[1]杨星,张丰深,孙海,等.胆囊收缩素对人胆管癌QBC939细胞分泌MMPs、TIMPs水平的影响[J].第三军医大学学报,2014,36(16):1684-1688.
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胆囊收缩素对人胆管癌QBC_{939}细胞分泌MMPs、TIMPs水平的影响

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Title: Effect of cholecystokinin on expression of MMPs and TIMPs in human cholangiocarcinoma QBC_{939} cells

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关键词：胆囊收缩素；基质金属蛋白酶；胆管癌；周围神经浸润

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摘要：目的　观察胆囊收缩素（cholecystokinin, CCK）对人胆管癌QBC_{939}细胞基质金属蛋白酶（mtrix metalloproteinases, MMPs）及其内源性组织抑制剂(tissue inhibitors of matrix metalloproteinases, TIMPs)表达的影响，阐明CCK促进胆管癌周围神经浸润（perineural invasion, PNI）可能的分子机制。　方法　将人胆管癌QBC_{939}细胞分为对照组与实验组。对照组为未经处理的胆管癌细胞QBC_{939}；实验1～8组分别加入10^{-13}、10^{-11}、10^{-9}、10^{-7} mol/L CCK_{8s}，10^{-7} mol/L L18（CCK-A受体拮抗剂）、10^{-7} mol/L L60（CCK-B受体拮抗剂）、10^{-7} mol/L CCK_{8s} +10^{-7} mol/L L18、10^{-7} mol/L CCK_{8s} +10^{-7} mol/L L60的QBC_{939}，孵育48 h，采用Transwell细胞侵袭实验比较各组QBC_{939}细胞侵袭能力的差异，并用Real-time PCR和Western blot检测各组MMP-2、MMP-9、TIMP-1、TIMP-2 mRNA和蛋白表达情况。各组间比较采用方差分析，采用Spearman法进行相关性检验。　结果　CCK_{8s}能够明显促进QBC_{939}细胞的侵袭能力，L18及L60均能明显减弱CCK_{8s}对QBC_{939}细胞的作用。与对照组相比，实验1～4组MMP-2及MMP-9 mRNA和蛋白的表达水平明显升高（P<0.01），TIMP-1及TIMP-2 mRNA和蛋白的表达水平明显降低（P<0.01）；实验5～6组MMP-2及MMP-9 mRNA和蛋白的表达水平明显降低（P<0.01），TIMP-1及TIMP-2 mRNA和蛋白的表达水平明显升高（P<0.01）；实验7～8组MMPs及TIMPs mRNA和蛋白表达水平与对照组比较差异无统

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计学意义（$P>0.05$）。实验1～4组MMP-2 mRNA和蛋白的表达水平与CCK浓度呈正相关（相关系数r分别为0.972、0.963，$P<0.01$），而TIMP-2 mRNA和蛋白的表达水平与CCK浓度呈负相关（相关系数r分别为-0.974、-0.952，$P<0.01$）。MMP-9、TIMP-1的变化与CCK浓度间无明显相关性；实验组5与实验组6比较、实验组7与实验组8比较，MMP-2、MMP-9、TIMP-1及TIMP-2 mRNA和蛋白表达水平差异无统计学意义（$P>0.05$）。

结论　CCK能明显促进QBC_{939}细胞侵袭能力及MMPs的表达、降低TIMPs的表达，可能主要通过影响MMP-2、TIMP-2平衡的途径增加胆管癌细胞的侵袭能力从而促进胆管癌的PNI。

Abstract: Objective To investigate the effect of cholecystokinin (CCK) on the expression of matrix metalloproteinases (MMPs) and tissue inhibitors of matrix metalloproteinases (TIMPs) in human cholangiocarcinoma QBC_{939} cells.

Methods Human cholangiocarcinoma QBC_{939} cells were assigned into a control group and 8 experimental groups. The experimental groups (group 1 to group 8) were treated with 10^{-13}, 10^{-11}, 10^{-9}, 10^{-7} and 10^{-7} mol/L L18 (a CCK-A receptor antagonist), 10^{-7} mol/L L60 (a CCK-B receptor antagonist), 10^{-7} mol/L CCK_{8s} + 10^{-7} mol/L L18, and 10^{-7} mol/L CCK_{8s} + 10^{-7} mol/L L60, respectively, for 48 h. Transwell Matrigel invasion assay was used to study the impact on the invasion ability of QBC_{939} cells. The mRNA and protein expression of MMP-2, MMP-9, TIMP-1 and TIMP-2 were detected by real-time PCR and Western blotting. All the data were analyzed using the analysis of variance and Spearman rank correlation.

Results CCK_{8s} could promote invasion ability of QBC_{939} cells significantly. The mRNA and protein expression of MMP-2 and MMP-9 in experimental groups 1 to 4 were significantly higher than those of the control group ($P<0.01$), and the mRNA and protein expression of TIMP-1 and TIMP-2 were significantly lower ($P<0.01$). The mRNA and protein expressions of MMP-2 and MMP-9 in experimental groups 5 and 6 were obviously lower than those of the control group ($P<0.01$), and the expression of TIMP-1 and TIMP-2 were obviously higher ($P<0.01$). There was no significant difference in the mRNA and protein expression of MMPs and TIMPs among experimental groups 7 and 8 and the control group. The mRNA and protein expression of MMP-2 in experimental groups 1 to 4 were positively correlated with the concentrations of CCK (r=0.972, 0.963; $P<0.01$), while those of TIMP-2 were negatively correlated with the concentrations of CCK (r=-0.974, -0.952; $P<0.01$). Conclusion CCK can significantly promote the invasion ability of QBC_{939} cells and the expression of MMPs, but decrease the expression of TIMPs in QBC_{939} cells. CCK may promote perineural invasion of cholangiocarcinoma mainly through impacting the balance between MMP-2 and TIMP-2.

参考文献/References:

杨星, 张丰深, 孙海, 等. 胆囊收缩素对人胆管癌QBC_{939}细胞分泌MMPs、TIMPs水平的影响[J].第三军医大学学报,2014,36(16):1684-1688.

相似文献/References:

[1]王春莉,梅虎,谢静,等.共培养下后交叉韧带成纤维细胞中MMPs的基因表达[J].第三军医大学学报,2012,34(16):1600.
Wang Chunli,Mei Hu,Xie Jing,et al.Matrix metalloproteinase expression in posterior cruciate ligament fibroblasts co-cultured with synovial cells[J].J Third Mil Med Univ,2012,34(16):1600.
[2]丁志祥,刘小琴,蒋辉.肝癌患者血清血管内皮细胞生长因子和基质金属蛋白酶-9的水平变化及其意义[J].第三军医大学学报,2005,27(21):2176.
[3]王海涛,刘昊,颜京霞,等.N-乙酰-L-半胱氨酸对染矽尘大鼠肺间质成纤维细胞MMP-2表达的影响[J].第三军医大学学报,2005,27(19):2003.

[4]黄刚,汤为学,陶小红.生长抑素对大鼠胰腺腺泡细胞内骨架蛋白F-actin分布和细胞外分泌功能的影响[J].第三军医大学学报,2010,32(07):672.

Huang Gang,Tang Weixue,Tao Xiaohong.Effect of somatostatin on F-actin distribution and exocrine function in rat pancreatic acinar cells[J].J Third Mil Med Univ,2010,32(16):672.

[5]焦海霞,王萍,马腾,等.强力霉素抑制MMP-9对大鼠局灶性缺血再灌注脑损伤的保护作用[J].第三军医大学学报,2010,32(24):2591.

Jiao Haixia,Wang Ping,Ma Teng,et al.Doxycycline-mediated inhibition of MMP-9 protects focal cerebral ischemia-reperfusion injury in rats[J].J Third Mil Med Univ,2010,32(16):2591.

[6]肖华,季爱民,李志梁,等.氧化低密度脂蛋白对大鼠心肌成纤维细胞胶原的影响[J].第三军医大学学报,2008,30(13):1252.

XIAO Hua,JI Ai-min,LI Zhi-liang,et al.Effects of oxidized low density lipoprotein ox-LDL on collagen synthesis and degradation in cultured cardiac fibroblasts[J].J Third Mil Med Univ,2008,30(16):1252.

[7]邓雪松,王立.HIF-1α516对人肝癌HepG2细胞侵袭转移能力的影响[J].第三军医大学学报,2011,33(02):169.

Deng Xuesong,Wang Li.Silencing HIF-1α516 inhibits invasive ability of HepG2 cells[J].J Third Mil Med Univ,2011,33(16):169.

[8]张涛,陈晓品,汤为学,等.MMP-2、MMP-9在高、低转移潜能乳腺癌细胞中的表达变化[J].第三军医大学学报,2011,33(06):600.

Zhang Tao,Chen Xiaopin,Tang Weixue,et al.Expressions of matrix metalloprotainese-2 and metalloprotainese-9 in human breast cancer cell line MDA-MB-435s with high- and low-metastatic potentiality[J].J Third Mil Med Univ,2011,33(16):600.

[9]倪志宇,丛斌,董玫,等.八肽胆囊收缩素对脂多糖诱导RAW 264.7细胞IL-1β、IL-6、IL-10表达的影响[J].第三军医大学学报,2011,33(22):2366.

Ni Zhiyu,Cong Bin,Dong Mei,et al.Effect of cholecystokinin octapeptide on IL-1β, IL-6 and IL-10 expression in LPS-induced RAW 264.7 cells[J].J Third Mil Med Univ,2011,33(16):2366.

[10]赵军,王宏伟,宋继伟,等.PTEN基因对胶质瘤细胞增殖及侵袭性影响的体外研究[J].第三军医大学学报,2012,34(06):542.

Zhao Jun,Wang Hongwei,Song Jiwei,et al.Effect of PTEN gene transfection on growth and invasion in human