mmu00650+2	Butanoate metabolism	27	2	0.212085	9.430199	0.0187942	0.566786
Significantly differential expressed genes in Pancreatic secretion							
Stable IDs	Feature gene name	baseMean	pvalue	padj	FoldChange		
ENSMUSG000000041193.15	Pla2g5	327.6929	0	0	21.6047666		
ENSMUSG00000007107.6	Atp1a4	258.5906	0	0.000001	4.8748642		
ENSMUSG000000071553.10	Cpa2	196.022	0	1.2E-06	3.7086972		
ENSMUSG000000020431.5	Adcy1	104.9107	0.00022	0.020501	0.3598145		
ENSMUSG00000001225.12	Sic26a3	29.11604	0.00024	0.021209	3.9715976		
Significantly differential expressed genes in Thiamine metabolism							
Stable IDs	Feature gene name	baseMean	pvalue	padj	FoldChange		
ENSMUSG000000026817.14	Ak1	1358.032	0.000292	0.023143	2.018769		
ENSMUSG000000028766.10	Alpl	237.0382	0.000518	0.034247	2.843773		
Significantly differential expressed genes in Neuroactive ligand-receptor interaction							
Stable IDs	Feature gene name	baseMean	pvalue	padj	FoldChange		
ENSMUSG000000056380.13	Gpr50	56.43242	1.47E-05	0.003859	571.2601672		
ENSMUSG000000040495.4	Chm4	14.01271	2.69E-05	0.005626	25.0694979		
ENSMUSG000000043017.9	Ptgir	400.5867	5.13E-05	0.008149	2.345606		
ENSMUSG000000044338.9	Aplnr	803.3216	0.00076	0.04297	2.7616563		
ENSMUSG000000021478.6	Drd1	52.58807	0.0009	0.048217	0.2768258		
ENSMUSG000000022041.10	Chma2	169.0929	0.000918	0.048854	2.1544465		
ENSMUSG000000045730.4	Adrb2	637.536	0.000944	0.049661	0.4256661		
Significantly differential expressed genes in Staphylococcus aureus infection							
Stable IDs	Feature gene name	baseMean	pvalue	padj	FoldChange		
ENSMUSG000000069441.3	Dsg1a	61.66929	4E-07	0.000233	5.596025		
ENSMUSG000000061928.6	Dsq1b	20.82076	2.61E-05	0.00562	19.302742		
ENSMUSG000000026580.16	Selp	210.5568	0.00073	0.04214	2.465355		
Significantly differential expressed genes in Glycerolipid metabolism							
Stable IDs	Feature gene name	baseMean	pvalue	padj	FoldChange		
ENSMUSG000000024768.5	Lipf	231.5996	0	0	16.1785033		
ENSMUSG000000012187.13	Mogat1	674.7383	0.000242	0.021213	0.4207309		
ENSMUSG000000020258.16	Glytk	130.4926	0.000281	0.022637	2.285067		
Significantly differential expressed genes in Proximal tubule bicarbonate reclamation							
Stable IDs	Feature gene name	baseMean	pvalue	padj	FoldChange		
ENSMUSG00000007107.6	Atp1a4	258.5906	0	0.000001	4.874864		
ENSMUSG000000010064.15	Sic38a3	708.3462	0.000883	0.048032	2.559326		
Significantly differential expressed genes in Phagosome							
Stable IDs	Feature gene name	baseMean	pvalue	padj	FoldChange		
ENSMUSG000000031849.9	Comp	138.805	0.000068	0.009699	2.5941413		
ENSMUSG000000020695.14	Mrc2	2506.892	0.00012	0.013829	2.7056454		
ENSMUSG000000067201.11	H2-M9	240.8505	0.000181	0.017773	0.4219105		
ENSMUSG000000028238.6	Atp6v0d2	208.2672	0.000195	0.018484	4.0684324		
ENSMUSG000000026390.7	Marco	155.7569	0.000613	0.037129	2.3528541		
Significantly differential expressed genes in Folate biosynthesis							
Stable IDs	Feature gene name	baseMean	pvalue	padj	FoldChange		
ENSMUSG000000006764.8	Tph2	959.1933	1E-07	5.16E-05	22.049261		
ENSMUSG000000028766.10	Alpl	237.0382	0.000518	0.034247	2.843773		
Significantly differential expressed genes in Bile secretion							
Stable IDs	Feature gene name	baseMean	pvalue	padj	FoldChange		
ENSMUSG00000007107.6	Atp1a4	258.5906	0	0.000001	4.8748642		
ENSMUSG000000038776.13	Ephx1	4746.016	9.4E-06	0.002761	2.0548225		
Significantly differential expressed genes in Butanoate metabolism							
Stable IDs	Feature gene name	baseMean	pvalue	padj	FoldChange		
ENSMUSG000000057880.12	Abat	3895.093	1E-07	4.84E-05	2.4992921		
ENSMUSG000000030972.6	Acsm5	1971.103	0.000482	0.032604	0.4485598		

Appendix C: Supplemental Figures for Chapter 3



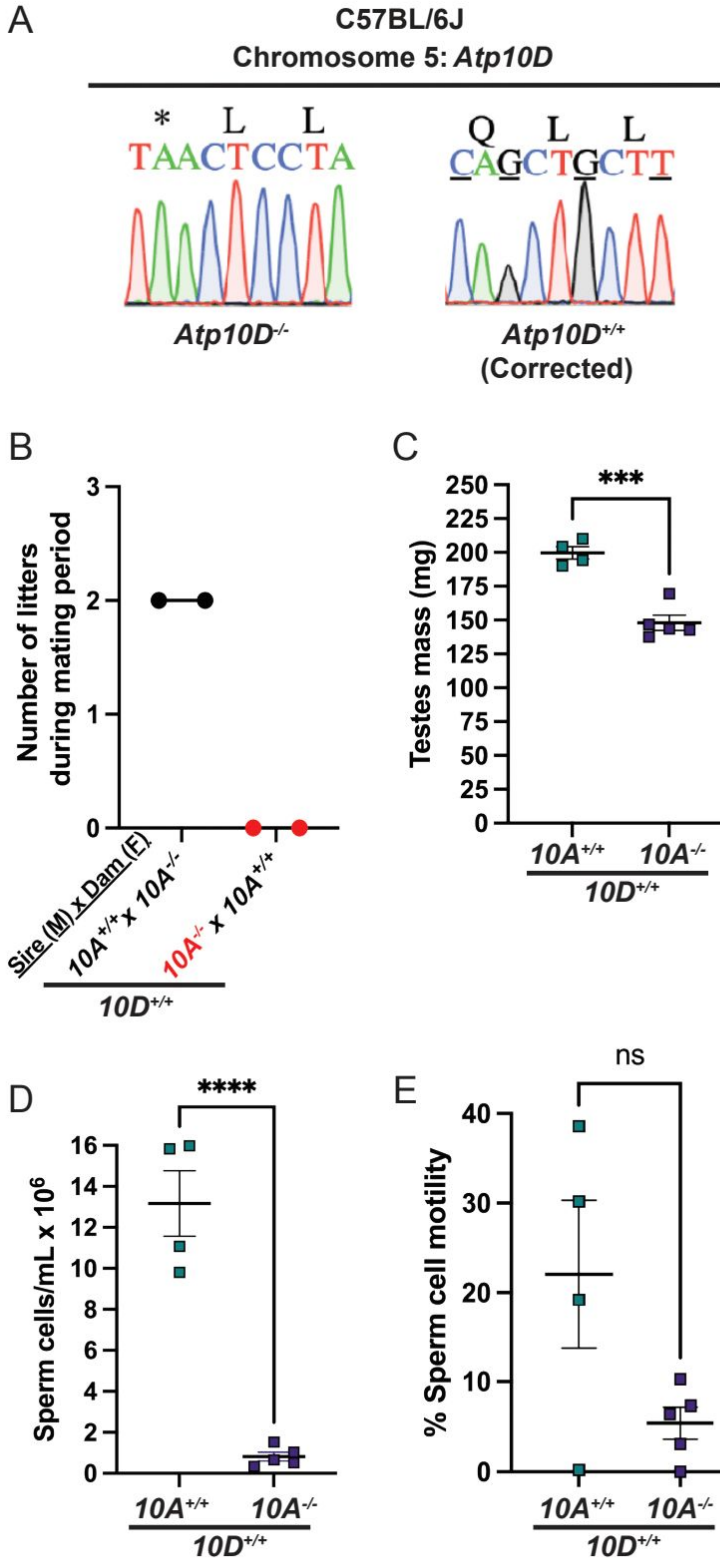
B

ATP10A Peptides	1	3	5
Amino acid number	27-38	41-53	498-509
Amino acid sequence	RTRTVRSNLLPP	TEDSTIGAPKGER	HKTQSIKSHRRT



Supplemental Figure 9. Validation of ATP10A antibody: expression of ATP10A in HeLa cells with over-expressed HA-ATP10A. (A) Alignments of human (h), rat (r), and mouse (m) ATP10A, ATP10B, and ATP10D created using Clustal Omega with the ATP10A peptides that were chosen for antibody production highlighted. These peptides were chosen based on conservation between mouse and human ATP10A orthologs and lack of conservation with ATP10B and ATP10D paralogs. (B) Table indicating the sequences of the ATP10A peptides that were injected into rabbits to produce antibodies, peptides are color-coded according to the colors of the peptides in (A). (C,D)

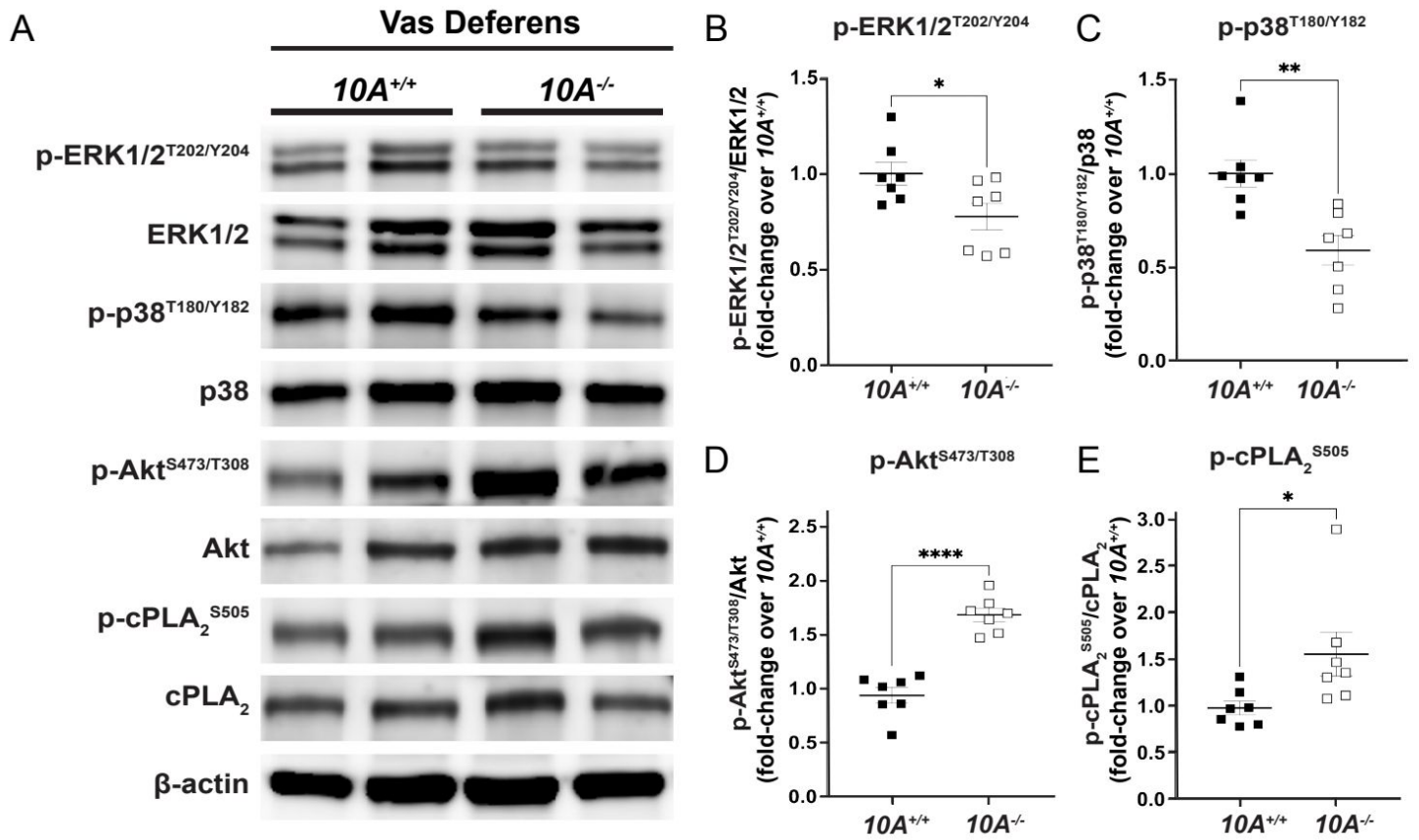
Representative images of total cell lysates from HeLa cells with (+) or without (-) over-expressed (OE) human HA-ATP10A (40 μ g total protein) that were subjected to Western immunoblot analyses for expression of ATP10A using antibody produced against (C) ATP10A peptides or (D) HA. Peptides are color-coded according to the colors of the peptides in (A).



Supplemental Figure 10. Reintroduction of *Atp10D*

does not rescue infertility, smaller testes, or low sperm cell count in *Atp10A* deficient mice. (A)

Sequencing was performed to validate the genotype of the *Atp10D* mice. Briefly, CRISPR was used to replace the premature stop codon found in the *Atp10D* sequence in C57BL/6J mice (*, *Atp10D*^{-/-} mice) to glutamine that is found in other mouse strains (Q, *Atp10D*^{+/+} (corrected) mice). The underlined nucleotides indicate CRISPR-mediated changes to the DNA sequence, for the purposes of changing the amino acid or introducing restriction digest cut sites for genotyping purposes. (B) Mating studies were done by housing pairs of mice together (without separating) for approximately 9 weeks and recording the number of litters produced. (See supplemental table 1 for more details). (C) The wet mass of testes was measured in 14-21-week-old mice, ***P=0.0002. (D) Sperm cells collected from the caput epididymis were counted using a hemocytometer, ****P<0.0001. (E) % Sperm cell motility was measured using the equation: motile sperm/total sperm counted. P-value by unpaired t-test.



Supplemental Figure 11. *Atp10A* deficiency results in changes to signaling in the vas

deferens. Total tissue lysates from vas deferens from the *10A*^{+/+} and *10A*^{-/-} mice were subjected

to Western immunoblot analysis. (A) Representative blots for total and/or phosphorylated

ERK1/2, p38, Akt, and cPLA₂. (B-E) The phosphoproteins were normalized to their respective

total protein, β-actin levels, and total protein from *10A*^{+/+} samples. Mean measurements of 4-6

independent experiments are shown. P value by unpaired t-test. (*10A*^{+/+} n=7, *10A*^{-/-} n=7, B:

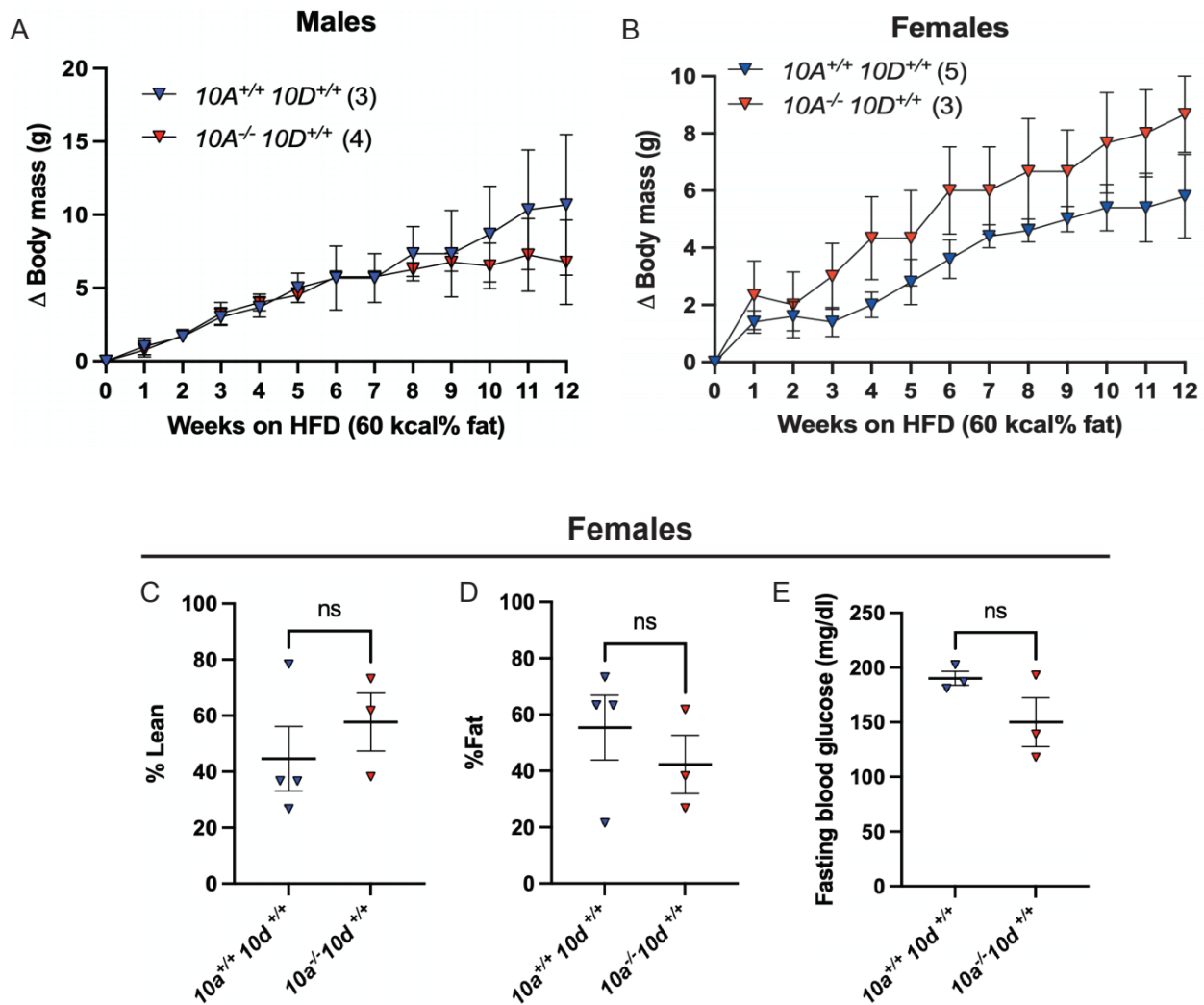
*P=0.0313, C: **P= 0.0024, D: ****P<0.0001, E: *P=0.0376).

Appendix D. Supplemental Table for Chapter 3

Supplemental Table 3

Figure 9 (all 10D -/-)	10A+/+ (WT)	10A-/- (KO)	
(A) Vas deferens WB			
n	1	1	
Body mass at sacrifice (g)	did not measure	did not measure	
Age at sacrifice (weeks)	7	8	
Outliers removed by ROUT analysis	N/A	N/A	
(B) Testes WB			
n	1	1	
Body mass at sacrifice (g)	23	24	
Age at sacrifice (weeks)	8	8	
Outliers removed by ROUT analysis	N/A	N/A	
(C) Vas deferens IF			
n	1	2	
Avg. body mass at sacrifice (g)	26	28.5	
Avg. age at sacrifice (weeks)	11	15	
Outliers removed by ROUT analysis	N/A	N/A	
(D) Testes IF			
n	2	2	
Avg. body mass at sacrifice (g)	27.5	28.5	
Avg. age at sacrifice (weeks)	17	17	
Outliers removed by ROUT analysis	N/A	N/A	
(E) Matino studies	Red text= Sire is 10A -/-		
Order: Sire (M) x Dam (F)	Average time parents housed together (weeks)		Average pups per litter
10A +/- x 10A +/- (1 pair)	20.9		6
10A +/- x 10A -/- (2 pairs)	6.6		5
10A -/- x 10A +/- (3 pairs)	9.8		N/A
10A +/- x 10A -/- (1 pair)	19.6		6
10A -/- x 10A +/- (3 pairs)	15.9		8.5
10A +/- x 10A +/- (5 pairs)	12		7.1
10A -/- x 10A -/- (3 pairs)	10.9		N/A
Figure 10 (all 10D -/-)	10A+/+ (WT)	10A+/- (Het)	10A-/- (KO)
(A) Testes mass (mg), (B) Sperm cells/mL, (C) % sperm cell motility			
n	9	2	14
Avg. body mass at sacrifice (g)	26.8	25	26.7
Avg. age at sacrifice (weeks)	13.8	10.5	12
Outliers removed by ROUT analysis	0	0	0
Figure 11 (all 10D -/-)	10A+/+ (WT)	10A-/- (KO)	
(C) Seminiferous tubule scoring			
n	6	12	
Avg. body mass at sacrifice (g)	26	26.8	
Avg. age at sacrifice (weeks)	12.2	11.6	
Outliers removed by ROUT analysis	N/A	N/A	
Figure 12 (all 10D -/-)	10A+/+ (WT)	10A-/- (KO)	
(B) Plasma T			
n	16	18	
Avg. body mass at sacrifice (g)	33.5	33.8	
Avg. age at sacrifice (weeks)	14.1	15.4	
Outliers removed by ROUT analysis	3	1	
(C) LH	10A+/+ (WT)	10A-/- (KO)	
n	11	17	
Avg. body mass at sacrifice (g)	26	27	
Avg. age at sacrifice (weeks)	11.5	11.1	
Outliers removed by ROUT analysis	0	1	
(D) FSH	10A+/+ (WT)	10A-/- (KO)	
n	11	18	
Avg. body mass at sacrifice (g)	26	27	
Avg. age at sacrifice (weeks)	11.5	11.1	
Outliers removed by ROUT analysis	1	0	
(E) Inhibin B	10A+/+ (WT)	10A-/- (KO)	
n	11	15	
Avg. body mass at sacrifice (g)	26	27.3	
Avg. age at sacrifice (weeks)	11.5	11.1	
Outliers removed by ROUT analysis	0	0	
Figure 13 (Testes, all 10D -/-)	10A+/+ (WT)	10A-/- (KO)	
n	6-8	6-8	
Avg. body mass at sacrifice (g)	27.4	28	
Avg. age at sacrifice (weeks)	14.5	14.1	
Outliers removed by ROUT analysis	0	LHR: 1, pAkt: 1	
Supplemental Figure 10 (All 10D +/-)			
(A) Mating studies	Red text= Sire is 10A -/-		
Order: Sire (M) x Dam (F)	Average time parents housed together (weeks)		Average pups per litter
10A +/- x 10A -/- (2 pairs)	8.9		5.8
10A -/- x 10A +/- (2 pairs)	9.1		N/A
(B,C,D) Testes mass, Sperm cells/mL, sperm cell motility	10A+/+, 10D +/-	10A-/-, 10D +/-	
n	4	5	
Avg. body mass at sacrifice (g)	29.8	29.4	
Avg. age at sacrifice (weeks)	18.5	17.8	
Outliers removed by ROUT analysis	0	0	
Supplemental Figure 11 (Vas deferens, all 10D -/-)	10A+/+ (WT)	10A-/- (KO)	
n	4-7	4-7	
Avg. body mass at sacrifice (g)	15.4	15	
Avg. age at sacrifice (weeks)	27.9	28.6	
Outliers removed by ROUT analysis	0	0	

Appendix E: Supplemental Figure for Chapter 5



Supplemental Figure 12. Metabolic parameters in the 10A Deficient, 10D Correction mouse model (A, B) Weight gain of (A) male and (B) female $10A^{+/+}10D^{+/+}$ and $10A^{-/-}10D^{+/+}$ over the course of 12 weeks on a HFD (60 kcal% fat, Ad lib feeding). P value by 2-way ANOVA with Sidak's multiple comparison. (C) Lean and (D) fat body mass were normalized to the sum of lean mass and fat mass to calculate % Lean and % Fat mass ($10A^{+/+}10D^{+/+}$ n=4, $10A^{+/+}10D^{-/-}$ n=3). (E) Fasting blood glucose was measured after a 5 hour fast, via a glucometer ($10A^{+/+}10D^{+/+}$ n=3, $10A^{+/+}10D^{-/-}$ n=3). (C-E) P value by unpaired t-test. Average body mass (g) at end of HFD: Males: $10A^{+/+}10D^{+/+}$: 36 g, $10A^{-/-}10D^{+/+}$: 33.3 g, Females: $10A^{+/+}10D^{+/+}$: 24.5 g, $10A^{+/+}10D^{+/+}$: 29 g

Appendix F: Supplemental Tables for Chapter 6

Supplemental Table 4

Target	Primer sequence (5' to 3')	
	F	R
Pla2g5	TGG GGT TCT GAG ATG TCA CT	TCT TCC TCC TGT CTT GTT GC
SCD1	CTGTACGGGATCATACTGGTTC	GCCGTGCCTTGTAAGTTCTG
LPL	TGG CAT AAG TCA GGT CCA TT	GAG CCA TGT CTT CAA CTG CT
Pla2g6	ATC CAC ACA GCC ATG AAG TT	TGT TCC CTG AGG AGC TAG TG
FAS	GCGATGAAGAGCATGGTTTAG	GGCTCAAGGGTCCATGTT
DGAT1	CCA GGT GGT GTC TCT GTT TC	AGA TGA TTG TGG CCA GGT TA
DGAT2	CTGTGCTCTACTTCACCTGGCT	CTGGATGGGAAAGTAGTCTCGG
PPARa	GCCTGTCTGTCCGGATGT	GGCTTCGTGGATTCTCTTG
PPARg	GCCCTTTGGTGACTTTATGGA	GCAGCAGGTTGTCTTGATG
LXRa	CCGACAGAGCTTCGTC	CCCACAGACACTGCACAG
Srebpc1	CGCGGAAGCTGTCGGGGTAG	AAATGTGCAATCCATGGCTCCGTGGTC
CGI58	GGT TAA GTC TAG TGC AGC	AAG CTG TCT CAC CAC TTG
	cca ggg aga tgc tct gtg ttc	acc cag aca gcg ctc ttt gat
Cyp7a1	AGCAACTAAACAACCTGCCAGTAC TA	GTCCGGATATTCGAAGGATGCA
Cyp8b1	GCCTTCAAGTATGATCGGTTCCCT	GATCTTCTTGCCCGACTTGTAGA
Hprt1 (housekeeping gene)	GCTGACCTGCTGGATTACAT	TTGGGGCTGTACTGCTTAAC

Supplemental Table 5

Visceral Fat, Females, 12 weeks after HFD feeding									
qPCR results					RNA sequencing results (10A+/+ n=3, 10A-/- n=3)				
Target	Sample size (n)		p-value (unpaired t-test of Relative mRNA level (2 ^{-ΔΔCq}))	Significant (Y or N)	Target	log2FoldChange	p-value	Significant (Y or N)	
	10A+/+	10A-/-							
Pla2g5	2	4	0.2526	N	Pla2g5	4.433277742	2.03E-17	Y	
SCD1	2	4	0.8275	N	SCD1	0.355620653	0.242335	N	
LPL	3	4	0.7678	N	LPL	0.294114678	0.123495	N	
Pla2g6	3	4	0.8143	N	Pla2g6	0.234601663	0.309316	N	
FAS	3	4	0.9077	N	FAS	-0.099676899	0.588441	N	
DGAT1	3	4	0.6299	N	DGAT1	0.017153228	0.933198	N	
DGAT2	3	4	0.6174	N	DGAT2	0.439107779	N/A	N/A	
PPARa	2	4	0.3831	N	PPARa	not found in data	---	---	
PPARg	3	4	0.3658	N	PPARg	not found in data	---	---	
LXRa	3	4	0.824	N	LXRa	not found in data	---	---	
SREBP-1C	2	4	0.824	N	Srebpc1	not found in data	---	---	
CGI58	2	4	0.824	N	CGI58	not found in data	---	---	
Liver qPCR Results, Females, 12 weeks after HFD feeding									
Target	Sample size (n)		p-value (unpaired t-test of Relative mRNA level (2 ^{-ΔΔCq}))	Significant (Y or N)					
	10A+/+	10A-/-							
PPARg	4	4	0.0474	Y *					
FAS	4	3	0.0067	Y **					
SCD1	4	5	0.4074	N					
Cyp7a1	4	4	0.7601	N					
Cyp8b1	4	4	0.1477	N					
LPL	4	3	0.2685	N					