

Supplementary information

Title

Released Mitochondrial DNA Following Intestinal Ischemia Reperfusion Induces the Inflammatory Response and Gut Barrier Dysfunction

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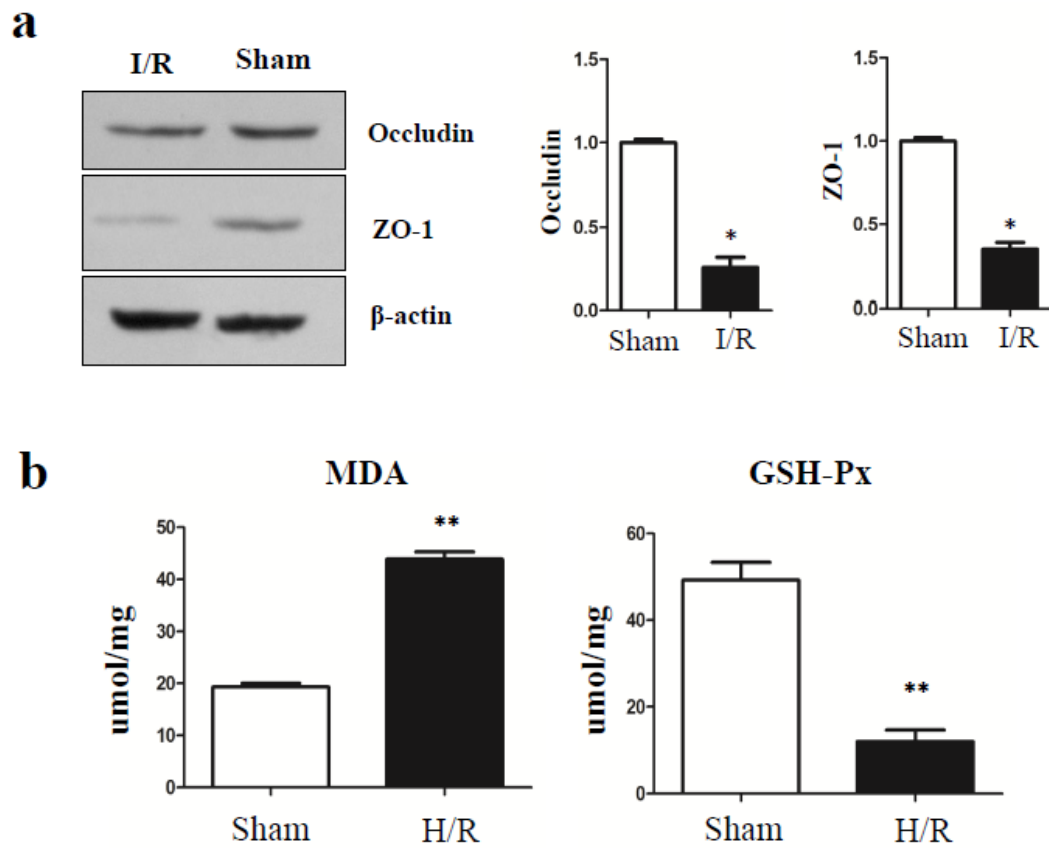
Table S1. Primers used for quantitative PCR analysis

| Genes | Primers sequence |
|--------------------|---|
| Human cytochrome B | F: 5'- ATGACCCCAATACGCAAAAT -3' R: 5'- ATGACCCCAATACGCAAAAT -3' |
| Human Cox3 | F: 5'- ATGACCCACCAATCACATGC -3' R: 5'- ATCACATGGCTAGGCCGGAG -3' |
| Human ND1 | F: 5'- ATACCCATGGCCAACCTCCT -3' R: 5'- GGGCCTTTGCGTAGTTGTAT -3' |
| Mouse cytochrome B | F: 5'- TGAGGGGGCTTCTCAGTAGA -3' R: 5'- CTGTTTCGTGGAGGAAGAGG -3' |
| Mouse Cox3 | F: 5'- CGTGAAGGAAACTACCCAGG -3' R: 5'- CGCTCAGAAGAATCCTGCAA -3' |
| Mouse ND1 | F: 5'- GGATCCGAGCATCTTATCCA -3' R: 5'- GGTGGTACTCCCTCTGTAAA -3' |
| TLR 9 | F: 5'- CCTGGCACACAATGACATTCA -3' R: 5'- TAAAGGTCCTCCTCGTCCCA -3' |
| MyD88 | F: 5'- GAGATCCGCGAGTTTGAGAC -3' R: 5'- TTGTCTGTGGGACACTGCTC -3' |
| NFκB | F: 5'- GAGGACTTGCTGAGGTTGG -3' R: 5'- TGGGGTGGTTGATAAGGAGTG -3' |
| GAPDH | F: 5'- GGCACAGTCAAGGCTGAGAATG -3' R: 5'- ATGGTGGTGAAGACGCCAGTA -3' |

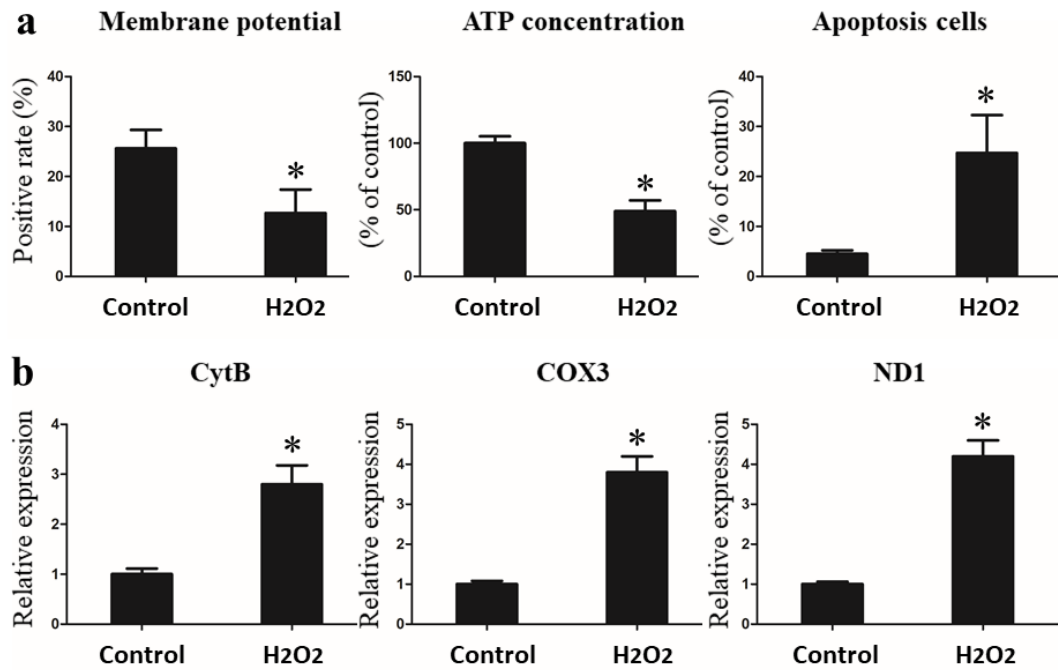
Supplementary Table 2. Oxidative and antioxidative parameters in sham and I/R group.

| Parameters | Sham | I/R | P value |
|--|-------------|------------|----------------|
| H ₂ O ₂ (mmol/l) | 17.5±5.1 | 25.7±6.5 | 0.029 |
| [•] OH (U/mggrot) | 236.5±75.1 | 321.9±66.6 | 0.047 |
| GSH (mg/gprot) | 3.9±0.8 | 2.2±1.1 | 0.012 |
| CAT (U/mggrot) | 21.1±6.5 | 13.6±2.1 | 0.035 |

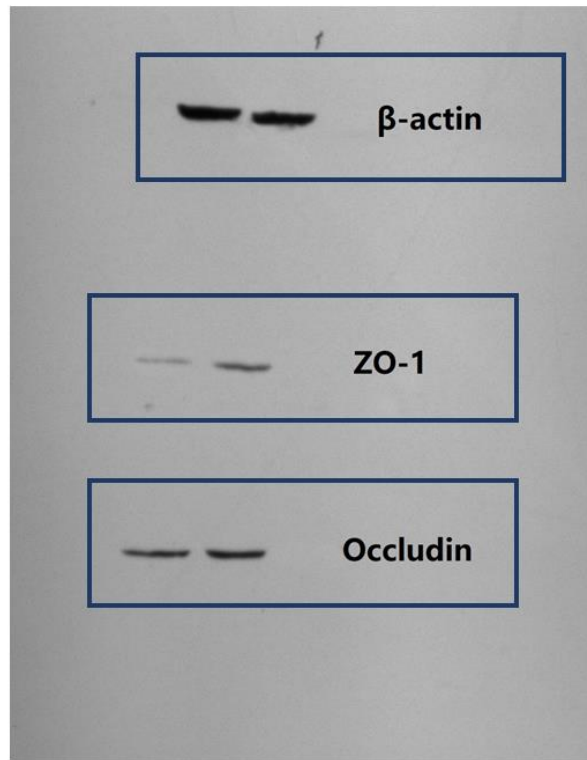
Values are mean ±SD. hydrogen peroxide (H₂O₂); hydroxyl radical ([•]OH); glutathione (GSH); catalase (CAT).



Supplementary Fig.S 1 Ischemia reperfusion significantly decreased the expression of tight junction proteins and elevated oxidative stress in vitro. (a) Occludin and ZO-1 expression were determined by western blot. (b) H/R of the IEC-6 cell lines significantly increased MDA, and the level of GSH-Px was obviously decreased. * $p < 0.05$, ** $p < 0.01$ when compared with sham group.



Supplementary Fig.S 2 H₂O₂-induced mitochondrial dysfunction, apoptosis, and mtDNA release. (a) mitochondrial membrane potential, ATP content, and cell apoptosis were quantified after H₂O₂ treatment. (b) detection of mtDNA levels in supernatant of H₂O₂-treated IEC-6 cells. *P<0.05 vs control.



Supplementary Fig.S 3 Unprocessed original scans for the bolts in Figure S1a.