Supplementary Materials

1. Supplemental Figures and Legends

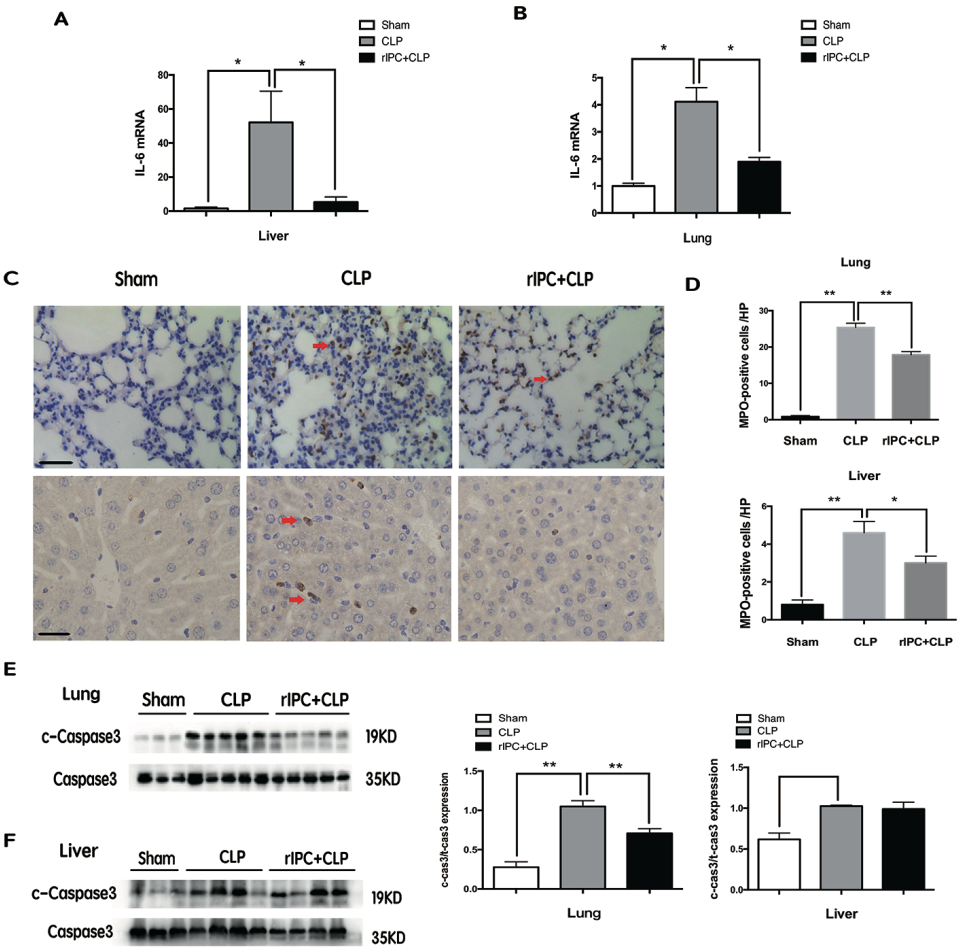

Figure S1.

Figure S1. Limb ripc effectively ameliorated inflammatory injury and cell apoptosis induced by CLP in mlti-organs. A and B, mRNA level of IL-6 and TNF-α in lungs and livers in different groups. C and D, MPO-positive staining for inflammatory cell infiltration in lungs and livers in different groups. (magnification × 400). E and F, Western blot analysis of cleaved-caspase 3 (c-caspase 3) in lungs and livers in different groups. (n≥3). *p < 0.05 and **p < 0.01. Data are the mean ± SEM.

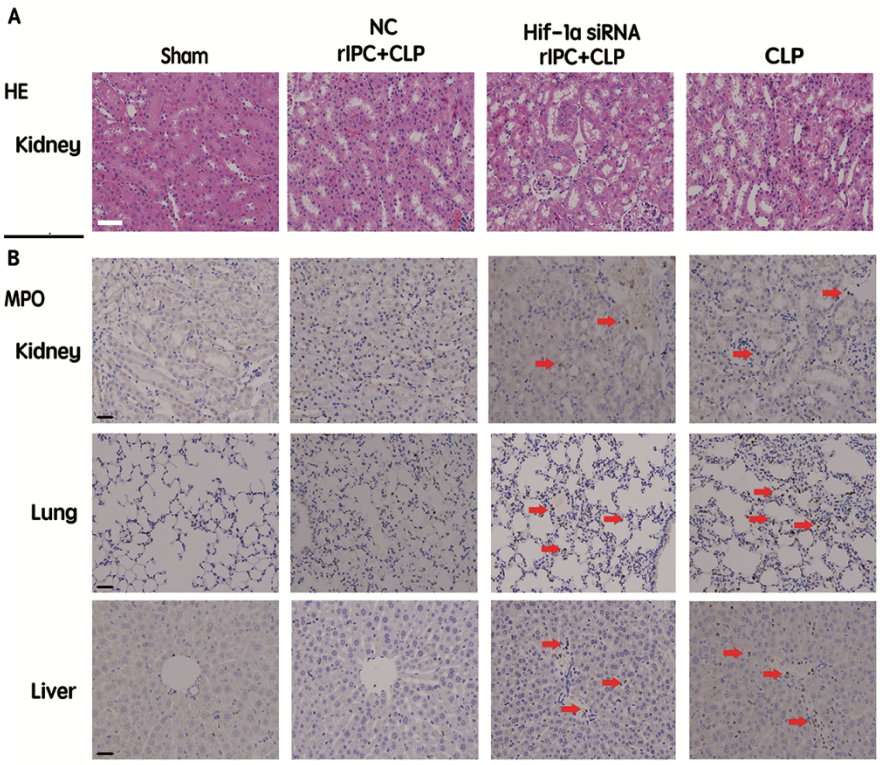

Figure S2.

Figure S2. HIF-1α siRNA inhibited the protective effect of limb rIPC against CLP injury in multi-organs. A, compared to NC prior to rIPC, HIF-1α siRNA administration aggravated the vacuolization of renal tubular cells induced by CLP, especially in the outer strip of the outer medulla. B, By MPO staining assay, increased number of MPO-positive cells induced by CLP were detected in kidney, lung and liver of the mice with HIF-1α siRNA prior to rIPC, when compared with NC group. (magnification × 200).

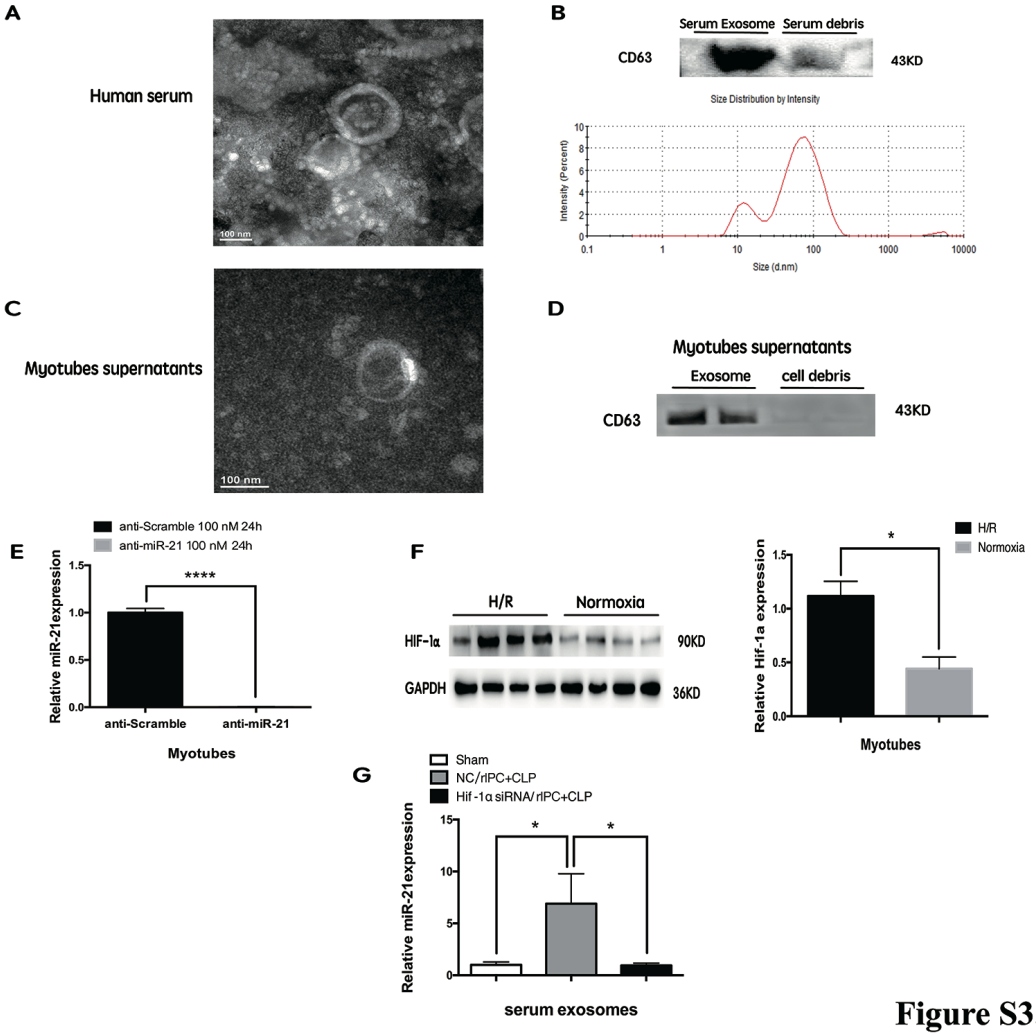

Figure S3.

Figure S3. Identification of exosomes and evaluation of miR-21/HIF-1α expression. A and B, the vesicles fraction extracted from human serum were identified as exosomes by TEM analysis and immunoblot assay; the size of the vesicles were distributed between 50-100nm. C and D, identification of exosomes derived from myotubes supernatants by TEM and immunoblot assay. E-G, C2C12 cells were differentiated into myotubes with conditional medium and then subjected to hypoxia for 6 hrs or 24 hrs followed by re-oxygen for 4 hrs (H/R). E, miR-21 level in differentiated C2C12 cells (myotubes). H/R increased the miR-21 content in cells at two time points, and the increase was prominent at 24 hrs. (n = 4). F, HIF-1α expression in myotubes. HIF-1α expression was up-regulated in the H/R group compared with the normoxia group. (n = 4). C, miR-21 expression in serum exosomes. HIF-1α knock-down inhibited miR-21 expression in mouse serum exosomes, although the pretreatment of limb rIPC. (n = 4). *p < 0.05 and **p < 0.01. Data are the mean ± SEM.

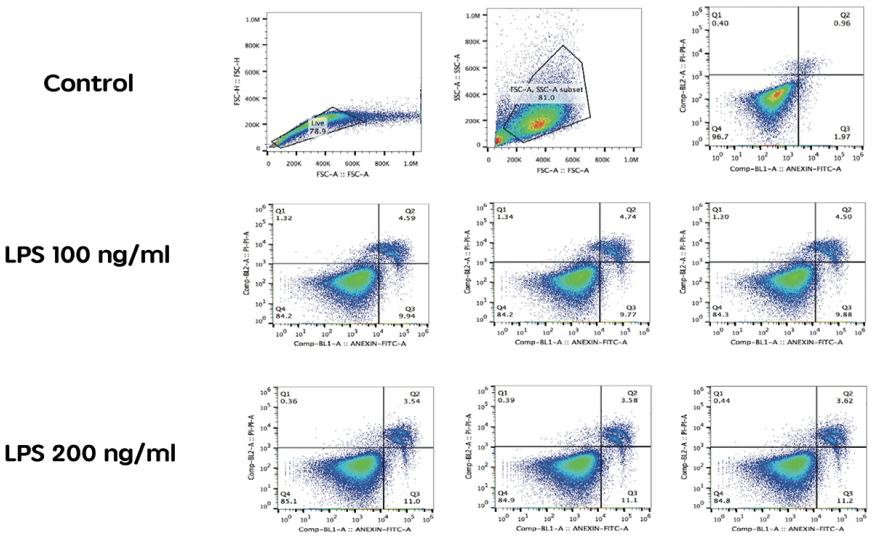

Figure S4.

Figure S4. 100ng/ml LPS was sufficient to trigger apoptosis injury in mouse renal tubular epithelial cells (mTECs). A, Annexin V/PI dual staining for cell apoptosis measurements. Representative density plots of cell apoptosis are shown after staining. The proportion of apoptotic cells is represented for both early (Annexin V+/PI −) and late (Annexin V+/PI+) apoptotic cells after the administration of LPS, in comparison with the proportions of live cells (Annexin V−/PI−). The results indicated that 100ng/ml was sufficient to trigger the apoptosis of mTECs. *p < 0.05 and **p < 0.01. Data are the mean ± SEM.

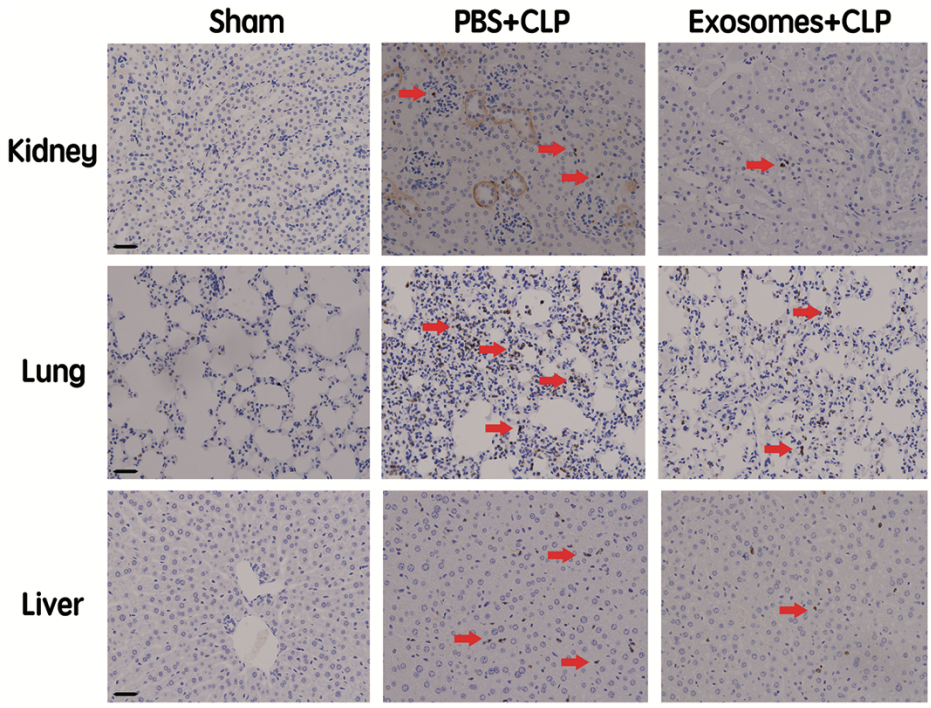

Figure S5.

Figure S5. Exosomes from H/R myotubes infusion rescued miR21$^{-/-}$ mice from pro-inflammatory cells infiltration in multi-organs. MPO-positive staining for inflammatory cell infiltration in kidneys, lungs and livers at with exosomes infusion after CLP in miR21$^{-/-}$ mice. (n = 4). (magnification × 200).

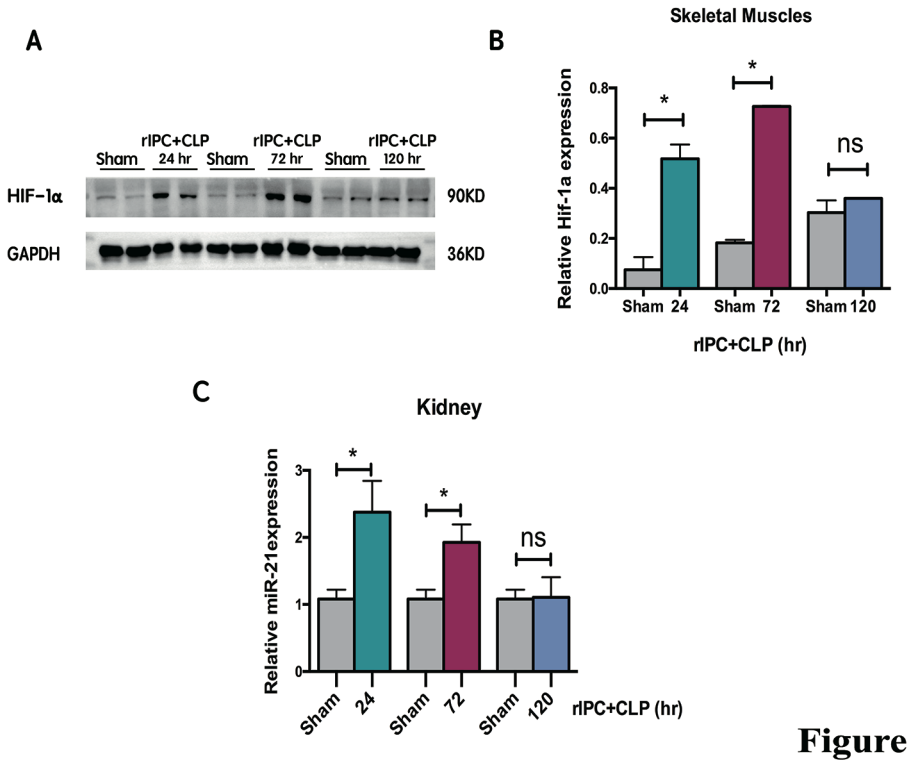

Figure S6.

Figure S6. The duration of the protective effectors from limb rIPC. We collected skeletal muscle tissues and renal tissues at 24, 72 and 120 h post rIPC+CLP. A and B, Immunoblot analysis of HIF-1a expression at different time points. (n = 6). C, Detection of miR-21 levels during the observed period in murine kidneys post rIPC+CLP (n = 6). *p < 0.05, and **p < 0.01. The data are presented as the mean ± SEM.

Table. Clinical information for volunteers

Patient NO.	Sex	Date of Birth
1	Male	1991-02-25
2	Female	1987-03-07
3	Male	1981-04-20
4	Female	1993-09-20
5	Male	1995-07-31
6	Female	1990-12-20

ALL the blood samples collected from the contralateral arm