SUPPLEMENT (Krupenko et al)

1. Supplementary figures S1-S12

2. Supplementary Tables S1-S4

Supplementary Data Files 1-6 (separate Excel files)

Supplementary Data Files 1: liver and pancreas metabolomic data (three genotypes)

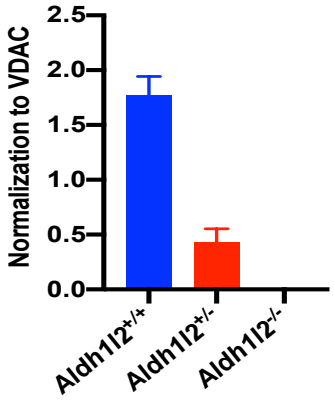
Supplementary Data File 2: VIP values (OPLSDA) for metabolomic data, liver and pancreas (three genotypes)

Supplementary Data Files 3: liver and plasma metabolomic data (wild type and Aldh1l2 KO, second experiment)

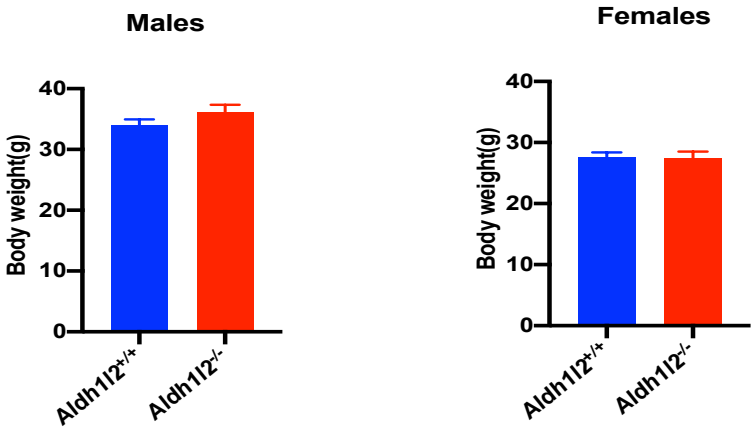
Supplementary Data Files 4: VIP values (OPLSDA) for metabolomic data, liver and plasma (wild type and Aldh1l2 KO, second experiment)

Supplementary Data File 5: list of metabolites overlapping between liver and plasma

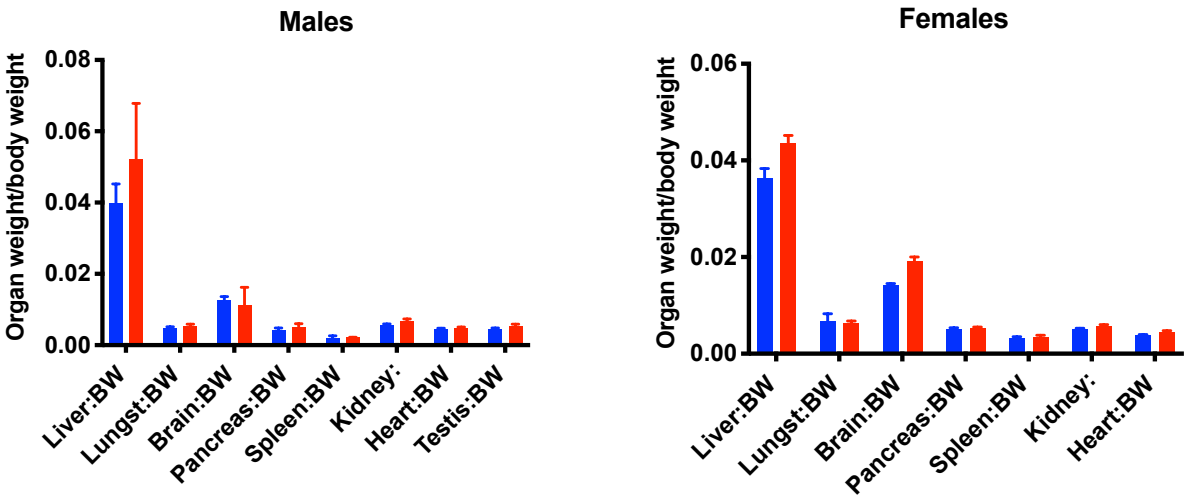
Supplementary Data File 6: NanoString data and VIP values (OPLSDA analysis)



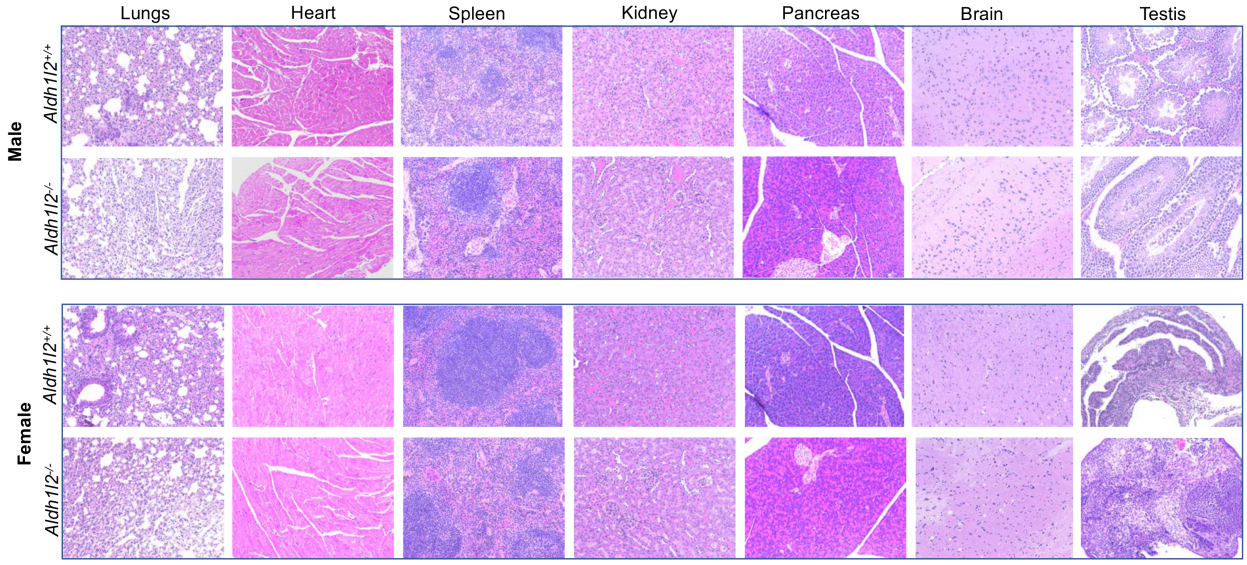
Supplementary Fig. S1. Intensity of ALDH1L2 bands (mean ± SE) relative to VDAC (from Fig. 1d; calculated using Image J).



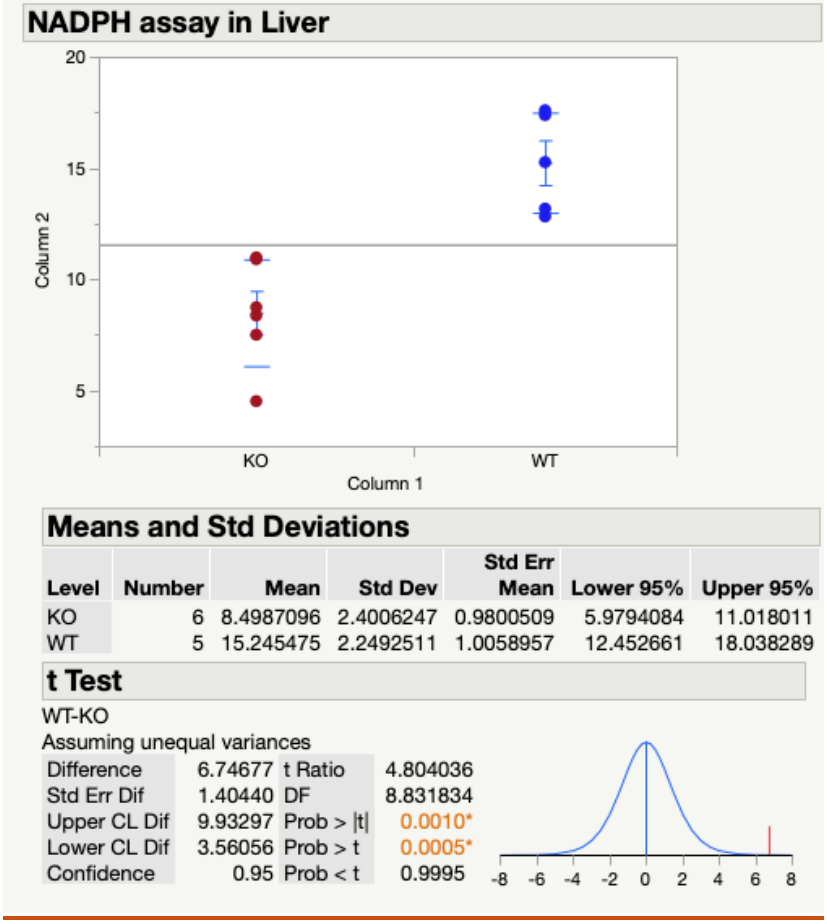
Supplementary Fig. S2. Body weights of *Aldh1l2*$^{+/+}$ and *Aldh1l2*$^{-/-}$ male and female mice at the age of 6 months.



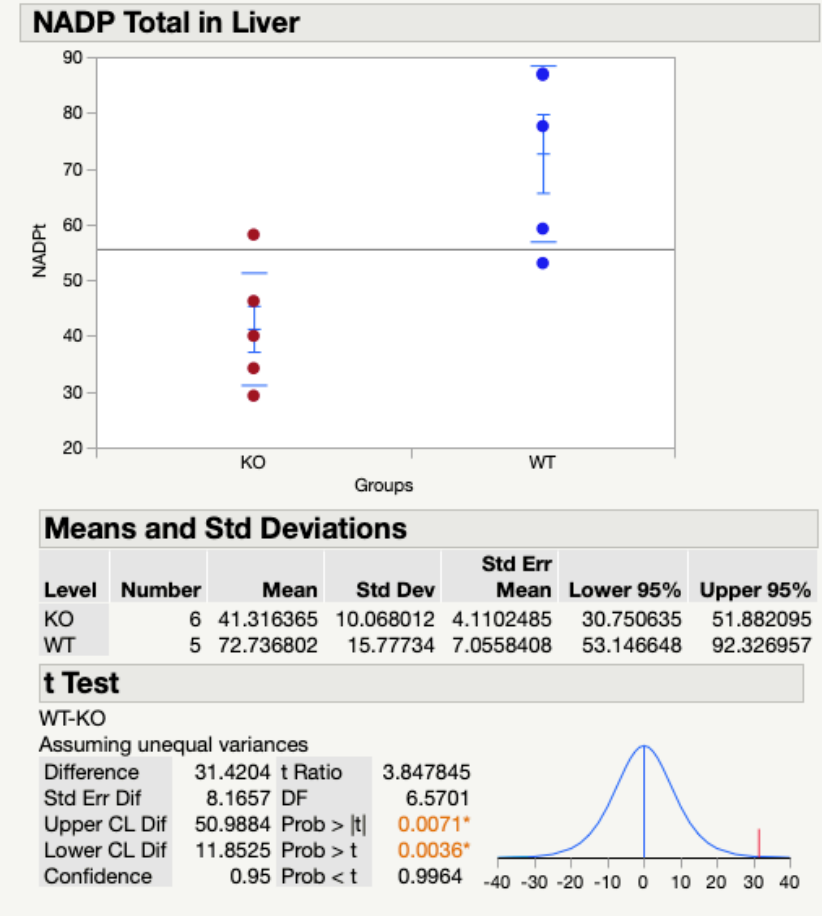
Supplementary Fig. S3. Organ to body weight ratio of *Aldh1l2*$^{+/+}$ and *Aldh1l2*$^{-/-}$ male and female mice at the age of 6 months.



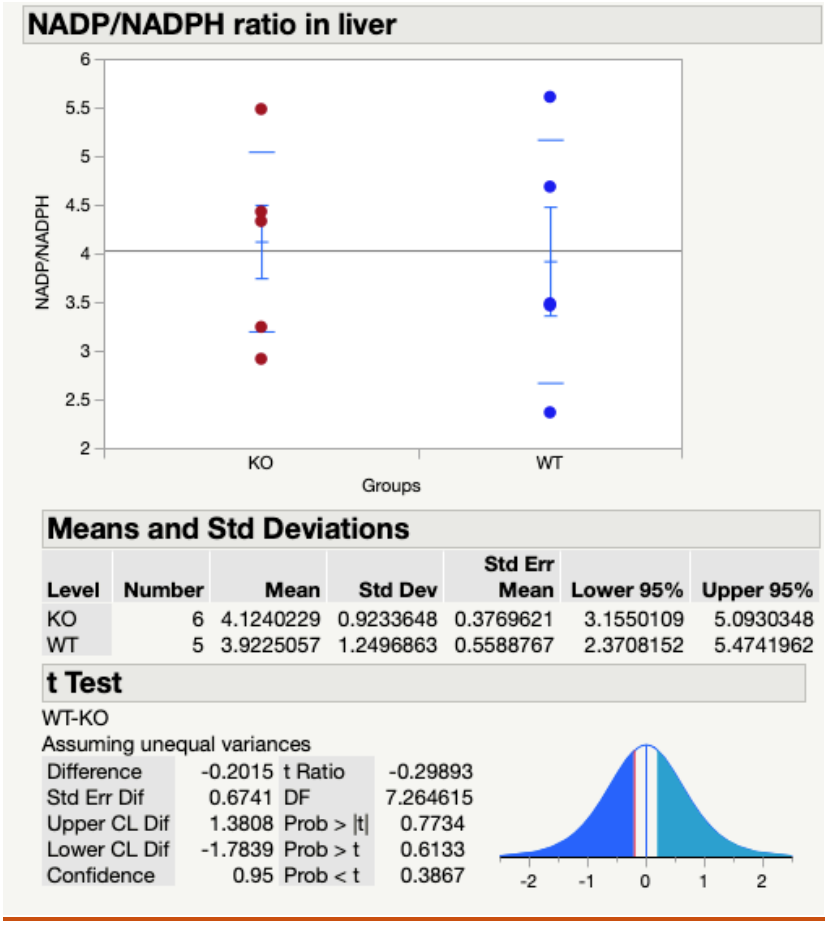
Supplementary Fig. S4. H&E staining of tissue sections from *Aldh1l2*$^{+/+}$ and *Aldh1l2*$^{-/-}$ mice.



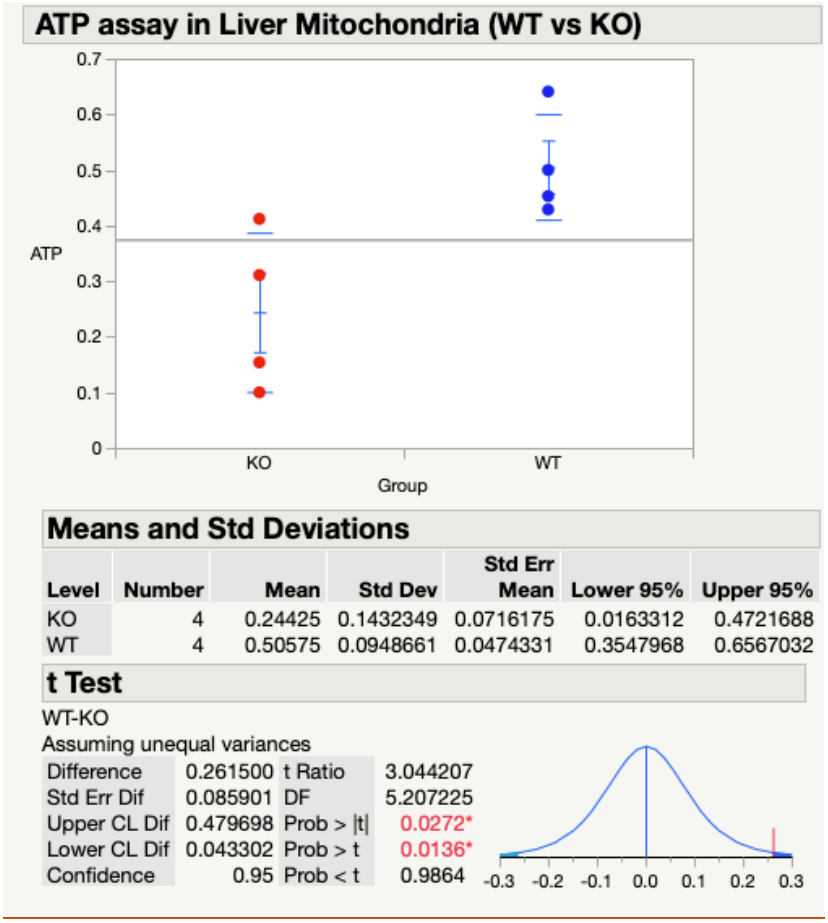
Supplementary Fig. S5. NADPH in liver of *Aldh1l2*$^{+/+}$(WT) and *Aldh1l2*$^{-/-}$(KO) mice measured by a fluorescence kit. 5-6 samples (mice) were analyzed for each genotype. Each sample was measured 4 times (technical replicate). Averages of technical replicates were used to calculate mean ± SE.



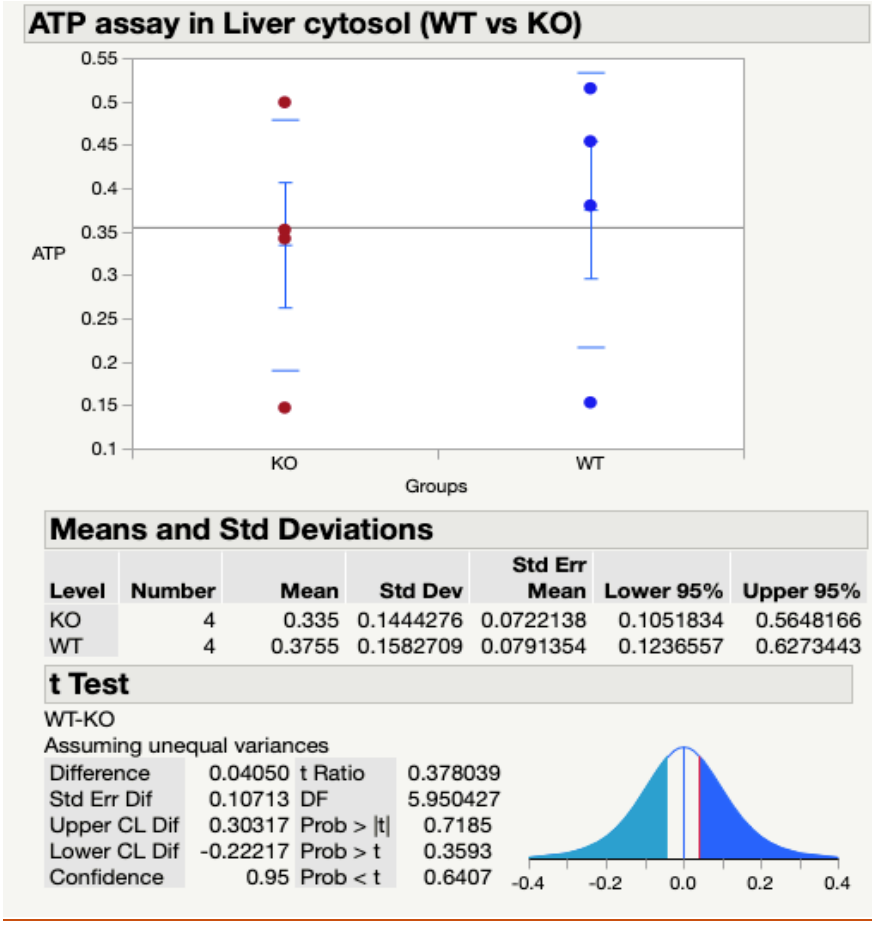
Supplementary Fig. S6. NADP Total (NADPt) in liver of *Aldh1l2*$^{+/+}$(WT) and *Aldh1l2*$^{-/-}$(KO) mice measured by a fluorescence kit. 5-6 samples (mice) were analyzed for each genotype. Each sample was measured 4 times (technical replicate). Averages of technical replicates were used to calculate mean ± SE



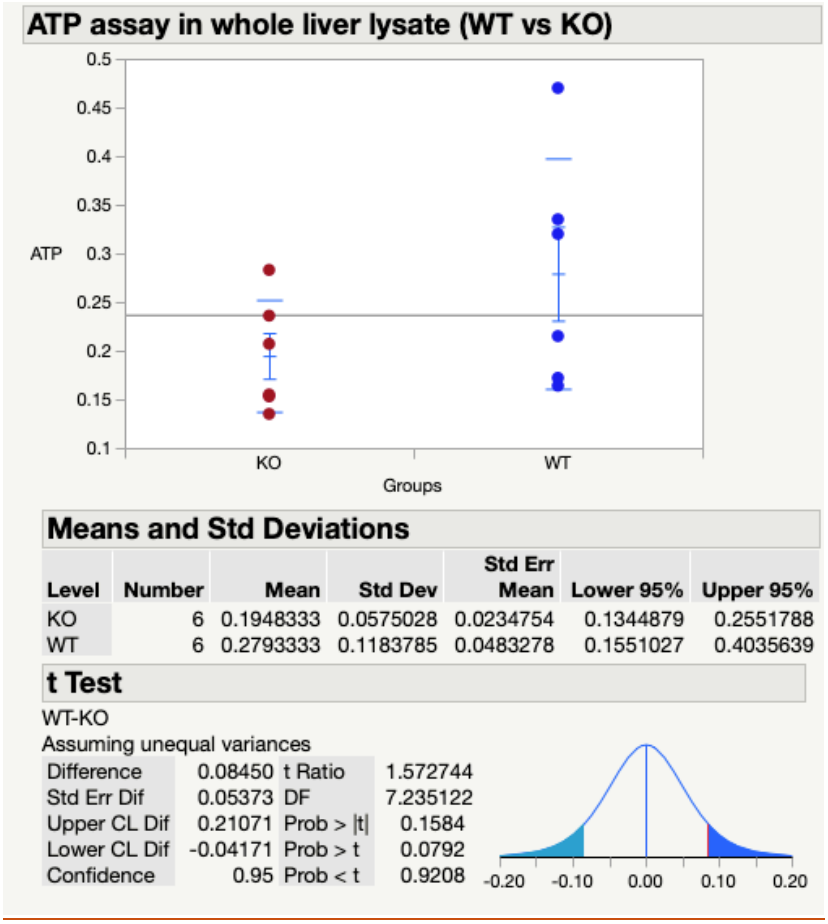
Supplementary Fig. S7. NADP/NADPH in liver of *Aldh1l2*$^{+/+}$(WT) and *Aldh1l2*$^{-/-}$(KO) mice measured by a fluorescence kit. 5-6 samples (mice) were analyzed for each genotype. Each sample was measured 4 times (technical replicate). Averages of technical replicates were used to calculate mean ± SE.



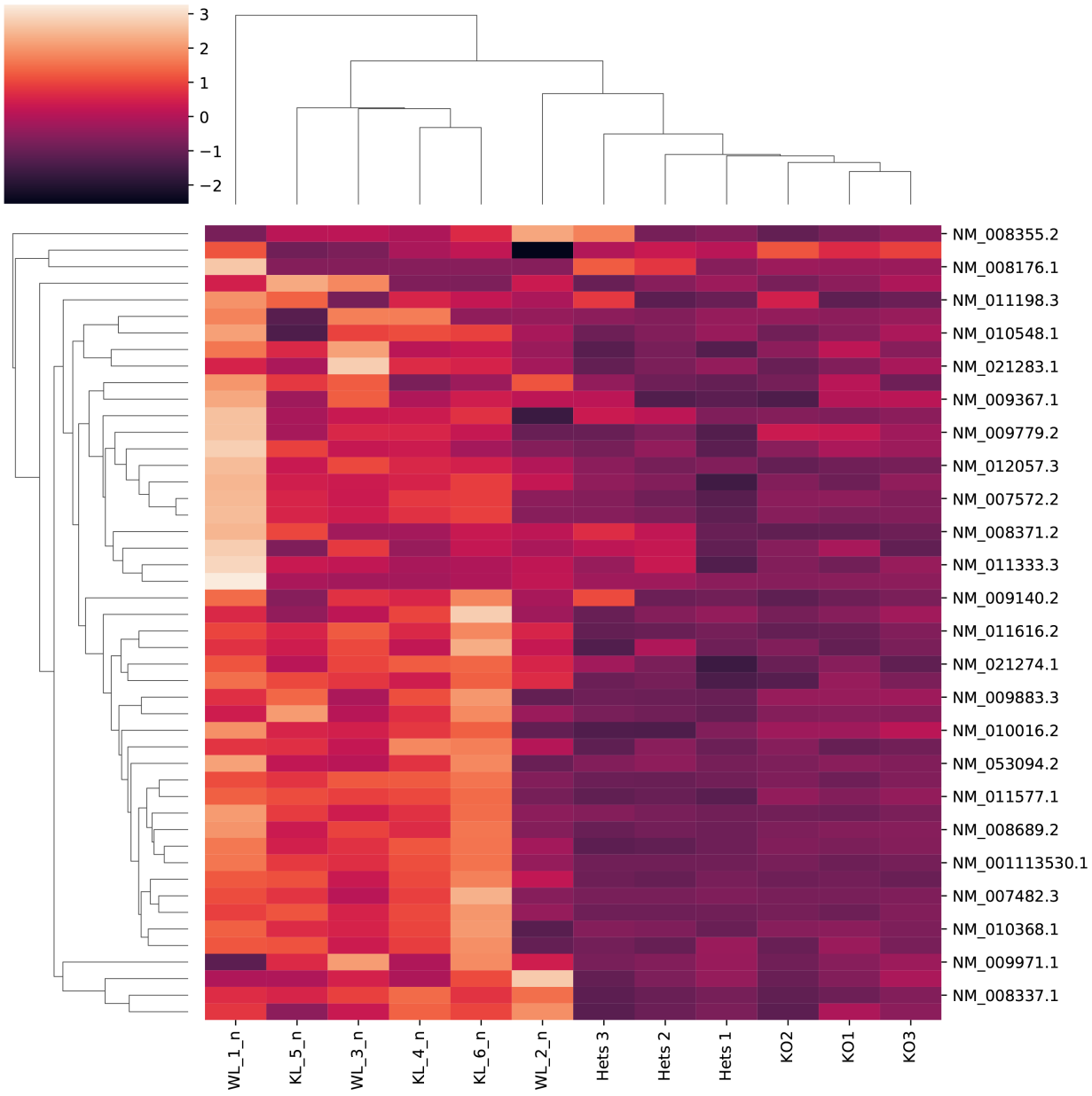
Supplementary Fig S8. Levels of ATP in mitochondria isolated from liver of *Aldh1l2*$^{+/+}$(WT) and *Aldh1l2*$^{-/-}$(KO) mice measured by a colorimetric assay. Four samples (mice) were analyzed for each genotype. Each sample was measured 4 times (technical replicate). Averages of technical replicates were used to calculate mean ± SE.



Supplementary Fig S9. Levels of ATP in cytosolic fraction isolated from liver of *Aldh1l2*$^{+/+}$(WT) and *Aldh1l2*$^{-/-}$(KO) mice measured by a colorimetric assay. Four samples (mice) were analyzed for each genotype. Each sample was measured 4 times (technical replicate). Averages of technical replicates were used to calculate mean ± SE.

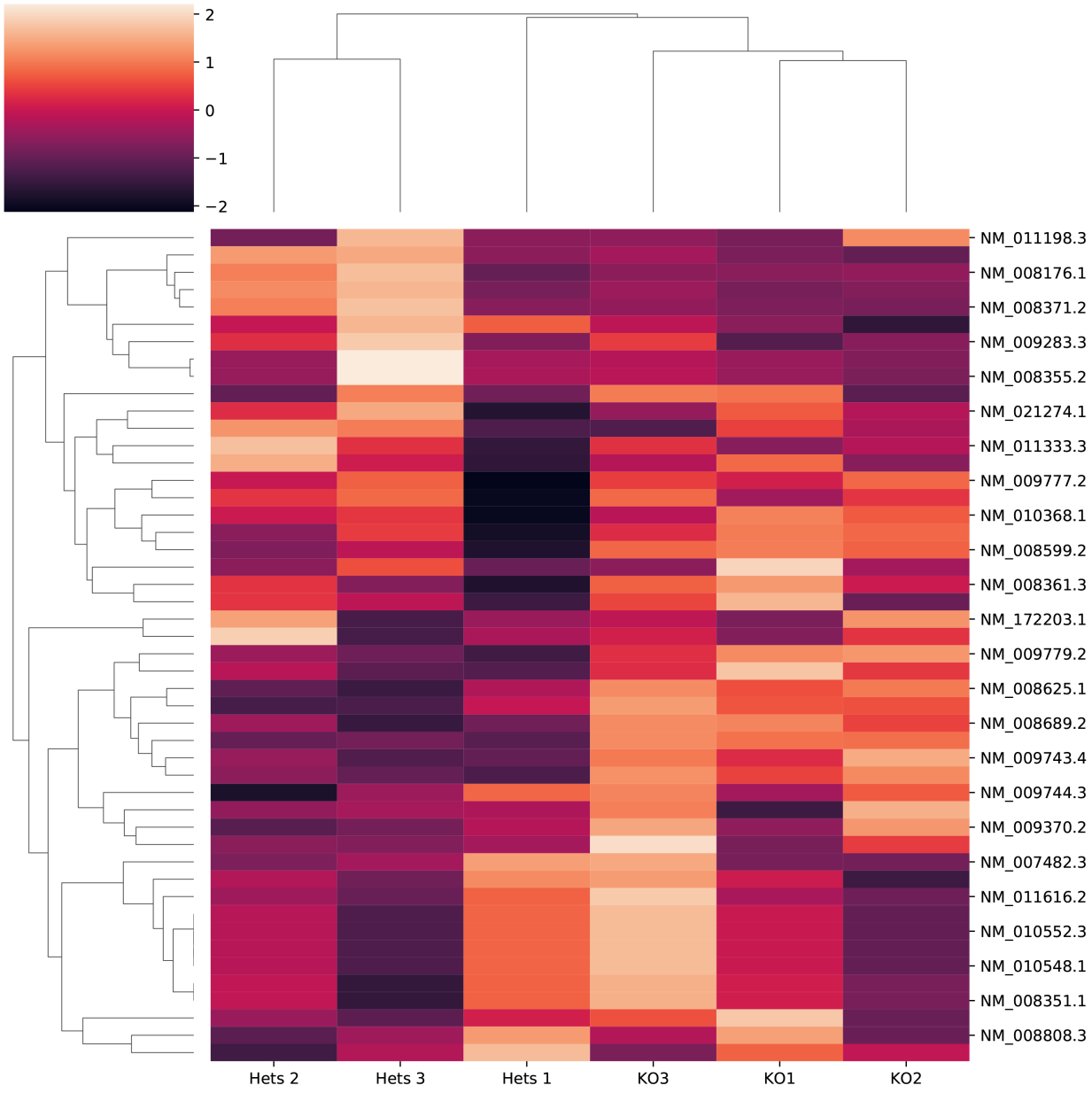


Supplementary Fig S10. Levels of ATP in whole liver lysate isolated from *Aldh1l2*$^{+/+}$(WT) and *Aldh1l2*$^{-/-}$(KO) mice measured by a colorimetric assay. Six samples (mice) were analyzed for each genotype. Each sample was measured 4 times (technical replicate). Averages of technical replicates were used to calculate mean ± SE.



Supplementary Fig S11. Heat map representation of the NanoString data for the four genotypes.

Each protein was standardized so that it has mean 0 and standard deviation 1.



Supplementary Fig S12. Heat map representation of the NanoString data for the *Aldh1l*$^{+/-}$ and *Aldh1l2*$^{-/-}$ genotypes (based on the entire panel of 242 genes).

Supplementary Table S1. Primers used for genotyping.

Primers	Sequence	Amplicon size (bp)
WTf	5′- AATTGGTGGTTCTCTCAAGTCTG -3′	338 bp
WTr	5′- GCACCCATAAAGGGGCTCAAG -3′	
RAF5-L2	5′- CACACCTCCCCCTGAACCTGAAA -3′	598 bp
TTR1-L2	5′- GACATATACTGACCTCTGAGGGTGGC-3′	

Supplementary Table S2. List of primary antibodies used for Western blot assays.

Target	Antibody	Source	Cat# or reference*	Dilution
ALDH1L1	Rabbit polyclonal	In-house	[1]	1:10,000
ALDH1L2	Rabbit polyclonal	In-house	[2]	1:10,000
Hsp60	Rabbit polyclonal	Abcam	ab46798	1:20,000
VDAC	Rabbit monoclonal	Abcam	ab154856	1:1,000
Actin	Mouse monoclonal	Abcam	ab8226	1:10,000

1 Krupenko, S. A. and Oleinik, N. V. (2002) 10-formyltetrahydrofolate dehydrogenase, one of the major folate enzymes, is down-regulated in tumor tissues and possesses suppressor effects on cancer cells. Cell Growth Differ. **13**, 227-236

2 Krupenko, N. I., Dubard, M. E., Strickland, K. C., Moxley, K. M., Oleinik, N. V. and Krupenko, S. A. (2010) ALDH1L2 is the mitochondrial homolog of 10-formyltetrahydrofolate dehydrogenase. J. Biol. Chem. **285**, 23056-23063

Supplementary Table S3. Fecundity, litter size weight and male/female numbers for different combinations of breeder genotypes. Data are expressed as mean ± SE.

Parameter	$M^{+/+}$ X $F^{+/+}$	$M^{-/-}$ X $F^{+/+}$	$M^{+/+}$ X $F^{-/-}$	$M^{-/-}$ X $F^{-/-}$
# of pairs	3	7	6	3
Litter freq/mo	1.4±0.05	1.1±0.05	1.2±0.08	0.95±0..07
Litter size	5.6±0.35	7±0.38	6.1±0.40	6.1±0.59
No of males	34	115	83	61
Weight(g)	14±0.25	13±0.20	12±0.21	13±0.20
No of females	24	74	78	47
Weight(g)	13±0.26	11±0.26	11±0.17	11±.0.18

Supplementary Table S4. Sex and genotype distribution of progeny from intercrosses of *Aldh1l1*$^{+/-}$ mice.

Genotype	Male	Female	Total
Aldh1l2$^{+/+}$	20(29%)	43(35%)	63(32%)
Aldh1l2$^{+/-}$	33(48%)	49(40%)	82(43%)
Aldh1l2$^{-/-}$	16(23%)	31(25%)	47(25%)
Total	69 (100%)	123 (100%)	192 (100%)