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论著

骨活素在急性环孢素中毒SD大鼠肾组织中的表达及其作用机制

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摘要：

目的:探讨骨活素(osteoactivin, OA)在急性环孢素(cyclosporine A,CsA)中毒SD大鼠肾组织中的表达及其作用机制。方法:在正常饮食1周后随机将SD大鼠分为3组:试验组(CsA＋橄榄油灌胃)、载体对照组(橄榄油灌胃)和空白对照组(生理盐水灌胃)。各组分别于灌药后2 d、1周、2周分批处死,测定血肌酐(serum creatinine,SCr)及体质量变化;HE染色评判肾组织病理学改变;免疫组织化学检测肾组织OA的着染情况;Western印迹检测OA蛋白的表达;RT-PCR检测*OA*、基质金属蛋白酶-13(matrix metalloproteinase-13, *MMP-13*)、Ⅲ型胶原(collagen type Ⅲ, Col Ⅲ)mRNA的表达。 结果:试验组SD大鼠的体质量和SCr在灌胃2 d和1周末与对照组相比差异无统计学意义(*P*>0.05),2周时体质量较对照组显著减轻(*P*<0.001),SCr显著升高(*P*<0.001)。试验组SD大鼠病理组织学改变主要为炎性细胞浸润、肾小管上皮细胞、间质细胞空泡变性及肾小管上皮细胞坏死脱落等。免疫组织化学显示试验组SD大鼠肾组织内OA的表达随CsA灌胃时间的延长而增加,其表达部位主要位于肾小管上皮细胞及间质细胞,对照组基本不表达。RT-PCR显示试验组*OA*, *MMP-13* 及 Col Ⅲ mRNA的相对表达量随着CsA灌胃时间的延长而增加。Western 免疫印迹检测显示,OA蛋白在空白对照组及载体对照组基本不表达,随着试验组CsA灌胃时间的延长,OA蛋白的表达增强。结论:OA在SD大鼠急性CsA肾中毒模型早、中期有表达且较SCr敏感,表达部位主要位于肾小管上皮细胞及间质细胞;在肾小管上皮细胞内早期反应性表达上调的OA可能通过激活MMPs的表达和胶原蛋白的重构从而在诱发肾间质纤维化中发挥关键作用。

关键词： 骨活素 急性环孢素中毒 基质金属蛋白酶-13 Ⅲ型胶原

Expression and mechanism of osteoactivin in the kidney of SD rats after acute cyclosporine A toxicity

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Abstract:

Objective To determine the expression and mechanism of osteoactivin (OA) in the kidney by establishing SD rat model of acute cyclosporine A (CsA) toxicity. Methods SD rats were fed with normal diet for a week, which they were then randomly divided into 3 groups: an experimental group (gavage with cycloporin A and olive oil), a vector group (gavage with olive oil), and a control group (gavage with normal saline). SD rats were killed 2 days, 1 week, or 2 weeks after the gavage to examine the serum creatinine (SCr) and body weight. HE staining was used to detect the kidney histopathological change. Immunohistochemistry was used to observe the staining degree and area of OA. Western blot was used to detect the OA protein.The mRNA expressions of the *OA*, matrix metalloproteinase-13(*MMP-13*), and collagen type Ⅲ(Col Ⅲ) were examined by RT-PCR. Results The body weight and SCr of the rats in the experimental group 1 week and 2 days after the gavage had no significant difference compared with the vector group or the control group (*P*>0.05).On the end of 2nd week, the rats' body weight was significantly reduced, and SCr significantly increased compared with the vector group or the control group (*P*<0.001).The main histopathological changes in the experimental group were inflammatory cell infiltration, vacuolar degeneration of interstitial cells, or tubular epithelial cell necrosis. Intense OA expression located in the tubular epithelium and interstitial fibroblasts in the kidney of the experimental group was observed by immunohistochemistry. After CsA gavage, the relative mRNA expressions of *OA*, *MMP-13* , and *Col* Ⅲ significantly increased with time. Western blot did not find the expression of OA protein in the control and the vector group, which increased with time in the experimental group. Conclusion OA expresses in the kidney of SD rats after acute CsA toxicity and mainly expresses in the tubular epithelial cells and renal interstitium. OA is more sensitive to the damage of kidney tissue caused by CsA than by SCr. The early-phase up-regulation of OA expression in the tubular epithelium in response to renal injury caused by acute CsA toxicity might play a key role in triggering the renal interstitial fibrosis via activating expression of MMPs and collagen remodeling in SD rats.

Keywords: osteoactivin acute cyclosporine A toxicity matrix metalloproteinase-13 collagen type Ⅲ

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