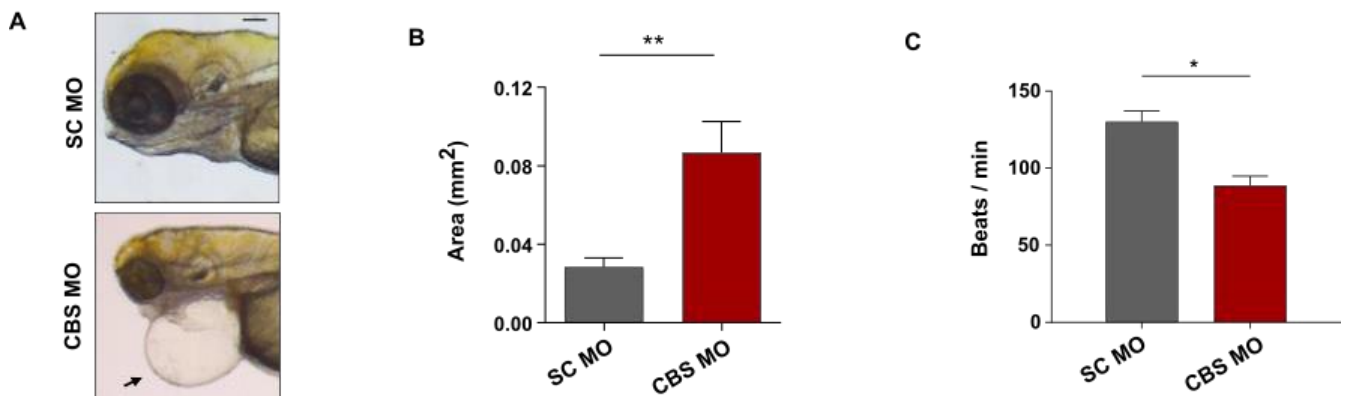
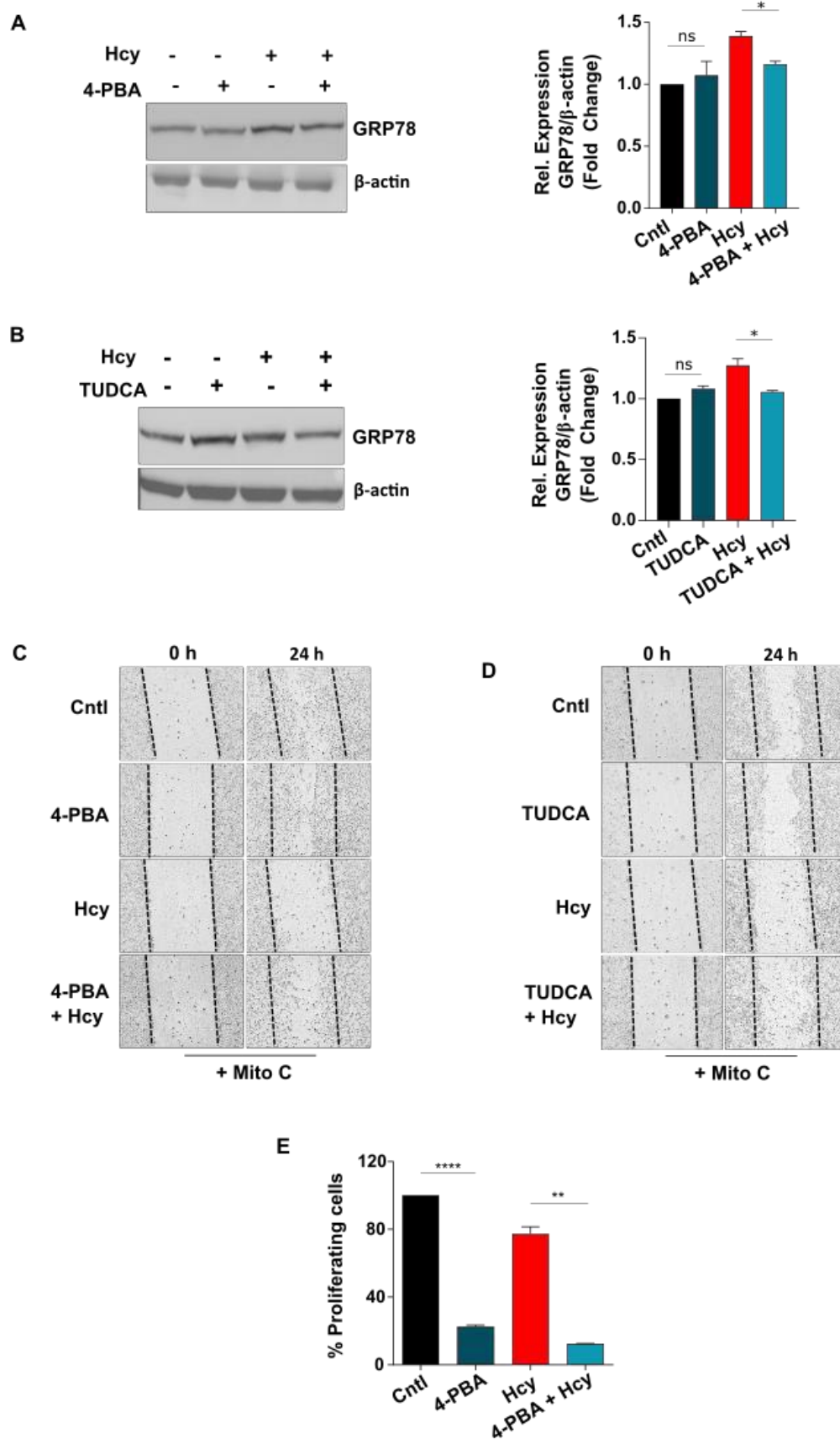


**Figure S1.** Primary HUVEC responds similarly to sub-lethal Hcy treatment. (A) Treatment of primary HUVEC cells with 2 mM Hcy for 24 h did not cause any cytotoxicity as measured by Trypan blue exclusion assay. (B) Bar plot showing that in presence of proliferation blocker Mitomycin C, migration of primary endothelial cells is drastically reduced upon sub-lethal Hcy treatment. (C) Percentage of proliferating cells, determined by BrdU cell proliferation assay, is significantly less in sub-lethal Hcy treated primary HUVEC compared to untreated control cells. Data are shown as Mean $\pm$ SEM with  $n \geq 3$ . \* $P \leq 0.05$ , \*\*\*\* $P \leq 0.0001$  and ns is non-significant ( $P > 0.05$ ).



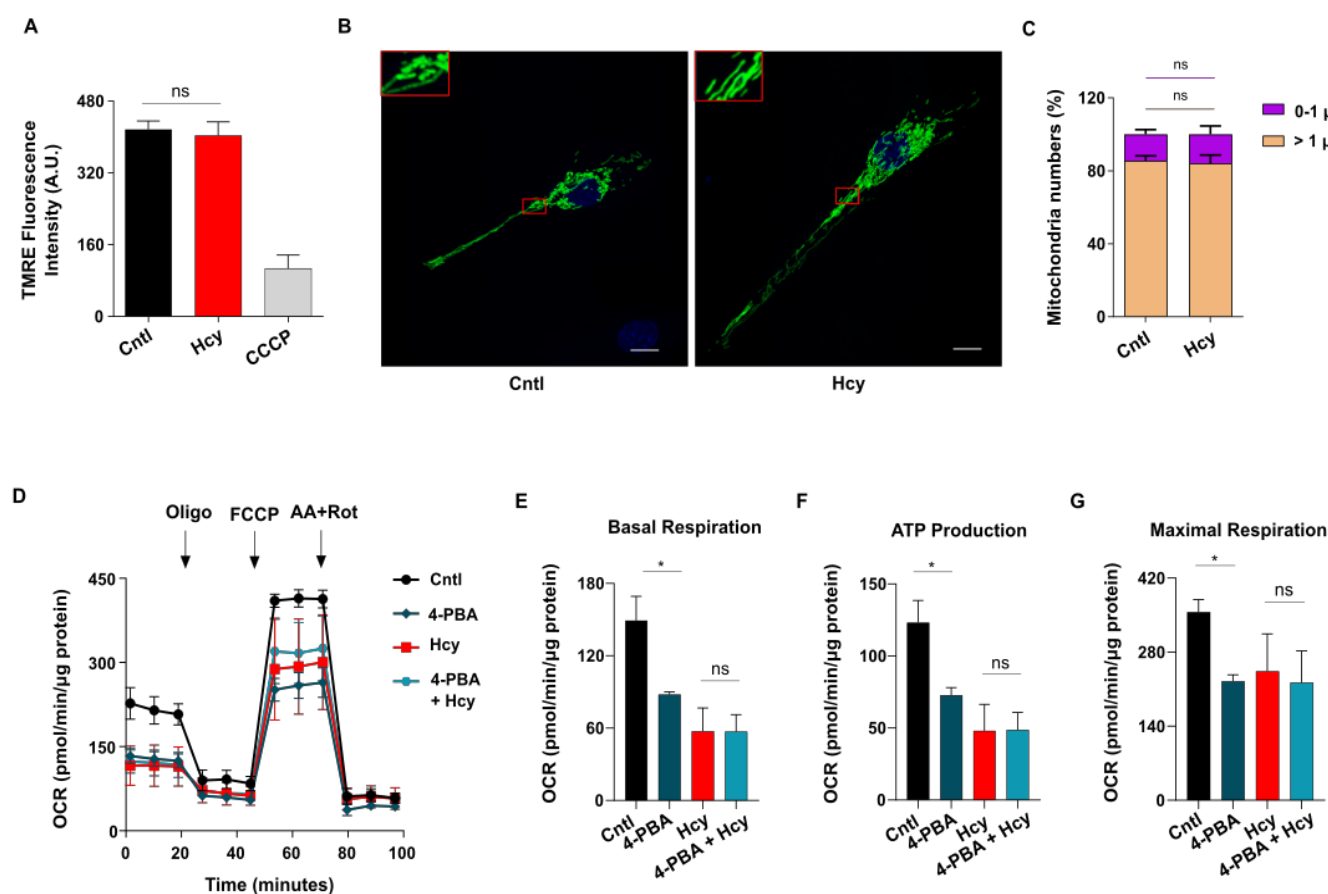
**Figure S2.** Sub-lethal HHcy induces abnormality in cardiac structure and function *in vivo*. (A) Representative brightfield images showing induction of severe pericardial edema in CBS MO injected embryos at 4 dpf. Arrow indicating enlarged pericardial area of CBS morphants. Scale bar, 0.1 mm. (B) Bar graph confirming pericardial area of CBS MO injected embryos is significantly higher compared to scrambled MO injected embryos at 4 dpf. (C) Bar graph showing that in comparison to SC MO injected embryos, heartbeat of CBS morphants is significantly reduced. Data are shown as Mean $\pm$ SEM with  $n \geq 3$ . \* $P \leq 0.05$  and \*\* $P \leq 0.01$ .

Figure S3



**Figure S3.** Effect of chemical chaperone mediated rescue of ER stress in sub-lethal HHcy treated endothelial cells. (A) & (B) Representative western blot showing sub-lethal Hcy treatment induced aberrant upregulation of GRP78 expression was rescued upon pre-treatment with 4-PBA (1 mM) and TUDCA (1 mM), respectively. As a loading control  $\beta$ -actin was used. Accompanying bar plots showing densitometric analysis (normalized to  $\beta$ -actin) of the protein bands. (C) & (D) Scratch wound assay images depicting that at 24 h in presence of proliferation blocker Mitomycin C, sub-lethal Hcy treatment induced endothelial migration defect is rescued upon pre-treatment with 4-PBA (1 mM) and TUDCA (1 mM), respectively. (E) Bar plot of BrdU cell proliferation assay showing no beneficial effect of 4-PBA (1 mM) on impairment of endothelial proliferation induced by 2 mM Hcy treatment at 24 h. Data are shown as Mean $\pm$ SEM with  $n \geq 3$ . \* $P \leq 0.05$ , \*\* $P \leq 0.01$ , \*\*\* $P \leq 0.0001$  and ns is non-significant ( $P > 0.05$ ).

Figure S4



**Figure S4.** Sub-lethal HHcy induced respiration defect neither influence mitochondrial health nor is it an outcome of ER stress. (A) Bar plot representing no alteration of mitochondrial membrane potential upon 2 mM Hcy treatment for 24 h, as measured using potentiometric dye TMRE. An uncoupler CCCP was used as a positive control. (B) Representative confocal images of mitoGFP transduced HUVEC/TERT2 cells showing that sub-lethal Hcy treatment did not alter integrity of mitochondrial network of endothelial cells. Scale bar, 10  $\mu$ m. (C) Measurement of mitochondria length by ImageJ analysis exhibiting no significant difference in between Hcy treated and non-treated cells. (D) OCR curves showing no improvement of sub-lethal Hcy treatment induced mitochondrial respiration defect in presence of chemical chaperone 4-PBA. (1 mM). (E), (F) & (G) Respective bars of basal respiration, ATP production and maximal respiration revealing no improvement upon 4-PBA (1 mM) pre-treatment as compared to only Hcy treated endothelial cells. Data are shown as Mean $\pm$ SEM with  $n \geq 3$ . \* $P \leq 0.05$  and ns is non-significant ( $P > 0.05$ ).

**Table S1.** List of Primers used in this study for qPCR analysis.

Gene	Primer sequence
Zebrafish - VEGFAA - forward	GCCCACATACCCAAAGAAGG
Zebrafish - VEGFAA - reverse	CTCATCGGGATACTCCTGGAT
Zebrafish - VEGFR2 - forward	TTTGGTAGAGGGATCTCGTC
Zebrafish - VEGFR2 - reverse	GCGTACCGATGACACATTTC
Zebrafish - VEGFR1 - forward	ATGGGAACAGCAGCACTCTT
Zebrafish - VEGFR1 - reverse	TGAAGACGGAGGGACAATC
Zebrafish - 18S - forward	TCGCTAGTTGGCATCGTTTATG
Zebrafish - 18S - reverse	CGGAGGTTCTGAAGACGATCA
Human - VEGFA - forward	TCCAACCTCTGGGCTGTTCT
Human - VEGFA - reverse	CCCCTCTCCTCTTCCTTCTC
Human - VEGFR2 - forward	TGGGGATTGACTTCAACTGG
Human - VEGFR2 - reverse	TTCTTGGTCATCAGCCCACT
Human - VEGFR1 - forward	ACCACGCCCAGTCAAATTAC
Human - VEGFR1 - reverse	TGGGAATTGCTTTGGTCAAT
Human - 18S - forward	CTACCACATCCAAGGAAGCA
Human - 18S - reverse	TTTTTCGTCACCTCCCCG

**Table S2.** Optimized parameters of different metabolites analyzed through targeted metabolomics.

Sr No.	Metabolite	Precursor (m/z)	Fragment (m/z)	Peak type	Charge	Collision Energy (V)
1	Pyruvate	87	42.99	Quantifier	-1	10
			42.99	Quantifier	-1	
2	Lactate	89	40.99	Qualifier	-1	20
			71.01	Quantifier	-1	
3	Fumarate	115	44.49	Qualifier	-1	20
			99	Qualifier	-1	
4	Succinate	117.01	73.01	Quantifier	-1	20
			87	Quantifier	-1	
5	Oxaloacetate	130.99	59	Qualifier	-1	20
			114.9	Quantifier	-1	
			72.98	Qualifier	-1	
6	Malate	133.01	71.0	Qualifier	-1	10
7	Phospho- enol pyruvate (PEP)	166.97	78.94	Quantifier	-1	10
8	Dihydroxyacetone phosphate (DHAP)	168.98	96.95	Qualifier	-1	
			78.94	Quantifier	-1	20
			96.95	Qualifier	-1	
9	3-Phosphoglycerate (3PG)	184.98	78.94	Quantifier	-1	20
			110.9	Quantifier	-1	
10	Citrate	191.01	86.99	Qualifier	-1	20
			138.9	Qualifier	-1	
			96.95	Quantifier	-1	
11	Glucose-6-phosphate	259.02	78.94	Qualifier	-1	20
			96.95	Quantifier	-1	
12	Fructose-1,6- biphosphate	338.98	78.94	Qualifier	-1	20