

Supplementary Materials

Table 1. Quantitative PCR primers

Gene	Forward sequence	Reverse sequence
PGC-1 α	5'-ACCCCAGAGTCACCAAATGA-3'	5'-GCAGTTCCAGAGAGTTCCAC-3'
PPAR α	5'-GGTCCTCTGGTTGTCCCCTT-3'	5'-GTGAGTTACGCCCCAAATGCA-3'
ERR α	5'-CACAAGGAGGAGGAGGATGG-3'	5'-AGGCGTTTGGGTAGAGAGCT-3'
ACADS	5'-CTGGATTGTGCCGTGAAGTAC-3'	5'-GCTTGAAGTGGATGTTTTGGA-3'
ACADM	5'-GACGGAGCAGCAGAAAGAGTT-3'	5'-CTTGATGAGAGGGAACGGGTA-3'
ACADL	5'-AAGTGAATGACCCTGCCAAGA-3'	5'-ATGCCCCCAGTTCTTTGAGTC-3'
CD36	5'-GTTATTGGTGCTGTCCTGGCT-3'	5'-CCGTTTTACCCAGTTTTTGA-3'
CPT-1a	5'-GGCATGATCGCAAAGATCAGT-3'	5'-GCCACCCAGAGCCCTGTACCA-3'
GLUT4	5'-AGAGTCTAAAGCGCCT-3'	5'-CCGAGACCAACGTGAA-3'
CS	5'-GCATGGACTAGCAAACCAGG-3'	5'-ATATCCTGGGACAACCCGTC-3'
PDH1	5'-CACGGACCATCTCATCACTG-3'	5'-TAGCACAGCCTCCTCTTCGT-3'
PDK1	5'-ACAATTCACGGAATGCCCCCT-3'	5'-TACTTGGCGTAGAGACGGGA-3'
PDK4	5'-CAAAGATGCTCTGCGATCAGTA-3'	5'-TGGTGAAGGTGTGAAGGAAC-3'
Hexokinase-2	5'-GCTGTGAAAATGTTGCCTACC-3'	5'-CATTGTCCGTCACCCTTACTC-3'
Mitochondrial-D loop region	5'-CCTCCCATTCATTATCGCCGCCCTTG C-3'	5'-GTCTGGGTCTCCTAGTAGGTCTGGGAA- 3'
18S rRNA	5'-GCAATAACAGGTCTGTGATGCC-3'	5'-CACGAATGGGGTTCAACG-3'
ANP	5'-GGTAGGATTGACAGGATTGGA-3'	5'-GCAGATTTGGCTGTTATCTTCG-3'
BNP	5'-CAGAACAATCCACGATGCCAGA-3'	5'-GCGCTGTCTTGAGACCTAAG-3'
β -MHC	5'-TGACCTGAAGCTGACCCA-3'	5'-CCTGCTCATCCTCAATCCTG-3'
Cpt-1b	5'-AGCAACTATTACGCCATGGATT-3'	5'-GTACCATACCCAGTGCCATC-3'
LCAD	5'-CACAAAAGAACAGATCGAGCAG-3'	5'-AGAATCCAATCACTCCCAGAC-3'
VLCAD	5'-CCTGCCAAGAATGACTCCTT-3'	5'-CACAATCTCTGCCAAGCGA-3'
RELM α	5'-GCTGGGATGACTGCTACTGG-3'	5'-AAACGGGGTTAATGGGCAAT-3'
POLG2	5'-CTGAGTAAGGAACAGCTAGTGG-3'	5'-ATCCAGGCAGTGAACATAGTG-3'
POLRMT	5'-GTGAGCAAGCTGTCCAGA-3'	5'-ACCTTCTTCACCCTCATCTCA-3'
TFAM	5'-AGCTAAACACCCAGATGCAA-3'	5'-GTACACCTTCCACTCAGCTTT-3'
TOP1MT	5'-CTGCATCTCGAGTGACGAC-3'	5'-GTGTGTGATGAGCTTCCTCTC-3'
IL-1 β	5'-GTGCTGTCTGACCCATGT-3'	5'-TTGTCGTTGCTTGTCTCTCC-3'
IL-6	5'-CAGAGCAATACTGAAACCCTAGT-3'	5'-CCTTCTGTGACTCTAACTTCTCC-3'
IL-18	5'-CGGAGCATAAATGACCAAGTTC-3'	5'-GCCAGTCCTCTTACTTACTTCACTATC-3'
NLRP3	5'-TTTCCAGACCCTCATGTTG-3'	5'-GTGCTGAGACTTGAGAAGAGAC-3'

Supplemental Figures:

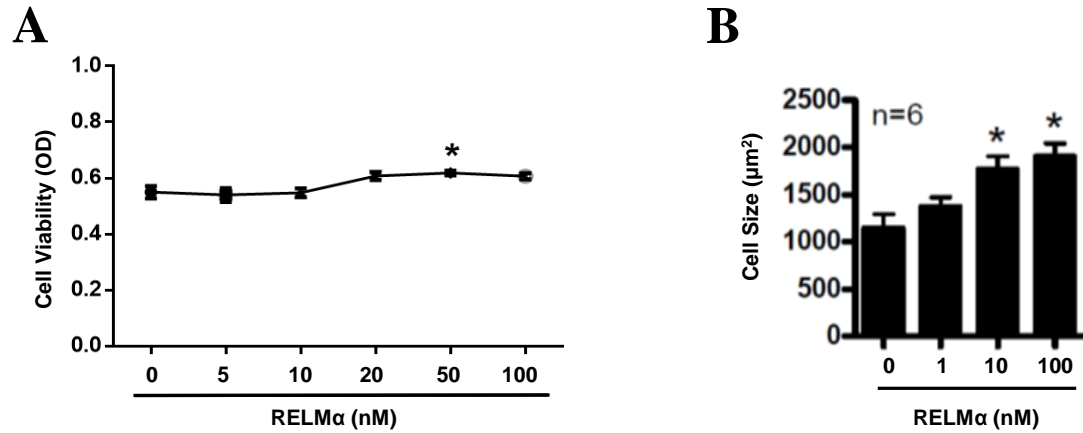


Figure S1: Effects of rRELMα on cardiomyocytes viability and size. (A) Cell viability evaluated by colorimetric BrdU assay in neonatal rat cardiomyocytes stimulated with 0 to 100 nM RELMα. n=6/group. (B) Quantification of the cell surface area with indicated doses of RELMα in NRCMs. n=50-100 cells per group and n= 6/group. * $p < 0.05$ vs without RELMα group.

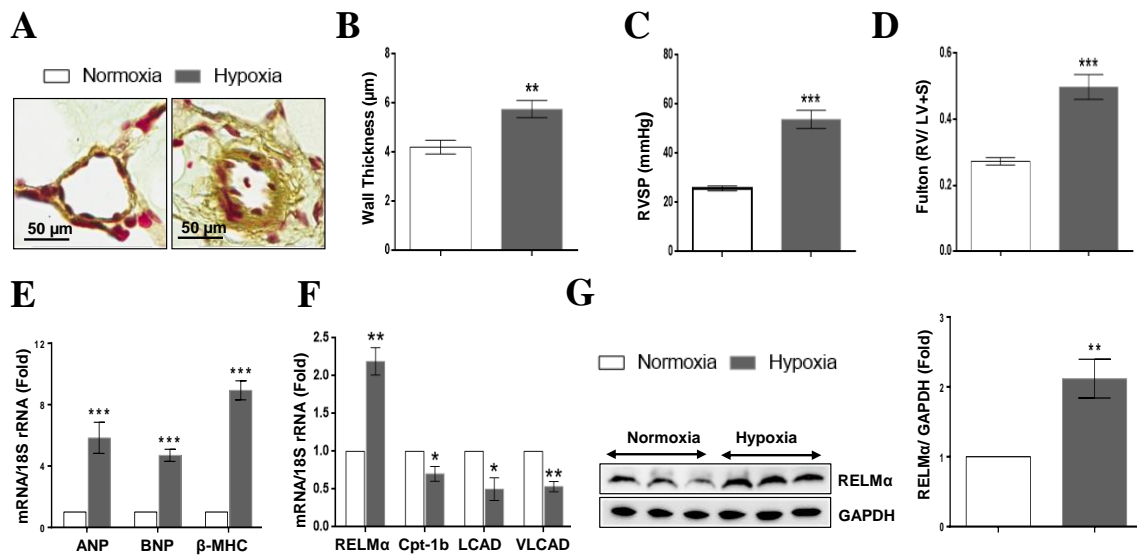


Figure S2: Effects of hypoxia-induced pulmonary hypertension and right ventricular hypertrophy on RELM α and mitochondrial metabolic genes. (A, B) Representative images and its relative quantification from rat lung tissue sections stained for VWF (von Willebrand factor; brown) and α -SMA (α -smooth muscle actin; red) to define the medial vessel wall thickness. $n=100-150$ medial vessel count per group and $N=6$ rats/group. (C, D) Hemodynamic analysis, right ventricular systolic pressure (RVSP), and Fulton index [right ventricle (RV) weight/ left ventricle (LV) + septum (S) weight] from rats RV tissue. $N=6$ rats/group. (E F) Quantitative PCR analyses of atrial natriuretic peptide (ANP), brain natriuretic peptide (BNP), beta-myosin heavy chain (β -MHC), RELM α (Resistin like molecule α), carnitine palmitoyltransferase-1b (CPT-1b), long-chain acyl-CoA dehydrogenase (LCAD), and very long-chain acyl-CoA dehydrogenase (VLCAD) in total mRNA derived from rats RV tissue. $N=3-4$ rats/group. (G) Immunoblotting (left) and statistical (right) analysis RELM α in RV tissue of indicated rat group. $N=6$ rats/group. *, **, *** $p<0.05$, 0.01, 0.001 vs. Normoxia.

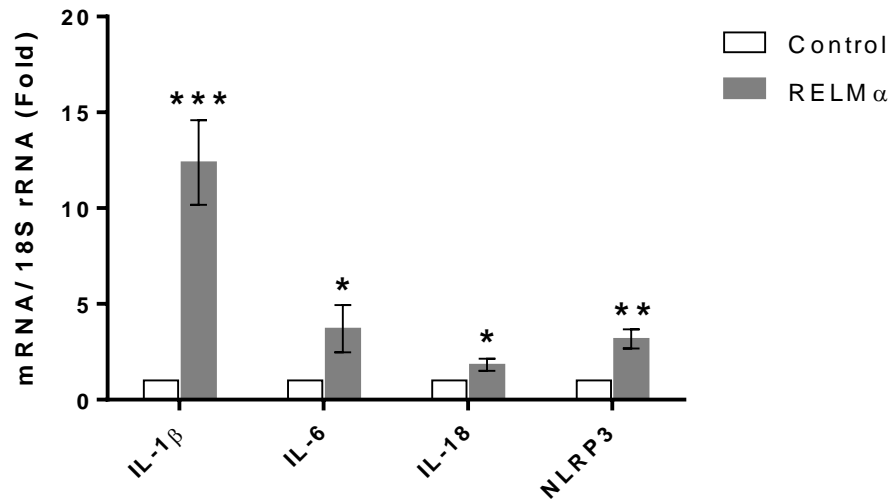


Figure S3: Effects of rRELM α on inflammatory cytokines and NLRP3 in NRCMs. qPCR analyses of cytokines (IL-1 β , IL-6, IL-18) and NLRP3 expression from total mRNA derived from NRCMs group treated with PBS or rRELM α (50 nM, 24h). n= 5/group. *,*** p <0.05, 0.001 vs. control.

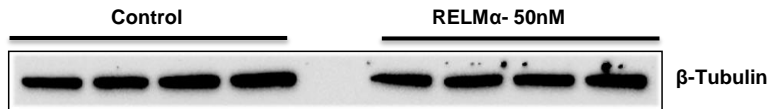


Figure S4: Representative immunoblotting of beta-Tubulin with or without RELM α (50 nM) stimulation in NRCMs. n=4/group.

Sea Horse Image (Figure 4A) Approval from Agilent Technologies, Inc.



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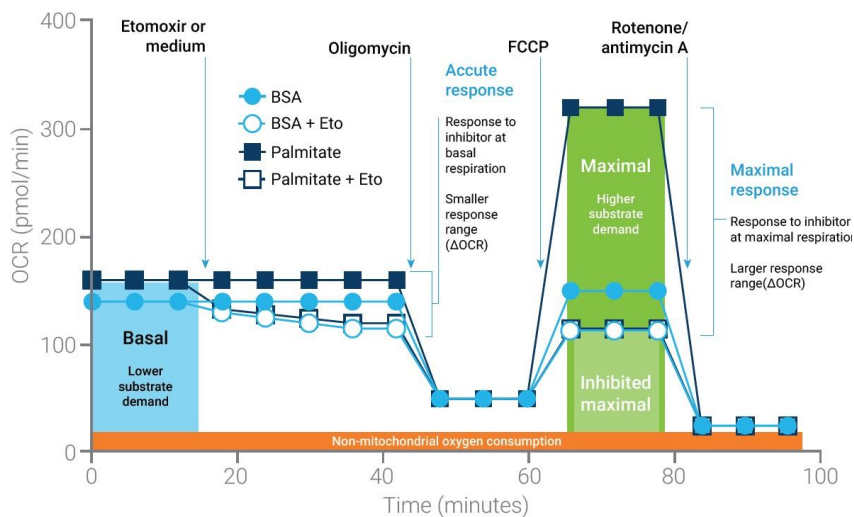


Figure 2. Advanced assay output

See: <https://www.agilent.com/cs/library/usermanuals/public/user-manual-substrate-oxidation-stress-test-kits-cell-analysis-5994-1164en-agilent.pdf> (page 7).



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