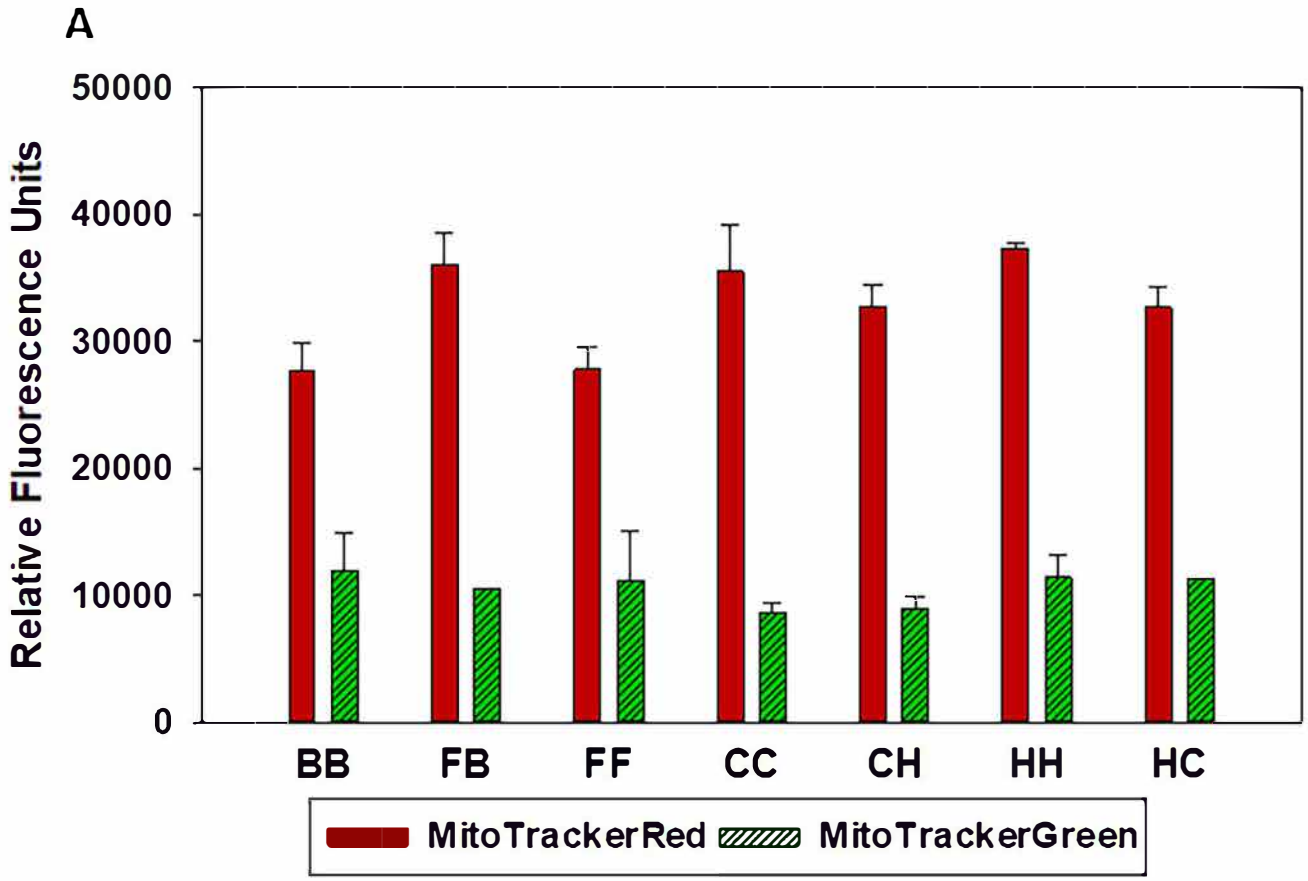
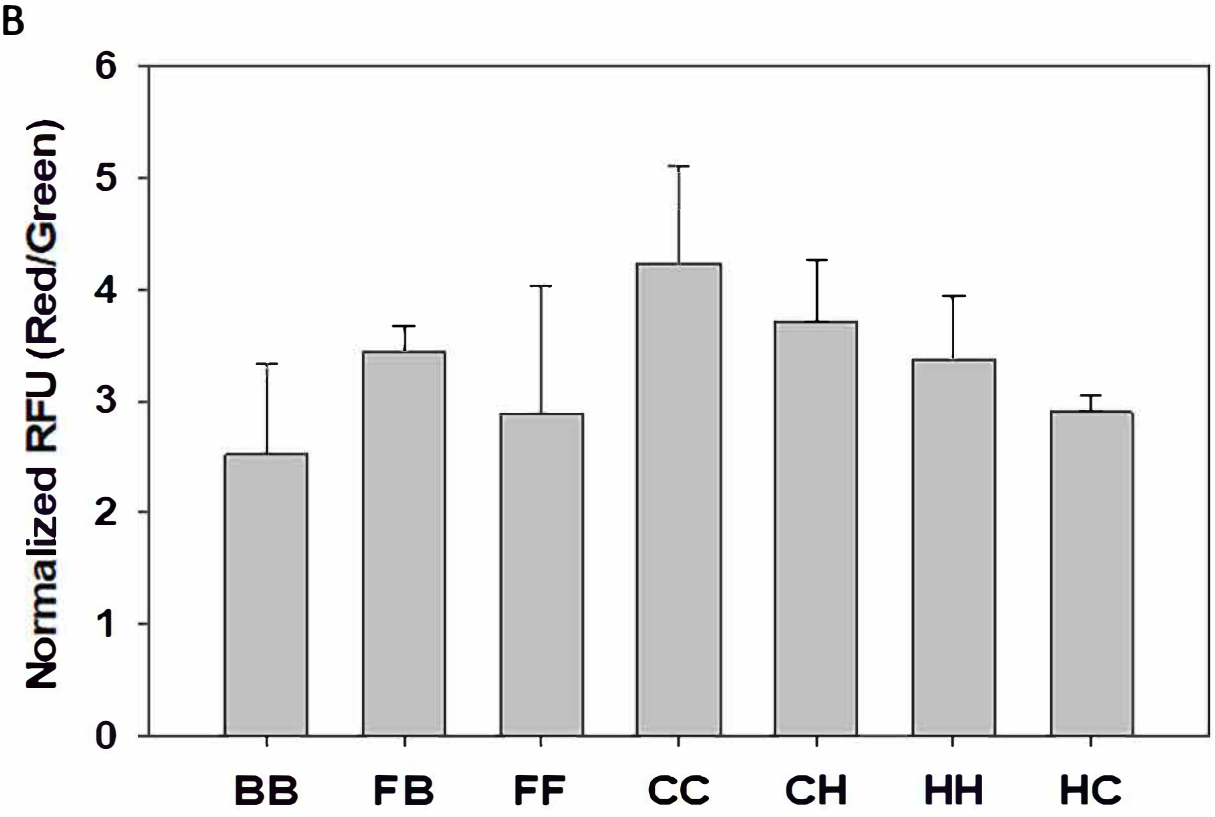
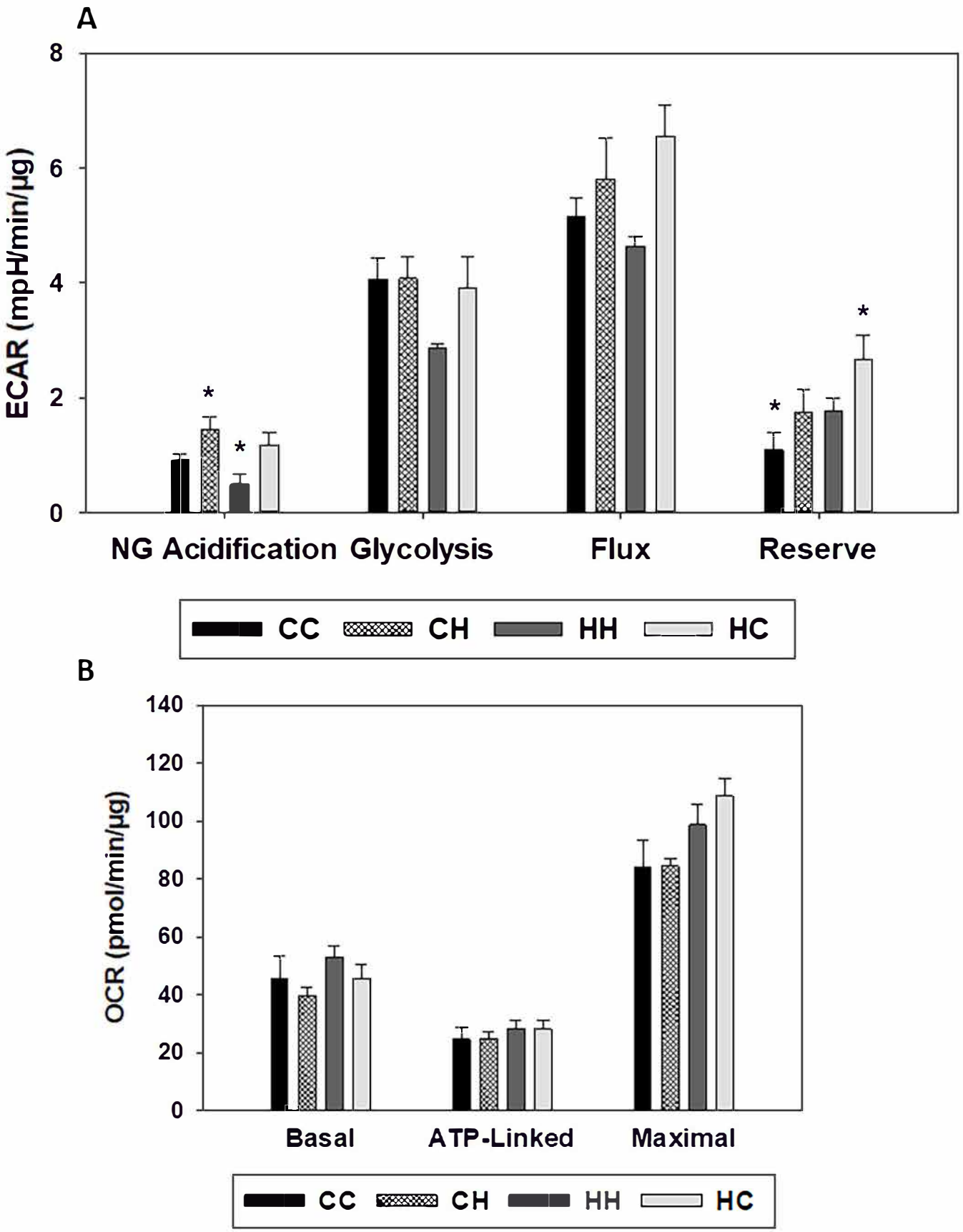
Brinker et al Supplemental Figure 1





S1: **Neither mitochondrial load nor membrane potential differ between MNX and wild-type strains**. Mouse embryonic fibroblasts were isolated from each MNX and wild-type strain and stained with MitoTracker Green FM to identify overall mitochondrial load and with MitoTracker Red CMXRos to identify membrane potential. Probe fluorescence in each cell was analyzed by flow cytometry. **A**: Relative fluorescence for each probe showing mitochondrial load and raw membrane potential. **B**: MitoTracker Red is normalized to MitoTracker Green to show membrane potential per mitochondrion. Error bars represent standard error of the mean.

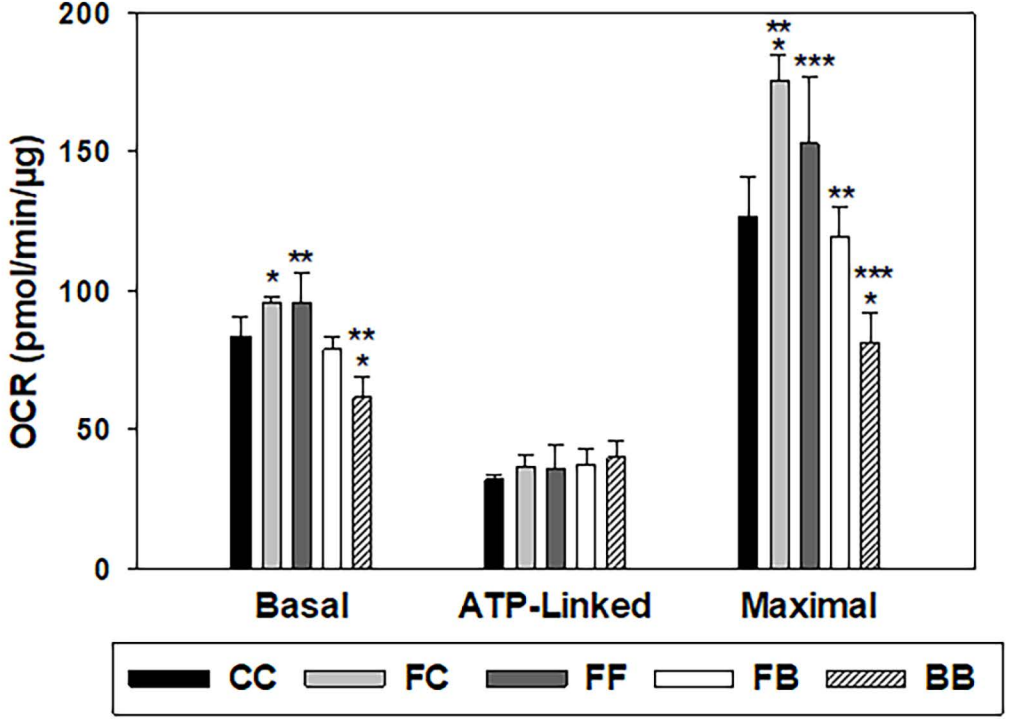
Brinker et al Supplemental Figure 2



S2: **OCR and ECAR profiles vary between C57BL/6J and C3H/HeN MNX and wild-type strains**. Mouse embryonic fibroblast from CC, CH, HH, and HC mouse strains were passaged once then analyzed for ECAR (Extracellular acidification rate) under serum starved conditions with no glucose to determine non-glycolytic acidification, after addition of glucose (glycolysis), after addition of oligomycin to determine glycolytic capacity (flux), and after addition of 2-DG (glycolytic reserve). * denotes $p<0.05$ and error bars represent standard error of the mean. CH and HH strains differ significantly in non-glycolytic acidification while CC and HC differ significantly in glycolytic reserve.

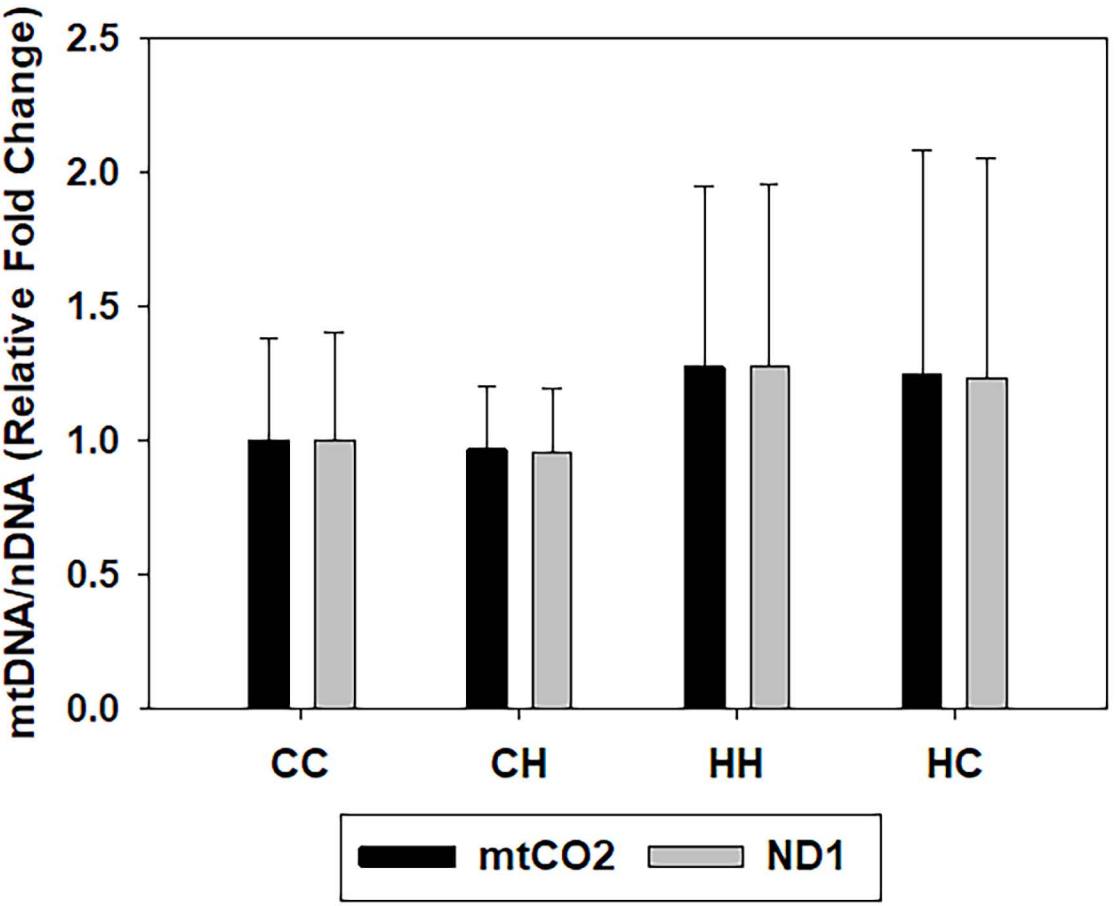
Supplemental Figure 3

A

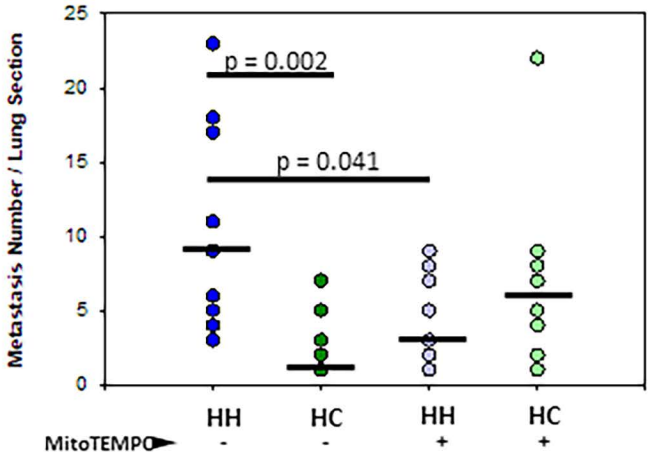


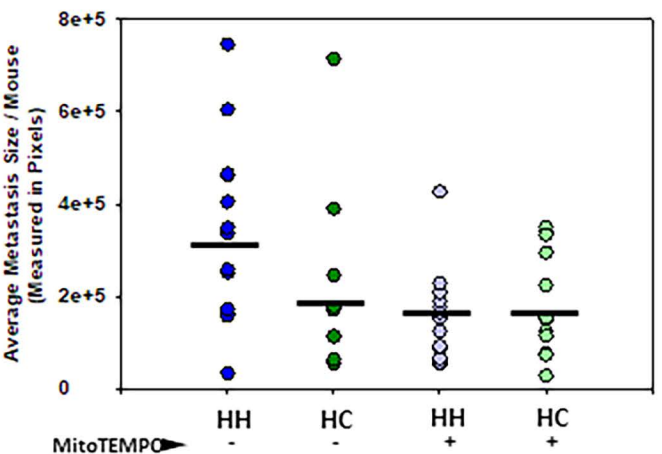
S3: **Mitochondrial flux of MNX and wild-type mice is variable**. MNX and wild-type mouse embryonic fibroblasts from each strain were plated and the Seahorse analyzer was used to measure oxygen consumption and extracellular acidification prior to compound addition (Basal respiration), after oligomycin addition (ATP-Linked respiration), and after addition of the uncoupler FCCP (Maximal respiration). * denotes $p<0.05$ and error bars represent standard error of the mean. **A**: Basal respiration differs significantly between * FC and BB, and ** FF and BB. Maximal respiration differs significantly between * FC and BB, ** FC and FB, as well as ***FF and BB. **B**: Basal, ATP-linked, and Maximal respiration does not differ significantly between CC, CH, HH, and HC strains

Supplemental Figure 4



S4: **Mitochondrial DNA content does not differ between C57BL/6J and C3H/HeN MNX and wild-type strains**. Lungs were isolated from four-week old CC (n=5), CH (n=7), HH (n=5), and HC (n=8) mice. Total DNA was isolated from each lung taking care to preserve mtDNA. qPCR utilizing Taqman primers for mitochondrial encoded mtCO2 and ND1 as well as nuclear encoded 18s was performed and fold change for mitochondrial genes was calculated relative to nuclear 18s. Error bars represent standard error of the mean.

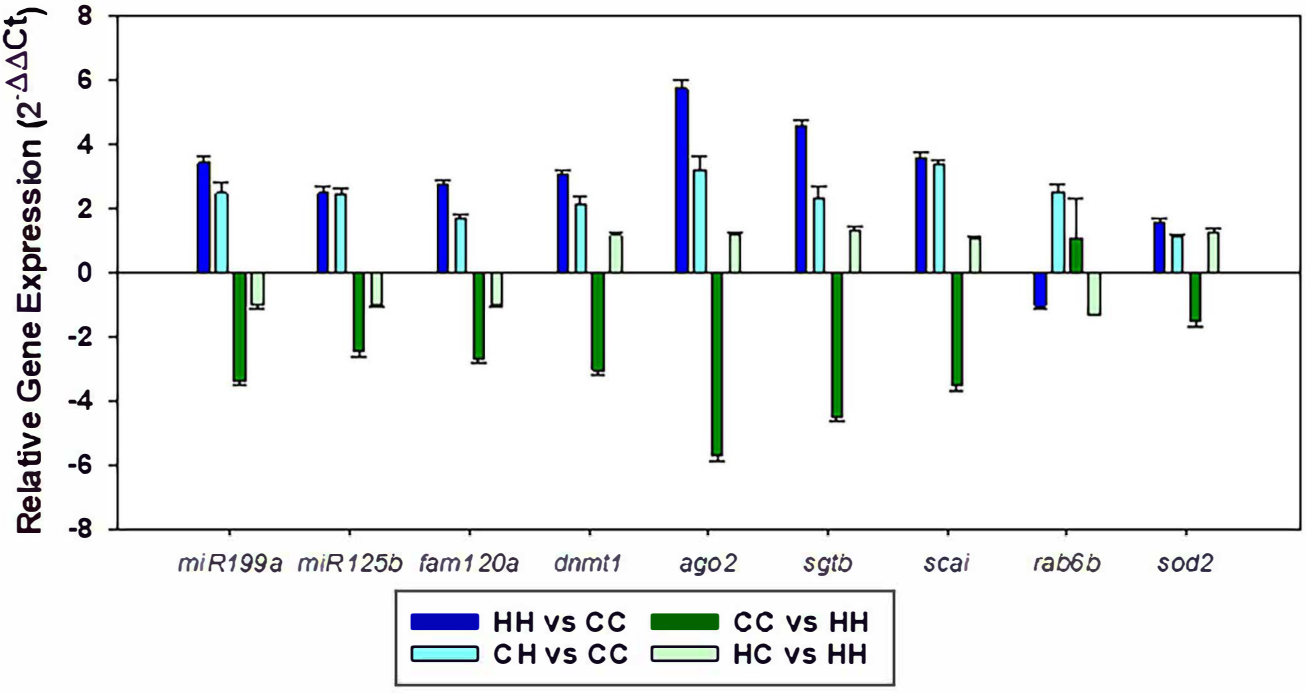




Supplemental Figure 5

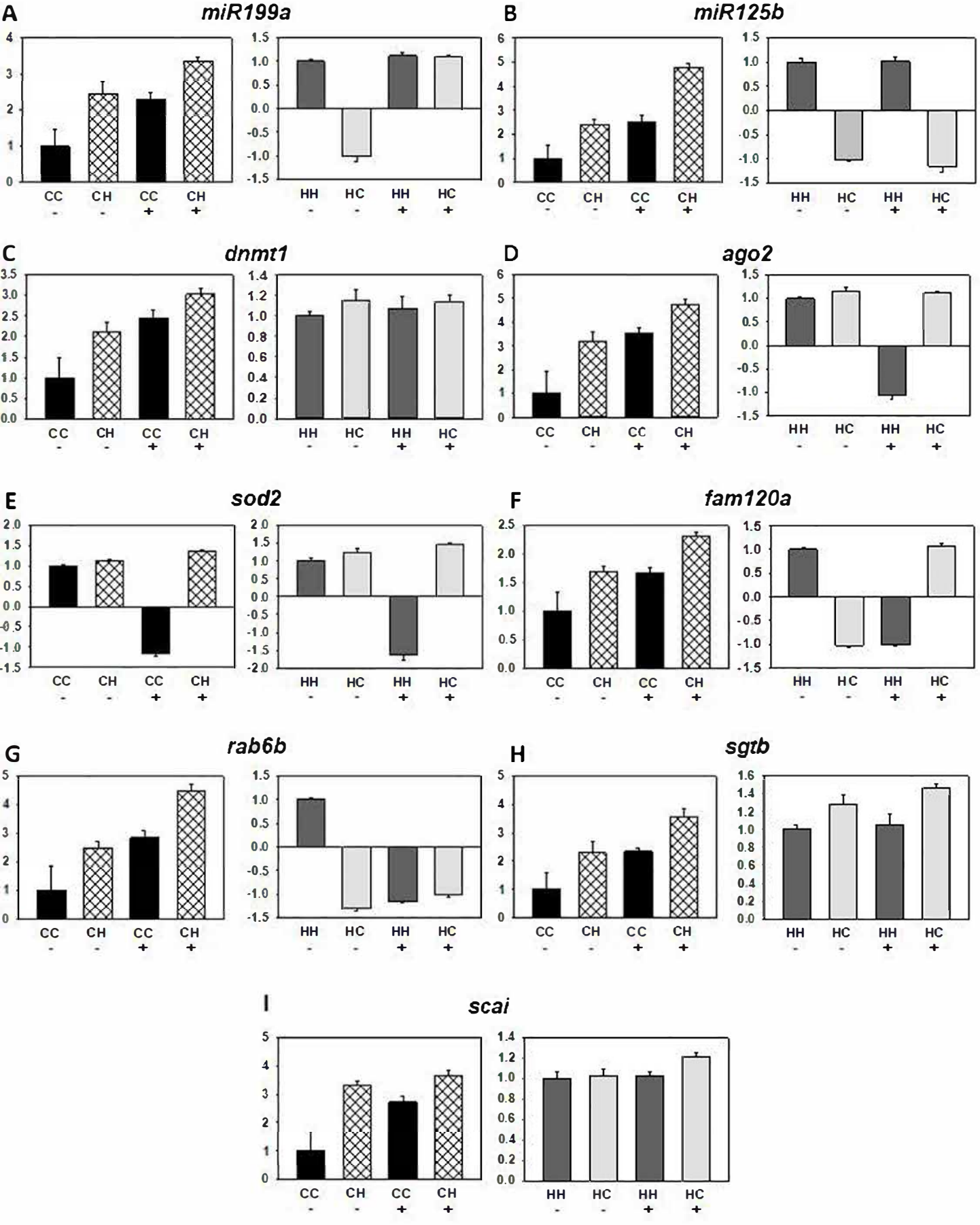
S5. Analysis of Metastatic size and area. Images (10x) from Figure 5B were analyzed using Photoshop and Image J for relative size and number of metastases. Analysis of formalin-fixed paraffin-embedded experimental lung metastases derived from K1735-M2 Melanoma cell intravenous injections. DMSO vehicle (-) or MitoTEMPO (+) was IP injected into 4-week-old wild-type or MNX mice 24 hours and again 1 hour prior to intravenous injection of cancer cells. Mice were euthanized two weeks post cell injection and lungs were harvested. Data as medians (n = 9-16; One-way ANOVA and multiple comparisons tests).

Supplemental Figure 6

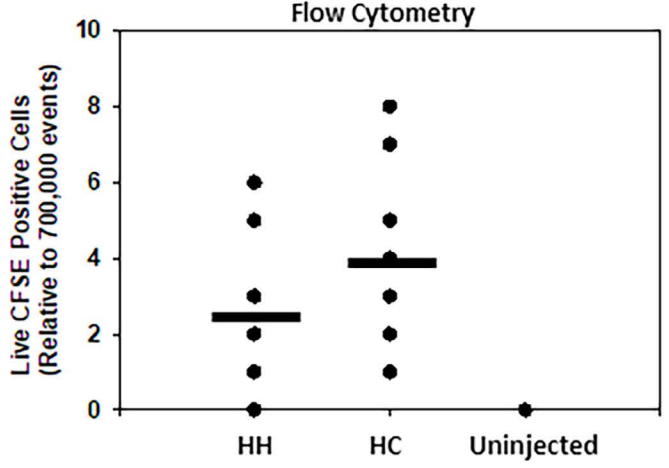


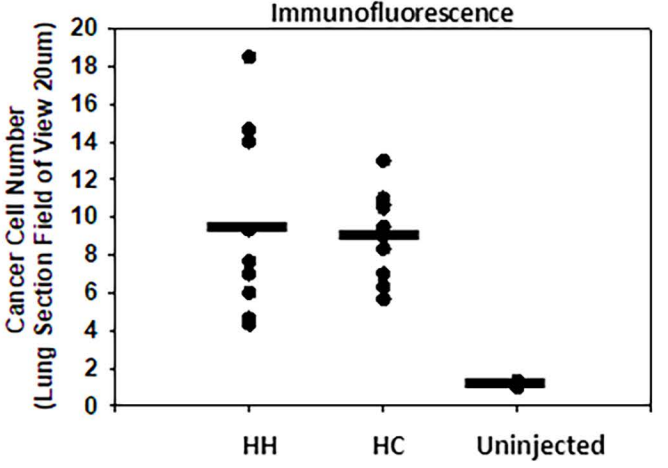
S6: **MtDNA affects expression of select nuclear genes**. RNA was isolated from lungs of 4-week old CC, CH, HH, and HC mice. qPCR was performed with Taqman primers to determine expression of nuclear genes normalized to the nuclear 18s housekeeping gene. Error bars represent standard error of the mean. Blue bars represent gene expression levels of HH and CH mice relative to CC while green bars represent gene expression levels of CC and HC mice relative to HH mice.

Supplemental Figure 7



S7: **Mitochondrial superoxide scavenging selectively alters nuclear gene expression**. RNA was isolated from lungs of 4-week old CC, CH, HH and HC mice treated with DMSO vehicle (-) or MitoTEMPO (+) 24 hours and 1 hour prior to euthanasia. qPCR was performed with Taqman primers to determine gene expression of **A**: miR199a **B**: miR125b **C**: dnmt1 **D**: ago2 **E**: sod2 **F**: fam120a **G**: rab6b **H**: sgtb and **I**: scai. All expression was normalized to 18s.





Supplemental Figure 8

S8: Mitochondria stromal haplotype does not alter cancer cell extravasation: Male and female wild-type (HH) and MNX (HC) mice were injected into the lateral tail vein with CFSE labeled K1735-M2 melanoma cells. Lungs were harvested 24 hours post-injection. **A**. The right lung lobes were analyzed by flow cytometry for CFSE positive cancer cells. Seven hundred thousand events were collected using the Acurri C6 cytometer and dead cells were identified using propidium iodide and excluded from further analysis. **B**. The left lung lobe was fixed in 10% formalin. Sections (3 x 20 μm) approximately 100 μm apart were analyzed from each mouse for detection of CFSE+ cancer cells. The number obtained from all three sections were then averaged and graphed in comparison to the uninjected controls