EFFECT OF MACROPHAGE TLR4/NF-KB PATHWAY ON RHABODMYOLYSIS-INDUCED ACUTE KIDNEY INJURY

Rongshuang Huang, Liang Ma, Ping Fu

Division of Nephrology, Kidney Research Institute, West China Hospital of Sichuan University, Chengdu, CHINA.

OBJECTIVES

Acute kidney injury (AKI) is the most common and life-threatening systemic complication of rhabdomyolysis. Inflammation plays an important role in the development of rhabdomyolysis-induced AKI. In the study, we investigated the kidney model of AKI caused by rhabdomyolysis to verify the role of macrophage TLR4/NF-κB signaling pathway.

METHODS

The mice were injected by 50% glycerin solution to double leg muscle to induce rhabdomyolysis, and CLI-095 or PDTC were intraperitoneally injected at 0.5h before molding. The serum level of Scr, CK and the expression of TNF-α, IL-1β, IL-6, as well as H&E stainings of kidney tissues were tested. The infiltration of macrophage, mRNA level and protein expression of TLR4 and NF-κB were investigated by immunofluorescence double staining technique, RT-qPCR and western blot, respectively. In vitro, macrophage RAW264.7 was stimulated by ferrous myoglobin, and the cytokines, TLR4 and NF-κB expression were also detected.

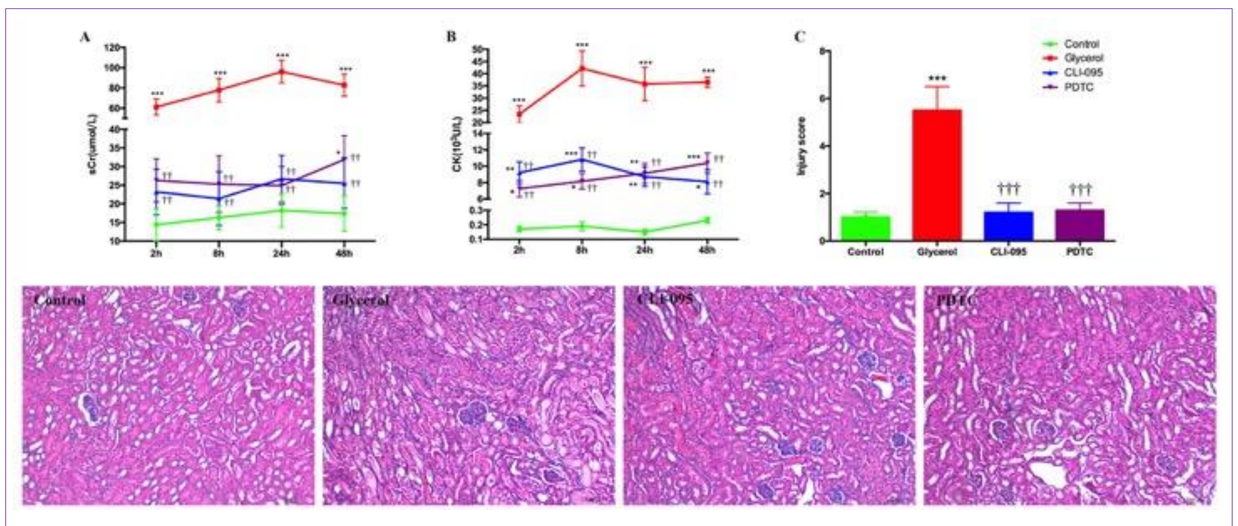

Figure1. TLR4 inhibitor CLI-095 and NF-κB inhibitor PTDC improved rhabdomyolysis-induced acute kidney dysfunction.

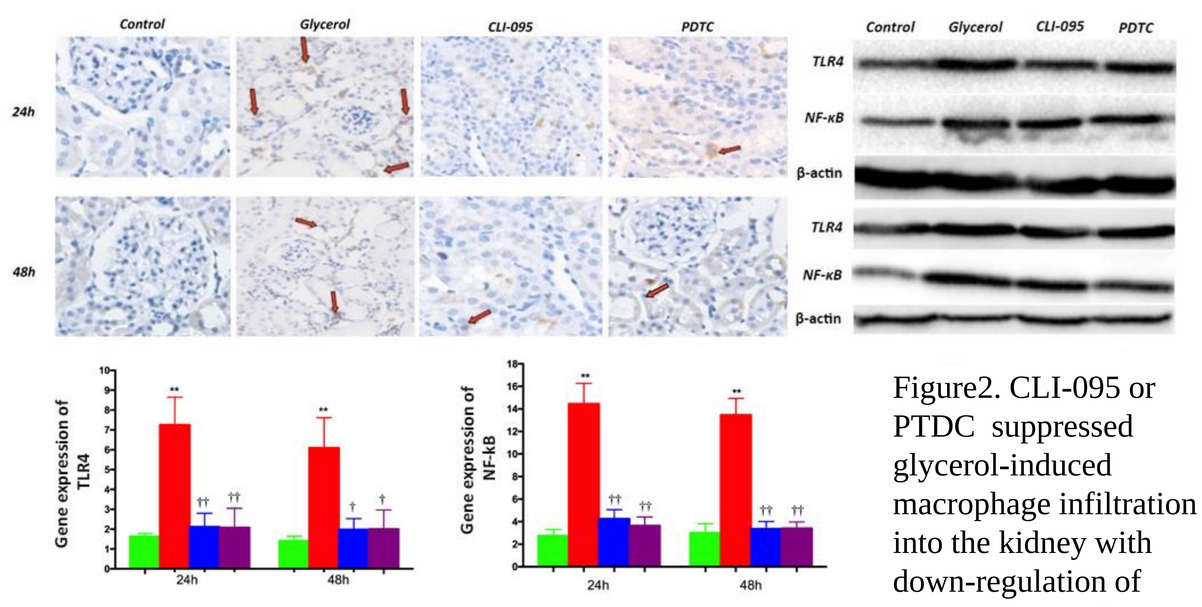

Figure2. CLI-095 or PTDC suppressed glycerol-induced macrophage infiltration into the kidney with down-regulation of TLR4 and NF-κB expression.

RESULTS

The animal model was built successfully with increased level of Scr and CK. The expression of TNF-α, IL-1β, and IL-6 in glycerol group was higher than in control group, CLI-095 and PDTC groups. Comparing with control group and blocker groups, the morphology and structure of kidney tissue were damaged more severely and the renal tubular Paller's score was higher in glycerol group. The infiltration of macrophage increased in glycerol group. The expression of TLR4 and NF-κB in rhabdomyolysis also increased. The mRNA and protein levels of TLR4 and NF-κB were higher in glycerol group than in control group and blocker groups. In intro, in ferrous myoglobin-treated RAW264.7 cell, the level of TNF-α, IL-1β and IL-6 were higher than other groups. The level of TLR4 and NF-κB mRNA, and TLR4 protein and NF-κB nuclear translocator were upregulated. The TLR4 and NF-κB blockers suppressed expression of TLR4 and NF-κB mRNA and TLR4 protein and NF-κB nuclear translocator.

CONCLUSIONS

In rhabdomyolysis-induced AKI, we found the infiltration of macrophages, along with increased TLR4 expression and NF-κB nuclear translocation, which mediated inflammatory response. These results may provide a new method to treat AKI.

ACKNOWLEDGEMENTS

This study was supported by Natural Science Foundation of China (No. 81570668) and Sichuan Science and Technology Support Program (2015SZ0135).

REFERENCES:

1. Panizo N, et al. Molecular Mechanisms and Novel Therapeutic Approaches to Rhabdomyolysis-Induced Acute Kidney Injury. Kidney Blood Press Res 2015;40:520-532.
2. Belliere J, et al. Specific macrophage subtypes influence the progression of rhabdomyolysis-induced kidney injury. J Am Soc Nephrol 2015; 26: 1363-77.
3. Gluba A, et al. The role of Toll-like receptors in renal diseases. Nat Rev Nephrol 2010; 6: 224-35.
4. Feng YY, et al. Rhabdomyolysis induced AKI via the regulation of endoplasmic reticulum stress and oxidative stress in PTECs. Rsc Adv 2016; 6: 109639-48.
5. Subeq YM, et al. Pentobarbital reduces rhabdomyolysis-induced acute renal failure in conscious rats. J Trauma 2009; 67: 132-8.

DOI: 10.3252/pso.eu.54ERA.2017