WCJD 世界华人消化杂志®

在线投稿: http://www.baishideng.com/wcjd/ch/index.aspx
帮助平台: http://www.wjgnet.com/esps/helpdesk.aspx
DOI: 10.11569/wcjd.v23.i25.4051

世界华人消化杂志 2015年9月8日; 23(25): 4051-4059
ISSN 1009-3079 (print) ISSN 2219-2859 (online)
© 2015年版权归百世登出版集团有限公司所有.

文献综述 REVIEW

EMT相关分子标志物在胆管癌侵袭转移中的研究进展

强 勇, 陈 钟

强勇, 陈钟, 南通大学附属医院肝胆外科 江苏省南通市 226001
强勇, 在读硕士, 主要从事肝胆肿瘤的临床与基础研究.
江苏省"科教兴卫工程"医学领军人才和创新团队项目, No. LJ201134
作者贡献分布: 本文综述由强勇完成; 文献资料收集由强勇完成; 陈钟审校.
通讯作者: 陈钟, 教授, 主任医师, 226001, 江苏省南通市西寺路20号, 南通大学附属医院肝胆外科. chenz9806@163.com
电话: 0513-81161006
收稿日期: 2015-06-20 修回日期: 2015-07-26
接受日期: 2015-07-30 在线出版日期: 2015-09-08

Epithelial mesenchymal transition related molecular markers and invasion and metastasis of cholangiocarcinoma

Yong Qiang, Zhong Chen

Yong Qiang, Zhong Chen, Department of Hepatobiliary Surgery, Affiliated Hospital of Nantong University, Nantong 226001, Jiangsu Province, China
Supported by: Jiangsu Province Outstanding Medical Academic Leader and Innovation Team Program, No. LJ201134
Correspondence to: Zhong Chen, Professor, Chief Physician, Department of Hepatobiliary Surgery, Affiliated Hospital of Nantong University, 20 Xisi Road, Nantong 226001, Jiangsu Province, China. chenz9806@163.com
Received: 2015-06-20 Revised: 2015-07-26
Accepted: 2015-07-30 Published online: 2015-09-08

Abstract

Tumor metastasis is a major cause of death in patients with solid tumors. Epithelial mesenchymal transition (EMT) is a process in which the epithelial cells are transformed into the stroma cells. This process is accompanied by changes in gene expression and cell phenotype, which are often activated during tumor invasion and metastasis. Cholangiocarcinoma is a kind of malignancy originating from the bile duct epithelium, and its main biological characteristics are early invasion, metastasis and recurrence. The research of cholangiocarcinoma metastasis could provide a theoretical basis for the development of new treatment strategies to manage this malignancy. This paper reviews the roles of EMT related molecular markers metastasis in the invasion and metastasis of cholangiocarcinoma.

© 2015 Baishideng Publishing Group Inc. All rights reserved.

Key Words: Epithelial mesenchymal transition; Tumor metastasis; Cholangiocarcinoma; Molecular markers

Qiang Y, Chen Z. Epithelial mesenchymal transition related molecular markers and invasion and metastasis of cholangiocarcinoma. Shijie Huaren Xiaohua Zazhi 2015; 23(25): 4051-4059 URL: http://www.wjgnet.com/1009-3079/23/4051.asp DOI: http://dx.doi.org/10.11569/wcjd.v23.i25.4051

摘要

肿瘤转移是导致实体瘤患者死亡的主要原因. 上皮-间质转化(epithelial mesenchymal transition, EMT)是上皮细胞转化为间质细胞伴随基因和表型改变的过程, 在肿瘤发生侵袭和转移过程中常常被激活. 胆管癌是起源于胆道上皮的一种恶性肿瘤, 早期易浸润、转移和复发是其主要生物学特点. 研究胆管癌的转移机制能够为未来治疗胆管癌的新方法, 建立新的治疗策略提供理论依据. 本

背景资料
胆管癌早期症状隐匿, 浸润转移发生早, 发现时往往已处于疾病晚期或发生了广泛转移, 许多患者丧失了手术机会. 而目前除了手术, 其他治疗效果都不尽如人意. 介于胆管癌的生物学特性与上皮-间质转化(epithelial mesenchymal transition, EMT)关系密切, 故研究EMT在胆管癌的转移机制能够为胆管癌的诊治提供新方法.

同行评议者
刘金钢, 教授, 中国医科大学附属盛京医院外科

研发前沿
EMT相关的分子标志物在胆管癌的诊断、治疗中的应用和研究受到国内外专家的广泛关注, 是胆管癌领域的研究热点.

文综述了近年来EMT相关标志物, 包括细胞表型分子标志物及EMT相关调控因子与胆管癌浸润转移的研究进展.

© 2015年版权归百世登出版集团有限公司所有.

关键词: 上皮-间质转化; 肿瘤转移; 胆管癌; 生物标志物

核心提示: 胆管癌是起源于肝内或肝外胆管上皮细胞的一种相对少见的恶性肿瘤, 其恶性程度高、预后差. 近年来发现, 上皮-间质转化(epithelial mesenchymal transition, EMT)在胆管癌的侵袭迁移过程中发挥重要作用. 因此, 研究EMT在胆管癌的转移机制能够为治疗胆管癌的提供新方法, 同时为建立新的治疗策略提供理论依据.

强勇, 陈钧. EMT相关分子标志物在胆管癌侵袭转移中的研究进展. 世界华人消化杂志 2015; 23(25): 4051-4059 URL: http://www.wjgnet.com/1009-3079/23/4051.asp DOI: http://dx.doi.org/10.11569/wcjd.v23.i25.4051

0 引言

胆管癌是起源于肝内或肝外胆管上皮细胞的一种相对少见的恶性肿瘤[1]. 占全部胃肠道恶性肿瘤的3%[2]. 过去30年来世界范围内胆管癌的发病率逐渐升高, 5年生存率不足10%[3]. 由于对放化疗不敏感, 根治性手术仍是目前唯一有效的治疗手段[4]. 胆管癌早期症状隐匿, 浸润转移发生早, 诊断时常处于晚期或已发生广泛转移, 丧失了手术机会; 几乎一半未经治疗的患者在3-4 mo内死于胆道梗阻、胆管感染、肝功能衰竭等并发症[5]. 而手术患者往往在2年内出现局部复发和远处转移, 预后很差[6].

近年来研究[7,8]表明上皮-间质转化(epithelial mesenchymal transition, EMT)与许多上皮源性肿瘤包括胆管癌的侵袭和转移密切相关. EMT是一个复杂、瞬时、可逆的生物学过程, 使肿瘤上皮细胞丧失极性, 黏附表型转化到间叶表型从而增加细胞侵袭和迁移能力, 具有细胞骨架重塑和抗凋亡的特性. 其中应力纤维的重新排列, 黏附分子的改变和细胞外基质(extracellular matrix, ECM)蛋白是肿瘤细胞发生EMT的关键[9]. 肿瘤细胞发生EMT除了形态学和生物学行为会发生改变外, 还有一些与EMT相关的分子标志物如细胞表面蛋白、细胞骨架蛋白、ECM蛋白、转录因子及microRNAs(miRNAs, miRs)的表达和/或功能会发生改变[10]. 本文对当前有关EMT及胆管癌的文献进行回顾, 就EMT相关分子标志物与胆管癌侵袭转移的关系作一综述.

1 细胞表面蛋白

1.1 钙黏素超家族 上皮细胞的特点是细胞间接触和黏附连接形成牢固. 钙黏素超家族是一类Ca^{2+}依耐的跨膜糖蛋白, 由E-cadherin、N-cadherin、P-cadherin组成, 参与细胞间黏附和连接. 上皮细胞间的黏附连接和接触主要是依赖E-cadherin通过α或β-catenin连接到肌动蛋白丝,从而维持细胞间的稳定性和细胞的极性. E-cadherin表达缺失是EMT发生并导致肿瘤侵袭和转移的标志性变化. 当发生EMT时常伴E-cadherin表达缺失, β-catenin从细胞膜转移到细胞核内, 使上皮细胞间的黏附减弱, 发生从良性非侵袭性向恶性侵袭性表型的转化[11]. 现已证实[12]在胆管癌中E-cadherin有着类似的改变, 而且与淋巴结转移, 总生存率和无病生存期明显相关. siRNA E-cadherin转染到胆管癌细胞中导致间叶标志物Vimentin蛋白上调, E-cadherin/β-catenin黏附复合物从细胞膜消失, 细胞的侵袭和迁移能力增强[13]. 总之, 上述研究表明E-cadherin是肿瘤细胞表型的主要调节因子, 也是胆管癌发生转移的标志.

P-cadherin最先在老鼠胎盘中发现[14], 在人类胎盘中并没有检测到其表达, 但在一些器官比如乳腺、前列腺中确有表达[15]. P-cadherin同E-cadherin一样与catenin蛋白连接保持上皮细胞间的黏附. 但在癌细胞迁移中P-cadherin的角色却有争议, 研究[15,16]已证实在卵巢癌和胰腺癌中是促进细胞的迁移. 相反, 在乳腺癌中却抑制乳腺上皮细胞的迁移[17]. 相关研究[18]发现*P-cadherin*在胆管癌组织和发育异常的胆管组织中往往表达高, 是一个癌基因. 虽然Obama等[19]在对肝内胆管癌进行全基因组分析认为P-cadherin的表达与淋巴结转移, 远处转移无关, 但这仅是在对23例肝内胆管癌组织进行免疫组织化学检查分析得出的结果. 而Baek等[20]研究认为P-cadherin下调虽然不能促进胆管癌上皮癌细胞细胞(HuCCT-1)的增殖, 但细胞的侵袭和迁移却明显降低; 此外, 其他EMT标志物(Snail1、Snail2和

Vimentin)没有发生改变, 这说明P-cadherin在胆管癌EMT中是一个独立调节因子.

在EMT中E-cadherin表达的下调往往伴随N-cadherin表达上调, 这种不同钙黏素表达水平的变化, 称为钙黏素转换, 近年来已经逐渐用来监测癌症进展中的EMT[21]. N-cadherin是另一个黏附分子, 形成嗜血细胞间黏附连接. N-cadherin通常在神经组织、血管内皮细胞及骨骼和心肌细胞中表达, 定位在片状伪足和丝状伪足上[20]. N-cadherin还在许多肿瘤细胞中表达, 能提高癌细胞的迁移和侵袭能力且不受E-cadherin的影响. 并且许多报道[22,23]N-cadherin在癌症转移中较E-cadherin和其他EMT诱导物更重要, 但Mosnier等[24]发现N-cadherin与肝内和肝门部胆管癌的许多临床病理参数无关. 值得注意的是Araki等[25]在肝外胆管癌的研究中发现N-cadherin表达增加和E-cadherin表达降低明显相关, 并且这个钙黏素转换可被转化生长因子-β(transforming growth factor-β, TGF-β)在转录水平调节, 促进肿瘤的进展, 是一个强有力的预后因素. 这表明不管肝外胆管癌中N-cadherin的含量如何, 其对肿瘤进展都具有重要作用.

1.2 syndecan-1 syndecan-1是一个细胞表面硫酸乙酰肝素蛋白多糖, 参与细胞间的凝聚, 调节细胞基质间黏附和控制生长因子信号. 主要在上皮细胞中表达, 调节众多生物过程与肿瘤进展有关[26,27], 是生长因子、血管生成因子、成形素、趋化因子经典的共同受体, 也是生长发育和癌症进展中EMT的一个相关标志物. Harada等[28]的实验证实了syndecan-1在肝内胆管癌中非癌胆道上皮细胞基侧膜上表达, 而在肝内胆管癌细胞膜和胞浆中都有表达. 表明肝内胆管癌中syndecan-1的表达在转录水平被调节. syndecan-1在肝内胆管癌中表达与肿瘤分化程度有关, 高分化的肝内胆管癌组织中syndecan-1广泛表达并且强烈. 阴性或弱表达常常与淋巴结转移和手术患者的预后不良相关, 而与肿瘤大小、间质结缔组织生成、大体分型、血管和神经侵犯相关. 总之syndecan-1是一个有用的预后标志.

2 细胞骨架蛋白

2.1 Vimentin蛋白 Vimentin蛋白在各种间叶细胞中都表达, 包括纤维母细胞、血管内皮细胞、造血系统细胞和胶质细胞. 通常在上皮细胞中没有表达, 但在一些上皮源性肿瘤细胞中却发现也有表达. Vimentin蛋白表达与肿瘤细胞的侵袭和转移能力增加正相关[29], 因此通常用来确定肿瘤细胞EMT的发生, 并作为间叶细胞表型的一个常用标志[9,30]. 研究[31]发现Vimentin蛋白可能是通过C-src调节E-cadherin/β-catenin复合物来促进肿瘤细胞的侵袭和转移. Vimentin蛋白在胆管癌中的表达具有一定临床的意义. Huang等[12]对140例肝内胆管癌组织芯片进行免疫组织化学检查, 其中55.7%癌组织中Vimentin蛋白表达增加, 并且与淋巴结转移、总生存率和无瘤生存率低下相关. Mao等[32]相关实验发现Vimentin蛋白与肿瘤分化、静脉侵犯和肝炎病毒感染相关. 在多因素分析中证实Vimentin蛋白过表达是胆管癌的一个独立预后因素.

2.2 β-catenin蛋白 β-catenin蛋白是一个细胞质斑蛋白, 在EMT中扮演了双重角色: 连接钙黏素和细胞骨架, 以保持正常的细胞结构及与T细胞因子(T-cell factor, TCF)/淋巴增强因子(lymphoid enhancing factor, LEF)共同组成转录调节的激活剂启动EMT. β-catenin蛋白的活动主要经胞浆内其水平控制来调节, 也通过自身补充钙黏素复合物或泛素化剂随后的降解来调节. β-catenin/TCF/LEF复合体直接控制EMT相关基因的表达, 特别是*Snail1*. 在胚胎发育、癌症和纤维化的各种研究中β-catenin蛋白已经作为EMT的标志物. 虽然β-catenin蛋白位于正常上皮细胞和非侵袭性肿瘤细胞的细胞膜上, 但是在发生EMT后β-catenin蛋白位于胞浆中或细胞核中. β-catenin蛋白在胆管癌侵袭和转移中有着一定的作用. β-catenin蛋白减少与肝内胆管癌分化程度低下、肿瘤大小和淋巴结转移明显相关[33]. Gu等[34]发现β-catenin蛋白在肝内胆管癌患者中下调32/83(38.6%), 其膜表达减弱倾向于与血管侵犯相关, 在男性患者中发生率更高, E-cadherin膜表达减少明显与β-catenin蛋白相关. 因此认为β-catenin对肿瘤分化和进展的影响超过了对肿瘤增殖和侵袭的影响. Abuetabh等[35]证实了β-catenin蛋白在2种不同的胆管癌细胞系HuCCT1和OZ中具有明显不同的表达模式, 在转移能力强的细胞株OZ中其仅在细胞膜表达, 而在具有中等侵袭性的胆管细胞株HuCCT1中其在细胞浆中表达.

相关报道
钙黏素超家族是一类Ca^{2+}依赖的跨膜糖蛋白, 而E-cadherin表达缺失是EMT典型的标志性变化, 也是促进EMT发生导致肿瘤侵袭和转移的先决条件.

创新盘点

本文综述了近年来EMT相关标志物, 包括细胞表型分子标志物及EMT相关调控因子与胆管癌浸润转移的研究进展. 明确了EMT过程在胆管癌侵袭和迁移中起重要作用.

2.3 FSP1肌动蛋白 肌动蛋白重构对EMT的进展非常重要[36,37]. 成纤维细胞特异蛋白FSP1(S100A4)是Ca^{2+}结合100家族成员之一, 在间叶细胞、巨噬细胞和发生间叶转化的上皮细胞中表达[38], 是癌症及纤维增生中检测EMT典型的成纤维细胞标志, 调节多种细胞的运动[39-41]. FSP1异位表达自身促进上皮细胞和癌细胞的EMT进程[40]. 在对86例胆管癌的临床资料进行分析发现没有S100A4核表达的平均存活时间是5.4年, 核表达弱阳性<30%是1.38年, ≥30%平均存活时间进一步下降为0.77年, 表明S100A4核表达明显降低了生存. 而敲低S100A4后, 发现CK19没有改变, 但抑制了细胞的迁移, 分泌MMP-9和侵入到ECM, 对增殖和细胞凋亡的活力没有影响; 这表明S100A4在胆管癌的侵袭中是一个机械因素, 他不仅是胆管癌发生EMT的标志, 还是胆管癌一个潜在的分子治疗靶点[42].

2.4 α-平滑肌肌动蛋白 α-平滑肌肌动蛋白(α-smooth muscle actin, α-SMA)是六个肌动蛋白家族成员之一, 在成人的血管平滑肌和肌上皮细胞中表达明显. α-SMA阳性的肌成纤维细胞对促进癌症的发生和发展具有重要的作用[43]. 在许多实体肿瘤的研究中已将α-SMA作为肌成纤维细胞表达的一个标志. 在胆管癌的研究中, Chuaysri等[44]研究证实α-SMA表达与肿瘤大小和生存明显相关. 在胆管癌基质成纤维细胞中α-SMA高表达可能是患者存活时间短的一个预后指标. 胆管癌成纤维细胞增生可能直接影响胆道上皮细胞肿瘤的发生和进展.

3 ECM蛋白

胶原蛋白、层黏连蛋白、巢蛋白和硫酸化蛋白聚糖是基底膜的主要组成成分. 层黏连蛋白是其中最确定的EMT标志物, 在EMT时下调[9]. 层黏连蛋白5(α3β3γ2)对上皮细胞与基底膜的黏附具有重要的作用, 与上皮细胞恶性转化和侵袭转移相关[45]. 层黏连蛋白5作为α3β1和α6β4整合素的配体, 在多种癌细胞中广泛表达, 包括胆管癌[46]. 与整合素结合导致细胞骨架重塑促进细胞的迁移[47], 通过磷脂酰肌醇3-激酶活化提高肿瘤细胞的侵袭和促进MMP-9产生并降解ECM, 促进肿瘤细胞的侵袭和转移[48]. 层黏连蛋白γ2只能由层黏连蛋白5产生, 是一种ECM蛋白, 对肿瘤细胞的迁移和侵袭有重要的作用[49]. 在肝内胆管癌中层黏连蛋白γ2的高表达与肿瘤恶变和预后不良明显相关, 层黏连蛋白γ2在导管内型、管周浸润型和肿块型3种不同类型的肝内胆管癌中分别呈基底膜染色、胞浆染色和基质染色的阶梯式表达[50]. 肝外胆管癌组织中层黏连蛋白γ2阳性染色有2种类型, 其中基质和胞浆染色都与淋巴结转移明显相关. 体外实验进一步验证了敲低层黏连蛋白γ2表达, 明显降低了QBC939细胞的侵袭和迁移能力[51].

4 转录因子

E-cadherin表达缺失是EMT的标志. 目前认为对E-cadherin表达下调的主要潜在机制是基因的突变、启动子甲基化和转录抑制. 其中, 转录抑制是最重要的调控机制. E-cadherin受到多种转录因子和miRNAs的调控[52], 其中Slug、Snail和Twist这3个是公认与EMT相关的转录因子, 这些转录因子能够识别位于E-cadherin转录起始位点的E-box DNA序列, 在那里他们补充辅因子和组蛋白去乙酰酶[7], 在转录抑制E-cadherin表达途径中起着关键性作用, 充当控制EMT过程分子开关的功能.

4.1 Slug与Snail Snail是被发现的第一个E-cadherin转录抑制物, 也是EMT中最重要的转录因子之一. Snail除了具有抑制E-cadherin功能外, Snail还下调其他上皮分子的表达, 包括封闭蛋白、闭合蛋白和角蛋白, 诱导间叶细胞和侵袭表型相关基因的表达[9]. 在肝门部胆管癌中E-cadherin下调的患者Slug mRNA水平明显高, Slug在癌组织和癌旁组织的mRNA比值明显与淋巴结转移、远处转移和生存时间有关, 但有趣的是Snail mRNA既不与E-cadherin表达相关也不和肿瘤侵袭力相关. siRNA干扰Slug表达后, E-cadherin表达上调, 降低了QBC939细胞的侵袭. 这表明胆管癌中Slug的表达与临床分期、侵袭、复发及远处转移有关. 在肝内胆管癌中Slug很少表达, Snail过表达预示着生存时间和无病存活时间短. Snail过表达与E-cadherin下调, Vimentin蛋白上调有关. 在RBE细胞中抑制Slug的表达, E-cadherin表达减少, Vimentin蛋白表达增加, 细胞的侵袭和迁移能力受到抑制. 这表明在肝内胆管癌EMT中Snail是主要的调节因子, 而不是Slug[53].

4.2 Twist蛋白 Twist是一个碱性螺旋-环-螺旋蛋白. 在早期胚胎成形, 组织纤维化和肿瘤转移中被下调[9]. 参与EMT过程, 在癌症转移中具有至关重要的作用[54]. Twist能够被多个信号转导通路激活, 包括Akt信号转导和转录激活因子3, 丝裂原活化蛋白激酶Ras和Wnt信号通路[55]. Twist能够不依赖Snail独立抑制E-cadherin的表达, 上调纤连蛋白和N-cadherin的表达[56]. Twist在胆管癌组织中表达上调, 其表达与胆管癌患者预后负相关, 而且Twist核表达样本中N-cadherin明显上调[57]. 已有研究[58]证实Twist可作为miR-214的直接作用靶点促进EMT. 而miR-214在调节肝内胆管癌的转移中具有重要的作用.

4.3 Smad7 Samds信号通路是涉及EMT过程的一个重要信号通路[59]. 转录调节因子Smad7能够抑制TGF-β, 在TGF-β/Smads通路扮演反馈调节作用来维持平衡[60]. 最近研究[61,62]已经证实Smad7具有促癌作用. 相关研究[63]发现, 在胆管癌组织中Smad7表达阳性率达68.3%, 明显高于邻近癌旁组织的表达, 而且在淋巴结转移, 神经浸润的胆管癌组织中表达明显增高, 其阳性表达与E-cadherin减少和Vimentin蛋白的增加明显相关; Smad7阴性表达患者总生存率和无病生存率较高. 因此认为Smad7在胆管癌侵袭转移发生EMT中是一个末期的反馈调节因子, 可作为临床上用于评估预后的一个指标.

4.4 FOXC2 FOXC2是另外一种转录因子充当EMT多效性诱导物. 在胚胎发育期间FOXC2广泛表达, 对血管、肌肉生成及多种器官发育都非常重要. 出生后, FOXC2通常只在脂肪细胞表达[64], 而在乳腺导管癌, 转移性乳腺癌细胞株中其也有表达[65]. EMT诱导物TGF-β1、Snail、Twist、Goosecoid中任何一种都能增加FOXC2的表达, 其本身超表达也能诱导EMT, 这表明FOXC2在EMT中具有重要作用[65]. FOXC2已经被报道对肿瘤细胞发生EMT[66]、促进肿瘤的血管生成[67]和多种癌症进展中具有重要的作用. FOXC2作为一个促癌因子, 在肝外胆管癌中高表达, 与淋巴管浸润和淋巴结转移明显相关. 敲低FOXC2表达后发现N-cadherin、Vimentin蛋白、MMP-2和Ang-2(一种淋巴管生成因子)明显增加, 然而E-cadherin却明显降低; 说明FOXC2在肝外胆管癌中调节MMP-2引发侵袭, 并通过Ang-2应答对淋巴管浸润和淋巴结转移的作用. 相比之下, TGF-β1和Snail不变, 而体外实验显示敲低FOXC2可抑制细胞的侵袭和迁移, 但对细胞增殖无明显影响; 表明FOXC2位于EMT诱导物(TGFβ1和Snail)的下游, 在肝外胆管癌中是通过调节N-cadherin和MMP-2来参与EMT应答的[68].

5 miRNAs

miRNAs是一种内源性非编码20-22个核苷酸的RNA, 以碱基互补配对的方式与信使RNA(mRNA)的3'端非翻译区(3' untranslated region, 3'UTR)相互作用导致mRNA降解和/或翻译抑制[69]. miRNAs几乎在每个生物过程调节中都发挥了重要的作用, 包括细胞增殖、凋亡、分化和迁移[70]. 同时, 大量的证据表明在多种癌症中miRNAs表达异常, 包括胆管癌. 许多miRNAs已被报道在肝内胆管癌中表达失调[58,71,72], 如miR-21、miR-124和miR-214等. miRNAs除了作为抑癌基因或促癌基因与肿瘤的进展相关外, 还可充当EMT重要的调节因子[73]. miR-200家族和miR-205已经被认定直接针对ZEB1、SIP1或NCAM1抑制EMT, 从而降低癌细胞的侵袭性[74,75].

miR-214在有转移的肝内胆管癌组织较无转移的癌组织表达明显减少. 在体外实验[58]中, 抑制miR-214的表达促进了人肝内胆管癌细胞的转移, 同时下调miR-214增加了EMT相关基因*Twist*的转录水平, 降低了E-cadherin的水平; 可认为下调miR-214直接作用于*Twist*基因促进了胆管癌EMT, 对调节肝内胆管癌的转移具有重要的作用.

肝内胆管癌中miR-204往往下调, 其低表达明显与淋巴结转移相关. miR-204过表达明显抑制了肝内胆管癌细胞的侵袭和迁移以及EMT过程. Slug为miR-204的直接靶点, 在HuH28细胞株中下调miR-204后, 上皮标记E-cadherin表达增加, 间叶标记vimentin蛋白表达减少, 发生了EMT逆转. 因此, miR-204可通过直接抑制Slug的表达来抑制肝内胆管癌的侵袭和或转移[76].

6 结论

鉴于EMT在肿瘤侵袭和转移过程中的重要性, 本文综述了近年来EMT相关标志物在胆管癌

■应用要点

研究EMT相关标志物在胆管癌的作用及细胞表型分子标志物和EMT相关调控因子与胆管癌浸润转移的关系, 有助于进一步探讨其在胆管癌发生发展中的作用, 为肿瘤转移的早期诊断、预后评估开辟新的视野, 并且有希望为肿瘤治疗提供新的靶点方向.

名词解释
EMT: 是上皮细胞转化为间质细胞伴随基因和表型改变的过程, 在肿瘤发生侵袭和转移过程中常常被激活.

的研究现状, 着重讨论了细胞表型分子标志物及EMT相关调控因子与胆管癌浸润转移的关系. 由于体内发生侵袭转移的肿瘤细胞只占原发肿瘤的很小一部分, 当他们发生EMT时的一些基因、转录、蛋白水平的改变往往为原发肿瘤所掩盖. 而且肿瘤细胞除了可以发生EMT外, 还可发生间质上皮转化, 两者的发生是一个动态、连续、可逆的过程, 并受到肿瘤微环境的影响. 因此, 如何确认和捕捉发生EMT的肿瘤细胞是一个难题. 而且在不同状态下的肿瘤细胞EMT的表达模式也不尽相同. 因此, 这些分子标志物在判断体内EMT的发生上仍具有一定的局限性. 未来仍需深入研究和寻找与EMT发生密切相关的特异性生物标志物来鉴定体内肿瘤EMT的发生, 进一步探讨其在胆管癌发生发展中的作用, 为早期诊断、预后评估开辟新视野, 并且有希望为胆管癌治疗提供新的靶点方向.

7 参考文献

1 Augustine MM, Fong Y. Epidemiology and risk factors of biliary tract and primary liver tumors. *Surg Oncol Clin N Am* 2014; 23: 171-188 [PMID: 24560105 DOI: 10.1016/j.soc.2013.10.001]
2 Gatto M, Bragazzi MC, Semeraro R, Napoli C, Gentile R, Torrice A, Gaudio E, Alvaro D. Cholangiocarcinoma: update and future perspectives. *Dig Liver Dis* 2010; 42: 253-260 [PMID: 20097142 DOI: 10.1016/j.dld.2009.12.008]
3 Khan SA, Davidson BR, Goldin RD, Heaton N, Karani J, Pereira SP, Rosenberg WM, Tait P, Taylor-Robinson SD, Thillainayagam AV, Thomas HC, Wasan H. Guidelines for the diagnosis and treatment of cholangiocarcinoma: an update. *Gut* 2012; 61: 1657-1669 [PMID: 22895392 DOI: 10.1136/gutjnl-2011-301748]
4 Murakami Y, Uemura K, Sudo T, Hashimoto Y, Nakashima A, Kondo N, Sakabe R, Ohge H, Sueda T. Prognostic factors after surgical resection for intrahepatic, hilar, and distal cholangiocarcinoma. *Ann Surg Oncol* 2011; 18: 651-658 [PMID: 20945107 DOI: 10.1245/s10434-010-1325-4]
5 Patel T. Cholangiocarcinoma--controversies and challenges. *Nat Rev Gastroenterol Hepatol* 2011; 8: 189-200 [PMID: 21460876 DOI: 10.1038/nrgastro.2011.20]
6 van der Gaag NA, Kloek JJ, de Bakker JK, Musters B, Geskus RB, Busch OR, Bosma A, Gouma DJ, van Gulik TM. Survival analysis and prognostic nomogram for patients undergoing resection of extrahepatic cholangiocarcinoma. *Ann Oncol* 2012; 23: 2642-2649 [PMID: 22532585 DOI: 10.1093/annonc/mds077]
7 Tsai JH, Yang J. Epithelial-mesenchymal plasticity in carcinoma metastasis. *Genes Dev* 2013; 27: 2192-2206 [PMID: 24142872 DOI: 10.1101/gad.225334.113]
8 Nitta T, Mitsuhashi T, Hatanaka Y, Miyamoto M, Oba K, Tsuchikawa T, Suzuki Y, Hatanaka KC, Hirano S, Matsuno Y. Prognostic significance of epithelial-mesenchymal transition-related markers in extrahepatic cholangiocarcinoma: comprehensive immunohistochemical study using a tissue microarray. *Br J Cancer* 2014; 111: 1363-1372 [PMID: 25077440 DOI: 10.1038/bjc.2014.415]
9 Zeisberg M, Neilson EG. Biomarkers for epithelial-mesenchymal transitions. *J Clin Invest* 2009; 119: 1429-1437 [PMID: 19487819 DOI: 10.1172/jci36183]
10 Creighton CJ, Gibbons DL, Kurie JM. The role of epithelial-mesenchymal transition programming in invasion and metastasis: a clinical perspective. *Cancer Manag Res* 2013; 5: 187-195 [PMID: 23986650 DOI: 10.2147/cmar.s35171]
11 Berx G, van Roy F. Involvement of members of the cadherin superfamily in cancer. *Cold Spring Harb Perspect Biol* 2009; 1: a003129 [PMID: 20457567 DOI: 10.1101/cshperspect.a003129]
12 Huang XY, Zhang C, Cai JB, Shi GM, Ke AW, Dong ZR, Zhang PF, Fan J, Peng BG, Zhou J. Comprehensive multiple molecular profile of epithelial mesenchymal transition in intrahepatic cholangiocarcinoma patients. *PLoS One* 2014; 9: e96860 [PMID: 24816558 DOI: 10.1371/journal.pone.0096860]
13 Techasen A, Loilome W, Namwat N, Khuntikeo N, Puapairoj A, Jearanaikoon P, Saya H, Yongvanit P. Loss of E-cadherin promotes migration and invasion of cholangiocarcinoma cells and serves as a potential marker of metastasis. *Tumour Biol* 2014; 35: 8645-8652 [PMID: 24867095 DOI: 10.1007/s13277-014-2087-6]
14 Nose A, Takeichi M. A novel cadherin cell adhesion molecule: its expression patterns associated with implantation and organogenesis of mouse embryos. *J Cell Biol* 1986; 103: 2649-2658 [PMID: 3539943]
15 Taniuchi K, Nakagawa H, Hosokawa M, Nakamura T, Eguchi H, Ohigashi H, Ishikawa O, Katagiri T, Nakamura Y. Overexpressed P-cadherin/CDH3 promotes motility of pancreatic cancer cells by interacting with p120ctn and activating rho-family GTPases. *Cancer Res* 2005; 65: 3092-3099 [PMID: 15833838 DOI: 10.1158/0008.5472.can-04-3646]
16 Cheung LW, Leung PC, Wong AS. Cadherin switching and activation of p120 catenin signaling are mediators of gonadotropin-releasing hormone to promote tumor cell migration and invasion in ovarian cancer. *Oncogene* 2010; 29: 2427-2440 [PMID: 20118984 DOI: 10.1038/onc.2009.523]
17 Simpson KJ, Selfors LM, Bui J, Reynolds A, Leake D, Khvorova A, Brugge JS. Identification of genes that regulate epithelial cell migration using an siRNA screening approach. *Nat Cell Biol* 2008; 10: 1027-1038 [PMID: 19160483 DOI: 10.1038/ncb1762]
18 Riener MO, Vogetseder A, Pestalozzi BC, Clavien PA, Probst-Hensch N, Kristiansen G, Jochum W.

Cell adhesion molecules P-cadherin and CD24 are markers for carcinoma and dysplasia in the biliary tract. *Hum Pathol* 2010; 41: 1558-1565 [PMID: 20621328 DOI: 10.1016/j.humpath.2009.12.016]
19 Obama K, Ura K, Li M, Katagiri T, Tsunoda T, Nomura A, Satoh S, Nakamura Y, Furukawa Y. Genome-wide analysis of gene expression in human intrahepatic cholangiocarcinoma. *Hepatology* 2005; 41: 1339-1348 [PMID: 15880566 DOI: 10.1002/hep.20718]
20 Baek S, Lee YW, Yoon S, Baek SY, Kim BS, Oh SO. CDH3/P-Cadherin regulates migration of HuCCT1 cholangiocarcinoma cells. *Anat Cell Biol* 2010; 43: 110-117 [PMID: 21189991 DOI: 10.5115/acb.2010.43.2.110]
21 Smith A, Teknos TN, Pan Q. Epithelial to mesenchymal transition in head and neck squamous cell carcinoma. *Oral Oncol* 2013; 49: 287-292 [PMID: 23182398 DOI: 10.1016/j.oraloncology.2012.10.009]
22 Nakajima S, Doi R, Toyoda E, Tsuji S, Wada M, Koizumi M, Tulachan SS, Ito D, Kami K, Mori T, Kawaguchi Y, Fujimoto K, Hosotani R, Imamura M. N-cadherin expression and epithelial-mesenchymal transition in pancreatic carcinoma. *Clin Cancer Res* 2004; 10: 4125-4133 [PMID: 15217949 DOI: 10.1158/1078-0432.ccr-0578-03]
23 Islam S, Carey TE, Wolf GT, Wheelock MJ, Johnson KR. Expression of N-cadherin by human squamous carcinoma cells induces a scattered fibroblastic phenotype with disrupted cell-cell adhesion. *J Cell Biol* 1996; 135: 1643-1654 [PMID: 8978829]
24 Mosnier JF, Kandel C, Cazals-Hatem D, Bou-Hanna C, Gournay J, Jarry A, Laboisse CL. N-cadherin serves as diagnostic biomarker in intrahepatic and perihilar cholangiocarcinomas. *Mod Pathol* 2009; 22: 182-190 [PMID: 18622386 DOI: 10.1038/modpathol.2008.123]
25 Araki K, Shimura T, Suzuki H, Tsutsumi S, Wada W, Yajima T, Kobayahi T, Kubo N, Kuwano H. E/N-cadherin switch mediates cancer progression via TGF-β-induced epithelial-to-mesenchymal transition in extrahepatic cholangiocarcinoma. *Br J Cancer* 2011; 105: 1885-1893 [PMID: 22068819 DOI: 10.1038/bjc.2011.452]
26 Yip GW, Smollich M, Götte M. Therapeutic value of glycosaminoglycans in cancer. *Mol Cancer Ther* 2006; 5: 2139-2148 [PMID: 16985046 DOI: 10.1158/1535-7163.mct-06-0082]
27 Ibrahim SA, Hassan H, Vilardo L, Kumar SK, Kumar AV, Kelsch R, Schneider C, Kiesel L, Eich HT, Zucchi I, Reinbold R, Greve B, Götte M. Syndecan-1 (CD138) modulates triple-negative breast cancer stem cell properties via regulation of LRP-6 and IL-6-mediated STAT3 signaling. *PLoS One* 2013; 8: e85737 [PMID: 24392029 DOI: 10.1371/journal.pone.0085737]
28 Harada K, Masuda S, Hirano M, Nakanuma Y. Reduced expression of syndecan-1 correlates with histologic dedifferentiation, lymph node metastasis, and poor prognosis in intrahepatic cholangiocarcinoma. *Hum Pathol* 2003; 34: 857-863 [PMID: 14562280]
29 McInroy L, Määttä A. Down-regulation of vimentin expression inhibits carcinoma cell migration and adhesion. *Biochem Biophys Res Commun* 2007; 360: 109-114 [PMID: 17585878 DOI: 10.1016/j.bbrc.2007.06.036]
30 Zhao J, Dong D, Sun L, Zhang G, Sun L. Prognostic significance of the epithelial-to-mesenchymal transition markers e-cadherin, vimentin and twist in bladder cancer. *Int Braz J Urol* 2014; 40: 179-189 [PMID: 24856504 DOI: 10.1590/S1677-5538.IBJU.2014.02.07]
31 Wei J, Xu G, Wu M, Zhang Y, Li Q, Liu P, Zhu T, Song A, Zhao L, Han Z, Chen G, Wang S, Meng L, Zhou J, Lu Y, Wang S, Ma D. Overexpression of vimentin contributes to prostate cancer invasion and metastasis via src regulation. *Anticancer Res* 2008; 28: 327-334 [PMID: 18383865]
32 Mao X, Chen D, Wu J, Li J, Zhou H, Wu Y, Duan X. Differential expression of fascin, E-cadherin and vimentin: Proteins associated with survival of cholangiocarcinoma patients. *Am J Med Sci* 2013; 346: 261-268 [PMID: 23221510 DOI: 10.1097/MAJ.0b013e3182707108]
33 Gu MJ, Choi JH. Epithelial-mesenchymal transition phenotypes are associated with patient survival in intrahepatic cholangiocarcinoma. *J Clin Pathol* 2014; 67: 229-234 [PMID: 24062361 DOI: 10.1136/jclinpath-2013-201806]
34 Gu MJ, Choi JH. Clinicopathological significance of E-cadherin, β-catenin and epidermal growth factor receptor expression in intrahepatic cholangiocarcinoma. *Hepatogastroenterology* 2012; 59: 1241-1244 [PMID: 22281980 DOI: 10.5754/hge11881]
35 Abuetabh Y, Persad S, Nagamori S, Huggins J, Al-Bahrani R, Sergi C. Expression of E-cadherin and β-catenin in two cholangiocarcinoma cell lines (OZ and HuCCT1) with different degree of invasiveness of the primary tumor. *Ann Clin Lab Sci* 2011; 41: 217-223 [PMID: 22075503]
36 Haynes J, Srivastava J, Madson N, Wittmann T, Barber DL. Dynamic actin remodeling during epithelial-mesenchymal transition depends on increased moesin expression. *Mol Biol Cell* 2011; 22: 4750-4764 [PMID: 22031288 DOI: 10.1091/mbc.E11-02-0119]
37 Liu SC, Jen YM, Jiang SS, Chang JL, Hsiung CA, Wang CH, Juang JL. G(alpha)12-mediated pathway promotes invasiveness of nasopharyngeal carcinoma by modulating actin cytoskeleton reorganization. *Cancer Res* 2009; 69: 6122-6130 [PMID: 19602597 DOI: 10.1158/0008-5472.can-08-3435]
38 Österreicher CH, Penz-Österreicher M, Grivennikov SI, Guma M, Koltsova EK, Datz C, Sasik R, Hardiman G, Karin M, Brenner DA. Fibroblast-specific protein 1 identifies an inflammatory subpopulation of macrophages in the liver. *Proc Natl Acad Sci U S A* 2011; 108: 308-313 [PMID: 21173249 DOI: 10.1073/pnas.1017547108]
39 Samejima K, Nakatani K, Suzuki D, Asai O, Sakan H, Yoshimoto S, Yamaguchi Y, Matsui M, Akai Y, Toyoda M, Iwano M, Saito Y. Clinical significance of fibroblast-specific protein-1 expression on podocytes in patients with focal segmental

同行评价

胆管癌易浸润转移, 其机制与EMT相关. 作者引用了大量文献, 介绍了多种胆管癌EMT相关分子标志物, 该综述内容较新, 对进一步研究有借鉴作用.

glomerulosclerosis. *Nephron Clin Pract* 2012; 120: c1-c7 [PMID: 22126861 DOI: 10.1159/000334184]
40 Xue C, Plieth D, Venkov C, Xu C, Neilson EG. The gatekeeper effect of epithelial-mesenchymal transition regulates the frequency of breast cancer metastasis. *Cancer Res* 2003; 63: 3386-3394 [PMID: 12810675]
41 Chen N, Sato D, Saiki Y, Sunamura M, Fukushige S, Horii A. S100A4 is frequently overexpressed in lung cancer cells and promotes cell growth and cell motility. *Biochem Biophys Res Commun* 2014; 447: 459-464 [PMID: 24732359 DOI: 10.1016/j.bbrc.2014.04.025]
42 Fabris L, Cadamuro M, Moserle L, Dziura J, Cong X, Sambado L, Nardo G, Sonzogni A, Colledan M, Furlanetto A, Bassi N, Massani M, Cillo U, Mescoli C, Indraccolo S, Rugge M, Okolicsanyi L, Strazzabosco M. Nuclear expression of S100A4 calcium-binding protein increases cholangiocarcinoma invasiveness and metastasization. *Hepatology* 2011; 54: 890-899 [PMID: 21618579 DOI: 10.1002/hep.24466]
43 Sirica AE, Gores GJ. Desmoplastic stroma and cholangiocarcinoma: clinical implications and therapeutic targeting. *Hepatology* 2014; 59: 2397-2402 [PMID: 24123296 DOI: 10.1002/hep.26762]
44 Chuaysri C, Thuwajit P, Paupairoj A, Chau-In S, Suthiphongchai T, Thuwajit C. Alpha-smooth muscle actin-positive fibroblasts promote biliary cell proliferation and correlate with poor survival in cholangiocarcinoma. *Oncol Rep* 2009; 21: 957-969 [PMID: 19287994]
45 Marinkovich MP. Tumour microenvironment: laminin 332 in squamous-cell carcinoma. *Nat Rev Cancer* 2007; 7: 370-380 [PMID: 17457303 DOI: 10.1038/nrc2089]
46 Bergamini C, Sgarra C, Trerotoli P, Lupo L, Azzariti A, Antonaci S, Giannelli G. Laminin-5 stimulates hepatocellular carcinoma growth through a different function of alpha6beta4 and alpha3beta1 integrins. *Hepatology* 2007; 46: 1801-1809 [PMID: 17948258 DOI: 10.1002/hep.21936]
47 Kariya Y, Miyazaki K. The basement membrane protein laminin-5 acts as a soluble cell motility factor. *Exp Cell Res* 2004; 297: 508-520 [PMID: 15212952 DOI: 10.1016/j.yexcr.2004.03.044]
48 Saito Y, Sekine W, Sano R, Komatsu S, Mizuno H, Katabami K, Shimada K, Oku T, Tsuji T. Potentiation of cell invasion and matrix metalloproteinase production by alpha3beta1 integrin-mediated adhesion of gastric carcinoma cells to laminin-5. *Clin Exp Metastasis* 2010; 27: 197-205 [PMID: 20352300 DOI: 10.1007/s10585-010-9314-3]
49 Tsubota Y, Ogawa T, Oyanagi J, Nagashima Y, Miyazaki K. Expression of laminin gamma2 chain monomer enhances invasive growth of human carcinoma cells in vivo. *Int J Cancer* 2010; 127: 2031-2041 [PMID: 20143393 DOI: 10.1002/ijc.25231]
50 Aishima S, Matsuura S, Terashi T, Taguchi K, Shimada M, Maehara Y, Tsuneyoshi M. Aberrant expression of laminin gamma 2 chain and its prognostic significance in intrahepatic cholangiocarcinoma according to growth morphology. *Mod Pathol* 2004; 17: 938-945 [PMID: 15105812 DOI: 10.1038/modpathol.3800143]
51 Liu W, Tian F, Jiang P, Zhao X, Guo F, Li X, Wang S. Aberrant expression of laminin γ2 correlates with poor prognosis and promotes invasion in extrahepatic cholangiocarcinoma. *J Surg Res* 2014; 186: 150-156 [PMID: 24124977 DOI: 10.1016/j.jss.2013.09.008]
52 Wang Y, Zhou BP. Epithelial-mesenchymal transition in breast cancer progression and metastasis. *Chin J Cancer* 2011; 30: 603-611 [PMID: 21880181 DOI: 10.5732/cjc.011.10226]
53 Zhang KJ, Wang DS, Zhang SY, Jiao XL, Li CW, Wang XS, Yu QC, Cui HN. The E-cadherin repressor slug and progression of human extrahepatic hilar cholangiocarcinoma. *J Exp Clin Cancer Res* 2010; 29: 88 [PMID: 20594328 DOI: 10.1186/1756-9966-29-88]
54 Zhang P, Hu P, Shen H, Yu J, Liu Q, Du J. Prognostic role of Twist or Snail in various carcinomas: a systematic review and meta-analysis. *Eur J Clin Invest* 2014; 44: 1072-1094 [PMID: 25257753 DOI: 10.1111/eci.12343]
55 Garg M. Epithelial-mesenchymal transition - activating transcription factors - multifunctional regulators in cancer. *World J Stem Cells* 2013; 5: 188-195 [PMID: 24179606 DOI: 10.4252/wjsc.v5.i4.188]
56 Yang Z, Zhang X, Gang H, Li X, Li Z, Wang T, Han J, Luo T, Wen F, Wu X. Up-regulation of gastric cancer cell invasion by Twist is accompanied by N-cadherin and fibronectin expression. *Biochem Biophys Res Commun* 2007; 358: 925-930 [PMID: 17512904 DOI: 10.1016/j.bbrc.2007.05.023]
57 Duangkumpha K, Techasen A, Loilome W, Namwat N, Thanan R, Khuntikeo N, Yongvanit P. BMP-7 blocks the effects of TGF-β-induced EMT in cholangiocarcinoma. *Tumour Biol* 2014; 35: 9667-9676 [PMID: 24969562 DOI: 10.1007/s13277-014-2246-9]
58 Li B, Han Q, Zhu Y, Yu Y, Wang J, Jiang X. Down-regulation of miR-214 contributes to intrahepatic cholangiocarcinoma metastasis by targeting Twist. *FEBS J* 2012; 279: 2393-2398 [PMID: 22540680 DOI: 10.1111/j.1742-4658.2012.08618.x]
59 Liu Q, Zhang Y, Mao H, Chen W, Luo N, Zhou Q, Chen W, Yu X. A crosstalk between the Smad and JNK signaling in the TGF-β-induced epithelial-mesenchymal transition in rat peritoneal mesothelial cells. *PLoS One* 2012; 7: e32009 [PMID: 22384127 DOI: 10.1371/journal.pone.0032009]
60 Drabsch Y, ten Dijke P. TGF-β signalling and its role in cancer progression and metastasis. *Cancer Metastasis Rev* 2012; 31: 553-568 [PMID: 22714591 DOI: 10.1007/s10555-012-9375-7]
61 Chen YK, Huang AH, Cheng PH, Yang SH, Lin LM. Overexpression of Smad proteins, especially Smad7, in oral epithelial dysplasias. *Clin Oral Investig* 2013; 17: 921-932 [PMID: 22669485 DOI: 10.1007/s00784-012-0756-7]
62 Zizi-Sermpetzoglou A, Myoteri D, Arkoumani E, Voultsos M, Marinis A. A study of Smad4 and

Smad7 expression in surgically resected samples of gastric adenocarcinoma and their correlation with clinicopathological parameters and patient survival. *J BUON* 2014; 19: 221-227 [PMID: 24659668]
63 Huang Q, Liu L, Liu CH, Shao F, Xie F, Zhang CH, Hu SY. Expression of Smad7 in cholangiocarcinoma: prognostic significance and implications for tumor metastasis. *Asian Pac J Cancer Prev* 2012; 13: 5161-5165 [PMID: 23244128]
64 Cederberg A, Grønning LM, Ahrén B, Taskén K, Carlsson P, Enerbäck S. FOXC2 is a winged helix gene that counteracts obesity, hypertriglyceridemia, and diet-induced insulin resistance. *Cell* 2001; 106: 563-573 [PMID: 11551504]
65 Mani SA, Yang J, Brooks M, Schwaninger G, Zhou A, Miura N, Kutok JL, Hartwell K, Richardson AL, Weinberg RA. Mesenchyme Forkhead 1 (FOXC2) plays a key role in metastasis and is associated with aggressive basal-like breast cancers. *Proc Natl Acad Sci U S A* 2007; 104: 10069-10074 [PMID: 17537911 DOI: 10.1073/pnas.0703900104]
66 Mortazavi F, An J, Dubinett S, Rettig M. p120-catenin is transcriptionally downregulated by FOXC2 in non-small cell lung cancer cells. *Mol Cancer Res* 2010; 8: 762-774 [PMID: 20460685 DOI: 10.1158/1541-7786.mcr-10-0004]
67 Kume T. The Role of FoxC2 Transcription Factor in Tumor Angiogenesis. *J Oncol* 2012; 2012: 204593 [PMID: 22174714 DOI: 10.1155/2012/204593]
68 Watanabe A, Suzuki H, Yokobori T, Altan B, Kubo N, Araki K, Wada S, Mochida Y, Sasaki S, Kashiwabara K, Hosouchi Y, Kuwano H. Forkhead box protein C2 contributes to invasion and metastasis of extrahepatic cholangiocarcinoma, resulting in a poor prognosis. *Cancer Sci* 2013; 104: 1427-1432 [PMID: 23919841 DOI: 10.1111/cas.12249]
69 Cho WC. MicroRNAs in cancer - from research to therapy. *Biochim Biophys Acta* 2010; 1805: 209-217 [PMID: 19931352 DOI: 10.1016/j.bbcan.2009.11.003]
70 Kong KL, Kwong DL, Chan TH, Law SY, Chen L, Li Y, Qin YR, Guan XY. MicroRNA-375 inhibits tumour growth and metastasis in oesophageal squamous cell carcinoma through repressing insulin-like growth factor 1 receptor. *Gut* 2012; 61: 33-42 [PMID: 21813472 DOI: 10.1136/gutjnl-2011-300178]
71 Karakatsanis A, Papaconstantinou I, Gazouli M, Lyberopoulou A, Polymeneas G, Voros D. Expression of microRNAs, miR-21, miR-31, miR-122, miR-145, miR-146a, miR-200c, miR-221, miR-222, and miR-223 in patients with hepatocellular carcinoma or intrahepatic cholangiocarcinoma and its prognostic significance. *Mol Carcinog* 2013; 52: 297-303 [PMID: 22213236 DOI: 10.1002/mc.21864]
72 Zeng B, Li Z, Chen R, Guo N, Zhou J, Zhou Q, Lin Q, Cheng D, Liao Q, Zheng L, Gong Y. Epigenetic regulation of miR-124 by hepatitis C virus core protein promotes migration and invasion of intrahepatic cholangiocarcinoma cells by targeting SMYD3. *FEBS Lett* 2012; 586: 3271-3278 [PMID: 22819820 DOI: 10.1016/j.febslet.2012.06.049]
73 Ding XM. MicroRNAs: regulators of cancer metastasis and epithelial-mesenchymal transition (EMT). *Chin J Cancer* 2014; 33: 140-147 [PMID: 24016392 DOI: 10.5732/cjc.013.10094]
74 Gregory PA, Bert AG, Paterson EL, Barry SC, Tsykin A, Farshid G, Vadas MA, Khew-Goodall Y, Goodall GJ. The miR-200 family and miR-205 regulate epithelial to mesenchymal transition by targeting ZEB1 and SIP1. *Nat Cell Biol* 2008; 10: 593-601 [PMID: 18376396 DOI: 10.1038/ncb1722]
75 Oishi N, Kumar MR, Roessler S, Ji J, Forgues M, Budhu A, Zhao X, Andersen JB, Ye QH, Jia HL, Qin LX, Yamashita T, Woo HG, Kim YJ, Kaneko S, Tang ZY, Thorgeirsson SS, Wang XW. Transcriptomic profiling reveals hepatic stem-like gene signatures and interplay of miR-200c and epithelial-mesenchymal transition in intrahepatic cholangiocarcinoma. *Hepatology* 2012; 56: 1792-1803 [PMID: 22707408 DOI: 10.1002/hep.25890]
76 Qiu YH, Wei YP, Shen NJ, Wang ZC, Kan T, Yu WL, Yi B, Zhang YJ. miR-204 inhibits epithelial to mesenchymal transition by targeting slug in intrahepatic cholangiocarcinoma cells. *Cell Physiol Biochem* 2013; 32: 1331-1341 [PMID: 24280681 DOI: 10.1159/000354531]

编辑: 郭鹏 电编: 都珍珍