中南大学学报(医学版) 2010, 35(1) 77-84 DOI: ISSN: 1672-7347 CN: 43-1427/R

论著

冬虫夏草提取液对NRK-52E缺血再灌注损伤时凋亡及TLR-4表达的干预作用

周巧玲1， 胡赛2

1.中南大学湘雅医院肾内科， 长沙 410008；2.长沙市第四医院急诊科， 长沙 410007

摘要：

目的：观察体外缺血再灌注损伤（IRI）时大鼠肾小管上皮细胞（NRK-52E）凋亡情况和Toll样受体4（TLR-4）基因表达，以及冬虫夏草提取液对其干预作用。方法：将体外培养的NRK-52E分成对照组、模型组和冬虫夏草干预组，均以去除培养基后经10-10 mol/L抗霉素A刺激1 h后恢复培养基的方法，模拟IRI过程。在多个时间点，以流式细胞仪检测凋亡率， RT-PCR法检测凋亡相关基因（Bax，Bcl-2）和TLR-4基因的表达变化。结果：与对照组相比，模型组细胞凋亡率明显增高(P＜0.01)，Bax mRNA表达上调(P＜0.05)，Bcl-2 mRNA表达下调(P＜0.05)以及TLR-4 mRNA表达上调(P＜0.05)。冬虫夏草干预可明显影响缺血再灌注损伤时Bax，Bcl-2的表达，降低凋亡水平，并能减轻TLR-4高表达程度(均P＜0.05)。结论：NRK-52E 在IRI时有明显的细胞凋亡，以及TLR-4基因表达异常。冬虫夏草提取液可能通过抗凋亡和影响TLR-4表达减轻肾IRI。

关键词： 缺血再灌注损伤 凋亡 Toll样受体4 冬虫夏草提取液

Effect of Cordyceps Cinensis extractant on apoptosis and expression of Toll-like receptor 4 mRNA in the ischemia-reperfusion injured NRK-52E cells

ZHOU Qiaoling1, HU Sai2

1.Department of Nephrology, Xiangya Hospital, Central South University, Changsha 410008;
2.Department of Emergency Medicine, The 4th Hospital of Changsha, Changsha 410007，China

Abstract:

ObjectiveTo observe the apoptosis and expression of Toll-like receptor (TLR)-4’s mRNA in ischemia-reperfusion injured rat renal tubular epithelia cells (NRK-52E) in vitro, and effect of Cordyceps Sinensis (CS) extractant.MethodsCultured NRK-52E cells were divided into a control group, a model group, and a CS preincubated group. All first removed media and incubated with antimycin A for 1 h, and then recovered the media to simulate the ischemia-reperfusion injury in vitro. We detected the apoptosis ratio of cells by flow cytometer and the mRNA expression of TLR-4, Bax, Bcl-2 gene by reverse transcription-polymerase chain reaction at different time points.ResultsIn ischemia-reperfusion injured NRK-52E, the apoptosis ratio rose as time passed (P＜0.05). We also observed increased mRNA expression of TLR-4, Bax (P＜0.05) and deceased expression of Bcl-2 (P＜0.05). Compared with the model grpup, the CS preincubated NRK-52E cells showed apparent tolerance to ischemia-reperfusion injury, which manifestated lower apoptosis ratio (P＜0.05), decreased expression of mRNA of TLR-4, Bax and increased expression of Bcl-2 (All Ps＜0.05). ConclusionThe number of apoptosis cells and the expression of TLR-4 mRNA inceased with ischemia-reperfusion injury of NRK-52E in vitro. CS can prevent the NRK-52E cells from ischemia-reperfusion injury by downregulating TLR-4 gene.

Keywords: ischemia-reperfusion injury;apoptosis;Toll like receptor-4;Cordyceps Sinensis extractant

收稿日期 2009-05-23 修回日期 网络版发布日期

DOI:

基金项目:

通讯作者: 周巧玲

作者简介:

作者Email: zhouqling@yahoo.com.cn

参考文献：

［1］Devarajan P.Update on mechanisms of ischemic acute kidney injury［J］. J Am Soc Nephrol, 2006,17(6):1503-1520.

［2］Padanilam B J. Cell death induced by acute renal injury: a perspective on the contributions of apoptosis and necrosis［J］.Am J Physiol Renal Physiol, 2003, 284 (4): F608-F627.
［3］Akira S. Toll-like receptor signaling［J］. J Biol Chem, 2003, 278(10):38105-38108.
［4］Tanaka T, Nangaku M, Miyata T, et al. Blockade of calcium influx through L-type calcium channels attenuates mitochondrial injury and apoptosis in hypoxic renal tubular cells［J］.J Am Soc Nephrol, 2004,15(9):2320-2333.
［5］Elmore S. Apoptosis: a review of programmed cell death［J］, Toxicol Pathol, 2007, 35(4): 495-516.
［6］Suen D F, Norris K L, Youle R J. Mitochondrial dynamics and apoptosis［J］. Genes Dev, 2008, 22 (6): 1577-1590.
［7］Gustafsson A B, Gottlieb R A. Bcl-2 family members and apoptosis, taken to heart［J］. Am J Physiol Cell Physiol, 2007, 292(1):C45-C51.
［8］Brunelle J K, Letai A, Control of mitochondrial apoptosis by the Bcl-2 family［J］. J Cell Sci, 2009, 122(2): 437-441.
［9］Oyama J, Blais C, Liu X,et al. Reduced myocardial ischemia-reperfusion injury in Toll-like receptor 4-deficient mice［J］. Circulation, 2004, 109(2): 784-789.
［10］Shimamoto A, Pohlman T H, Shomura S,et al. Toll-like receptor 4 mediates lung ischemia-reperfusion injury［J］. Ann Thorac Surg, 2006, 82(12): 2017-2023.
［11］Caso J R, Pradillo J M, Hurtado O,et al. Toll-like receptor 4 is involved in brain damage and inflammation after experimental stroke［J］. Circulation, 2007, 115(3): 1599-1608.
［12］Peng Y, Gong J P, Liu C A, et al. Expression of toll-like receptor 4 and MD-2 gene and protein in Kupffer cells after ischemia-reperfusion in rat liver graft［J］. World J Gastroenterol, 2004, 10(19): 2890-2893.
［13］崔敏，郭啸华，李恒，等.百令提取液对环孢素A导致肾小管损伤的防治作用［J］.肾脏病与透析肾移植杂志，2004,13(1):44-48.
CUI Min, GUO Xiaohua, LI Heng, et al. Cordyceps Sinensis extractant protect against cyclosporine induced injury of tubular epithelium cell［J］. Chin J Nephrol Dial Transp,2004,13(1):44-48.
［14］Shahed A R, Kim S I, Shoskes D A. Down-regulation of apoptotic and inflammatory genes by Cordyceps sinensis extract in rat kidney following ischemia/reperfusion［J］. Transplant Proc,2001, 33 (6):2986-2987.

本刊中的类似文章

1．冉珂1，段开明2，邹定全1，李志坚1，金丽艳1，常业恬1.异氟醚预处理延迟相对兔心肌缺血再灌注损伤的保护作用[J]. 中南大学学报(医学版), 2008,33(02): 146-150

2．李杰群，齐海智，贺志军，胡伟，司中州，李一宁．[异丙肾对肝脏热缺血再灌注损伤的保护作用[J]．中南大学学报(医学版)，2006,31(05)：792-796

3．周杰斌1,2,杨学抗2, 叶启发1,明英姿1,夏宗江1.银杏叶提取物在大鼠肝移植缺血再灌注损伤中的预处理效用[J]. 中南大学学报(医学版), 2007,32(01): 54-58

4．叶治， 郭曲练， 王锣， 石敏， 潘韫丹 .七氟醚预处理对大鼠局灶性脑缺血再灌注损伤的延迟性保护作用[J]. 中南大学学报(医学版), 2009,34(02): 152-157

5．叶治， 郭曲练， 王锣 .七氟醚迟发性预处理对大鼠局灶性脑缺血损伤后NF-κB表达的影响[J]. 中南大学学报(医学版), 2010,35(3): 262-266

6．贺志飚1， 彭再梅1, 金丽艳2， 徐军美2， 柴湘平1.吗啡预处理对大鼠缺血再灌注损伤心肌线粒体通透性转换孔的影响[J]. 中南大学学报(医学版), 2010,35(8): 800-

7．胡尔滨； 蒋海河；.还原型谷胱甘肽预处理对体外循环肺缺血再灌注损伤的保护作用[J]. 中南大学学报(医学版), 2003,28(6): 619-

8．张航， 刘思祎， 王昕， 张春芳.钙通道激动剂BayK8644在兔肺缺血再灌注损伤中的保护作用[J]. 中南大学学报(医学版), 2010,35(10): 1091-

9．张位星； 龚光甫； 陈胜喜；.吸入NO对体外循环术中肺缺血再灌注损伤的保护作用[J]. 中南大学学报(医学版), 2000,25(5): 483-