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【基础研究】

假木豆提取物对四氯化碳致大鼠肝纤维化的保护作用

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摘要: **目的** 探讨假木豆提取物抗四氯化碳(CCl_4)所致大鼠肝纤维化的作用机制。**方法** 将60只Sprague Dawley大鼠随机分为空白组、模型组、水飞蓟素组、低剂量假木豆组、中剂量假木豆组和高剂量假木豆组,每组10只。水飞蓟素组、低剂量假木豆组、中剂量假木豆组、高剂量假木豆组和模型组大鼠通过背部皮下注射CCl_4建立肝纤维化模型,空白组大鼠背部皮下注射花生油,每隔2 d注射1次,共8周。从第3周起,水飞蓟素组大鼠每日灌服水飞蓟素胶囊水溶液(50 mg·kg^{-1})1次;假木豆低、中、高剂量组大鼠每日灌服12.5、25.0、50.0 g·L^{-1}的假木豆水提液(20 mL·kg^{-1});空白组和模型组大鼠灌服等体积的蒸馏水;各组大鼠均连续给药6周。观察并记录各组大鼠饮食活动情况、体质量、毛色光亮程度、精神状况等一般情况。末次给药1 h后采集各组大鼠腹主动脉血5 mL,采用酶联免疫吸附试验检测各组大鼠血清中白细胞介素(IL)-1β、IL-6、肿瘤坏死因子-α(TNF-α)和转化生长因子-β_1(TGF-β_1)水平。采血后处死大鼠,取肝脏,观察肝脏大体形态,然后取大鼠肝脏左叶,制备肝组织切片,行苏木精-伊红染色,于光学显微镜下观察大鼠肝脏病理形态学变化。**结果** 空白组大鼠饮食正常,活动频繁,毛色洁白有光泽,精神状况正常。模型组大鼠饮食减少,活动量明显减少,毛色暗淡、不清洁,精神状况萎靡;水飞蓟素组大鼠毛色欠缺光泽但较清洁,其余一般情况同空白组;假木豆各剂量组大鼠一般情况较模型组有改善,而较空白组差。模型组、水飞蓟素组、低剂量假木豆组、中剂量假木豆组、高剂量假木豆组大鼠体质量增长量少于空白组($P<0.05$),低剂量假木豆组大鼠体质量增长少于模型组($P<0.05$)。模型组、水飞蓟素组、低剂量假木豆组、中剂量假木豆组、高剂量假木豆组大鼠血清中IL-1β、IL-6、TNF-α水平均高于空白组($P<0.05$);水飞蓟素组与空白组大鼠血清中TGF-β_1水平比较差异无统计学意义($P>0.05$);模型组、低剂量假木豆组、中剂量假木豆组、高剂量假木豆组大鼠血清中TGF-β_1水平均高于空白组($P<0.05$)。水飞蓟素组、低剂量假木豆组、中剂量假木豆组、高剂量假木豆组大鼠血清中IL-1β、IL-6、TNF-α、TGF-β_1水平均低于模型组($P<0.05$)。低剂量假木豆组大鼠血清中IL-1β、IL-6、TNF-α、TGF-β_1水平显著高于水飞蓟素组($P<0.05$)。中剂量假木豆组与水飞蓟素组大鼠血清中IL-1β、IL-6水平比较差异无统计学意义($P>0.05$),中剂量假木豆组大鼠血清中TNF-α、TGF-β_1水平显著高于水飞蓟素组($P<0.05$);高剂量假木豆组与水飞蓟素组大鼠血清中IL-1β、IL-6、TNF-α、TGF-β_1水平比较差异无统计学意义($P>0.05$)。低、中、高剂量假木豆组大鼠血清中IL-1β水平比较差异均无统计学意义($P>0.05$)。高剂量假木豆组大鼠血清中IL-6、TGF-β_1水平显著低于低剂量假木豆组($P<0.05$);中剂量假木豆组与低剂量假木豆组、高剂量假木豆组与中剂量假木豆组大鼠血清中IL-6、TGF-β_1水平比较差异均无统计学意义($P>0.05$)。低、中、高剂量假木豆组大鼠血清中TNF-α水平随剂量的增加逐渐降低,两两比较差异均有统计学意义($P<0.05$)。空白组大鼠肝组织结构完整,肝小叶结构正常;模型组大鼠肝组织肝纤维化明显;低剂量假木豆组大鼠肝组织病理学形态较模型组有轻微改善;水飞蓟素组和中、高剂量假木豆组大鼠肝组织病理学形态较模型组有较大的改善,肝纤维形成减轻或消失。**结论** 假木豆提取物对大鼠肝纤维化有保护作用,其作用机制可能与降低血清中IL-1β、IL-6、TNF-α及TGF-β_1水平有关。

关键词: 假木豆提取物;肝纤维化;白细胞介素-1β;白细胞介素-6;肿瘤坏死因子-α;转化生长因子-β_1

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Protective effect of *Jiamudou* extract on hepatic fibrosis induced by carbon tetrachloride

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Abstract: **Objective** To explore the mechanism of *Jiamudou* extract anti-hepatic fibrosis induced by carbon tetrachloride (CCl_4) in rats. **Methods** Sixty Sprague Dawley rats were randomly divided into blank group, model group, silymarin group, low-dose *Jiamudou* group, middle-dose *Jiamudou* group and high-dose *Jiamudou* group, with 10 rats in each group. The rats in the model group, silymarin group, low-dose *Jiamudou* group, middle-dose *Jiamudou* group and high-dose *Jiamudou* group were

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injected subcutaneously with CCl_4 every 2 days for 8 weeks to establish the liver fibrosis model; the rats in the blank group were injected subcutaneously with peanut oil every 2 days for 8 weeks. From the third week, the rats in the silymarin group were given silymarin capsule aqueous solution (50 mg · kg^{-1}), once a day; the rats in the low-dose *Jiamudou* group, middle-dose *Jiamudou* group and high-dose *Jiamudou* group were given 12.5, 25.0, 50.0 g · L^{-1} of *Jiamudou* water extract (20 mL · kg^{-1}) by gavage, respectively; the rats in the blank group and model group were given equal volume of distilled water; all rats were continuously administered for 6 weeks. The general conditions such as diet and activity, body weight, hair color brightness and mental state of rats in each group were observed and recorded. One hour after the last administration, 5 mL of abdominal aorta blood of rats in each group was collected, and the levels of serum interleukin (IL)-1β, IL-6, tumor necrosis factor-α (TNF-α) and transforming growth factor-β_1 (TGF-β_1) were detected by enzyme-linked immunosorbent assay. The rats were killed after blood collection, and the liver was taken to observe the general morphology. Then, the left lobe of liver was taken, and the liver tissue sections were prepared and stained with hematoxylin-eosin. The pathological changes of the liver tissues of rats were observed under the light microscope. **Results** The rats in the blank group had normal diet, frequent activities, white and shiny hair, and normal mental status. In the model group, the diet of rats was reduced, the activity was significantly reduced, the hair color was dark and unclean, and the mental state was depressed; in the silymarin group, the hair color of rats was less shiny but cleaner, and the other general conditions were the same as those of rats in the blank group; the general conditions of rats in the different dose of *Jiamudou* group were better than those in the model group but worse than those in the blank group. The growth of body mass of rats in the model group, silymarin group, low-dose *Jiamudou* group, middle-dose *Jiamudou* group and high-dose *Jiamudou* group was significantly less than that in the blank group ($P < 0.05$); the growth of body mass of rats in the low-dose *Jiamudou* group was less than that in the model group ($P < 0.05$). The levels of serum IL-1β, IL-6 and TNF-α of rats in the model group, silymarin group, low-dose *Jiamudou* group, middle-dose *Jiamudou* group and high-dose *Jiamudou* group were significantly higher than those in the blank group ($P < 0.05$). There was no significant difference in the level of serum TGF-β_1 of rats between silymarin group and blank group ($P > 0.05$). The levels of serum TGF-β_1 of rats in the model group, low-dose *Jiamudou* group, middle-dose *Jiamudou* group and high-dose *Jiamudou* group were significantly higher than those in the blank group ($P < 0.05$). The levels of serum IL-1β, IL-6, TNF-α and TGF-β_1 of rats in the silymarin group, low-dose *Jiamudou* group, middle-dose *Jiamudou* group and high-dose *Jiamudou* group were significantly lower than those in the model group ($P < 0.05$). The levels of serum IL-1β, IL-6, TNF-α and TGF-β_1 of rats in the low-dose *Jiamudou* group were significantly higher than those in the silymarin group ($P < 0.05$). There was no significant difference in the levels of serum IL-1β, IL-6 of rats between the middle-dose*Jiamudou* group and the silymarin group ($P > 0.05$); the levels of serum TNF-α and TGF-β_1 of rats in the middle-dose *Jiamudou* group were significantly higher than those in the silymarin group ($P < 0.05$); there was no significant difference in the levels of serum IL-1β, IL-6, TNF-α and TGF-β_1 of rats between the high-dose *Jiamudou* group and the silymarin group ($P > 0.05$). There was no significant difference in the level of serum IL-1β of rats among low-dose *Jiamudou* group, middle-dose *Jiamudou* group and high-dose *Jiamudou* group ($P > 0.05$). The levels of serum IL-6, TGF-β_1 of rats in the high-dose *Jiamudou* group were significantly lower than those in the low-dose *Jiamudou* group ($P < 0.05$); there was no significant difference in the levels of serum IL-6, TGF-β_1 of rats between the middle-dose *Jiamudou* group and the low-dose *Jiamudou* group, the middle-dose *Jiamudou* group and the high-dose *Jiamudou* group ($P > 0.05$). The level of serum TNF-α of rats in the low-dose *Jiamudou* group, middle-dose *Jiamudou* group and high-dose *Jiamudou* group gradually decreased with the dose, and the difference was statistically significant ($P < 0.05$). In the blank group, the liver tissue structure of rats was intact and the liver lobule structure was normal; the liver fibrosis of rats in the model group was obvious; compared with the model group, the histopathological morphology of the liver tissue of rats in the low-dose *Jiamudou* group was slightly improved, the histopathological morphology of liver tissue of rats in the silymarin group, middle-dose *Jiamudou* group and high-dose *Jiamudou* group were significantly improved, and the formation of liver fibers was reduced or disappeared. **Conclusion** *Jiamudou* extract has a protective effect on hepatic fibrosis of rats, and its mechanism may be related to the decrease of the levels of serum IL-1β, IL-6, TNF-α and TGF-β_1.

Key words: *Jiamudou* extract; hepatic fibrosis; interleukin-1β; interleukin-6; tumor necrosis factor-α; transforming growth factor-β_1

肝纤维化是指在各种慢性肝病中,肝细胞发生持续、反复的坏死,导致机体发生修复反应,大量纤维增生,同时伴有纤维降解相对或绝对不足,细胞外基质(extracellular matrix,ECM)在肝内大量沉积,超过肝脏自身降解能力所发生的病变。多种因素导致的慢性肝病在发展过程中往往伴随肝纤维化的发生,活动性肝纤维化如不能得到有效控制,将最终发展为肝硬化,甚至肝癌[1-2]。大部分肝纤维化患者治疗后肝纤维化可逆转至正常[3]。因此,阻断肝纤维化的发生、发展将成为慢性肝病治疗的关键环节。目前,临床上用于治疗肝纤维化药物的疗效不十分理想,且药物价格昂贵、不良反应多。近几十年来,中医学对肝纤维化的研究累积了丰富的经验,应用中药防治肝纤维化具有费用低、不良反应少、疗效好等优势,具有广阔的应用前景。

假木豆又名假绿豆、白毛千斤拔、野蚂蝗,其味辛、甘,性寒,具有清热凉血、舒经活络、健脾利湿之功效,主治咽喉肿痛、内伤吐血、跌打损伤、骨折、风

湿骨痛、瘫痪、泄泻、小儿疳积。假木豆主要分布于福建、广东、广西、海南、贵州、云南等地[4]。本课题组前期研究发现,假木豆对肝损伤有保护作用[5],但假木豆治疗肝纤维化的机制尚未见相关报道。水飞蓟素是从菊科植物水飞蓟中分离提取的一种黄酮类化合物,其在保肝抗炎方面的作用突出,基于此,本研究采用皮下注射四氯化碳(carbon tetrachloride,CCl_4)的方法制备大鼠肝纤维化模型,以水飞蓟素作为阳性对照观察假木豆水提物对大鼠肝纤维化的保护作用及相关机制。

1 材料与方法

1.1 实验动物 清洁级 Sprague Dawley(SD)大鼠 60 只,雌雄各半,体质量(200 ± 20)g,购自广西医科大学实验动物中心,许可证号:SCXK 桂 2014-0002。

1.2 试剂与仪器 水飞蓟素胶囊购自德国马博士大药厂(进口药品注册证号 H20181067)。CCl_4、无水乙醇购自天津市富宇精细化工有限公司,水合氯醛购自成都市科龙化工试剂厂,白细胞介素(interleukin,IL)-1β、IL-6、肿瘤坏死因子-α(tumor necrosis factor-α,TNF-α)和转化生长因子-β_1(transforming growth factor-β_1,TGF-β_1)酶联免疫吸附试验(enzyme-linked immunosorbent assay,ELISA)试剂盒购自深圳欣博盛生物科技有限公司;电子天平购自赛多利斯科学仪器(北京)有限公司,离心机购自上海安亭科学仪器厂,Infinite M200 酶标仪购自瑞士 Tecan 公司。

1.3 药物提取 取假木豆叶(采集自广西南宁市高峰林场)以水提方式处理(分 3 次先后用 8 倍量水、5 倍量水、3 倍量水提取),合并提取液,浓缩成浸膏,每毫升相当于生药材 6.22 g。低温储存,用时用纯水稀释。

1.4 实验方法

1.4.1 实验分组及干预措施 取 SD 大鼠 60 只,雌雄各半,适应性饲养 3 d 后,按性别随机分为空白组、模型组、水飞蓟素组、低剂量假木豆组、中剂量假木豆组和高剂量假木豆组,每组 10 只。水飞蓟素组、低剂量假木豆组、中剂量假木豆组、高剂量假木豆组和模型组大鼠参照文献[6]制备肝纤维化模型,具体方法为:第 1 周背部皮下注射含体积分数 40% CCl_4 的花生油溶液 5 mL · kg^{-1},第 2 ~ 8 周背部皮下注射含体积分数 40% CCl_4 的花生油 3 mL · kg^{-1},每隔 2 d 注射 1 次。空白组大鼠背部皮下注射花生油,方法同上述各组。第 1 ~ 8 周,以体积分数 5% 乙醇作为大鼠唯一饮水。从第 3 周起,水飞蓟素组大鼠每日灌服水飞蓟素胶囊水溶液(50 mg · kg^{-1})1 次;低、中、高剂量假木豆组大鼠每日灌服 12.5、25.0、50.0 g · L^{-1} 的假木豆水提液(20 mL · kg^{-1});空白组和模型组大鼠灌服等体积的蒸馏水;各组大鼠均连续给药 6 周。

1.4.2 各组大鼠一般状况观察 实验期间观察并记录各组大鼠饮食活动情况、体质量、毛色光亮程度、精神状况等一般情况。

1.4.3 ELISA 法检测各组大鼠血清中 IL-1β、IL-6、TNF-α、TGF-β_1 水平 末次给药 1 h 后,各组大鼠腹腔注射 100 g · L^{-1} 水合氯醛溶液(3 mL · kg^{-1})进行麻醉,采集大鼠腹主动脉血 5 mL,室温下静置 2 h,3 000 r · min^{-1} 离心 10 min,收集血清,采用 ELISA 法检测各组大鼠血清中 IL-1β、IL-6、TNF-α 和 TGF-β_1 水平,严格按照试剂盒说明书进行操作。

1.4.4 各组大鼠肝脏大体形态及病理组织学形态观察 采血后处死大鼠,取肝脏观察大体形态;然后取大鼠肝脏左叶,于体积分数 10% 甲醛溶液中固定 24 h 以上,然后制备肝组织切片,苏木精-伊红(hematoxylin-eosin,HE)染色,光学显微镜下观察并拍照,观察大鼠肝脏病理形态学变化。

1.5 统计学处理 应用 SPSS 19.0 软件进行统计学分析。计量资料以均数 ± 标准差($\bar{x} \pm s$)表示,多组间比较采用方差分析,组间两两比较采用最小显著性差异法 t 检验,$P < 0.05$ 为差异有统计学意义。

2 结果

2.1 6 组大鼠一般情况比较 空白组大鼠饮食正常,活动频繁,毛色洁白有光泽,精神状况正常。模型组大鼠饮食减少,活动量明显减少,毛色暗淡、不清洁,精神状况萎靡;水飞蓟素组大鼠饮食正常,活动正常,毛色欠缺光泽但较清洁,精神状况较好;各剂量假木豆组大鼠饮食略有减少,活动量一般,皮毛较暗淡,略发黄,精神状况尚可,总体情况较模型组佳而比空白组差。模型组、水飞蓟素组、低剂量假木豆组、中剂量假木豆组、高剂量假木豆组大鼠体质量增长量少于空白组,差异有统计学意义($P < 0.05$);低剂量假木豆组大鼠体质量增长少于模型组,差异有统计学意义($P < 0.05$)(表 1)。

表 1 6 组大鼠体质量增长量比较

Tab. 1 Comparison of growth of body mass of rats among the six groups ($\bar{x} \pm s$)

组别	n	体质量增长/g
空白组	10	129.74 ± 64.77
模型组	10	62.52 ± 33.95[a]
水飞蓟素组	10	61.82 ± 43.06[a]
低剂量假木豆组	10	35.98 ± 16.33[ab]
中剂量假木豆组	10	38.01 ± 16.33[a]
高剂量假木豆组	10	49.44 ± 27.58[a]

注:与空白组比较[a] $P < 0.05$;与模型组比较[b] $P < 0.05$。

2.2 6 组大鼠血清中 IL-1β、IL-6、TNF-α、TGF-β_1 水平比较 结果见表 2。模型组、水飞蓟素组、低剂量假木豆组、中剂量假木豆组、高剂量假木豆组大鼠

血清中 IL-1β、IL-6、TNF-α 水平均高于空白组,差异有统计学意义($P<0.05$);水飞蓟素组与空白组大鼠血清中 TGF-β_1 水平比较差异无统计学意义($P>0.05$);模型组、低剂量假木豆组、中剂量假木豆组、高剂量假木豆组大鼠血清中 TGF-β_1 水平均高于空白组,差异有统计学意义($P<0.05$)。水飞蓟素组、低剂量假木豆组、中剂量假木豆组、高剂量假木豆组大鼠血清中 IL-1β、IL-6、TNF-α、TGF-β_1 水平均低于模型组,差异有统计学意义($P<0.05$)。低剂量假木豆组大鼠血清中 IL-1β、IL-6、TNF-α、TGF-β_1 水平显著高于水飞蓟素组,差异有统计学意义($P<0.05$)。中剂量假木豆组与水飞蓟素组大鼠血清中 IL-1β、IL-6 水平比较差异无统计学意义($P>0.05$),中剂量假木豆组大鼠血清中 TNF-α、TGF-β_1 水平显著高于水飞蓟素组,差异有统计学意义($P<0.05$);高剂量假木豆组与水飞蓟素组大鼠血清中 IL-1β、IL-6、TNF-α、TGF-β_1 水平比较差异无统计学意义($P>0.05$)。低、中、高剂量假木豆组大鼠血清中 IL-1β 水平比较差异均无统计学意义($P>0.05$)。高剂量假木豆组大鼠血清中 IL-6、TGF-β_1 水平显著低于低剂量假木豆组,差异有统计学意义($P<0.05$);中剂量假木豆组与低剂量假木豆组、高剂量假木豆组与中剂量假木豆组大鼠血清中 IL-6、TGF-β_1 水平比较差异均无统计学意义($P>0.05$)。低、中、高剂量假木豆组大鼠血清中 TNF-α 水平两两比较差异均有统计学意义($P<0.05$)。

表 2　6 组大鼠血清中 IL-1β、IL-6、TNF-α、TGF-β_1 水平比较

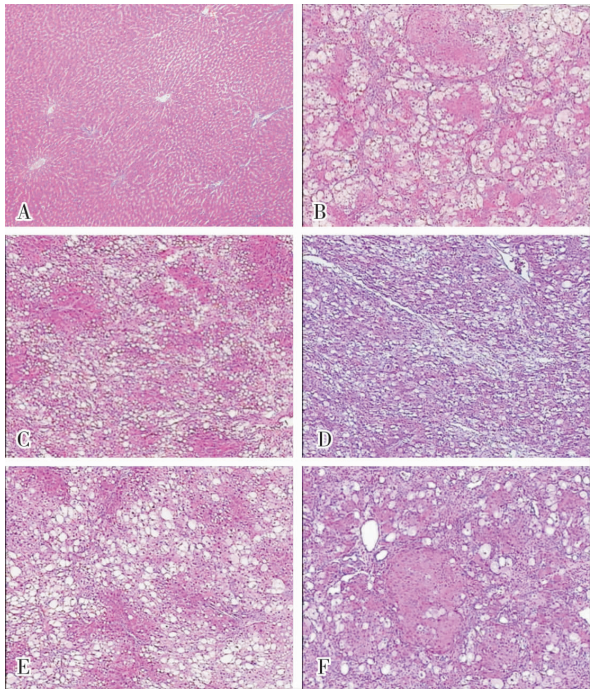
Tab. 2　Comparison of the levels of serum IL-1β, IL-6, TNF-α, TGF-β_1 of rats among the six groups　($\bar{x}\pm s$)

组别	n	IL-1β/(ng·L^{-1})	IL-6/(ng·L^{-1})	TNF-α/(ng·L^{-1})	TGF-β_1/(ng·L^{-1})
空白组	10	77.61±12.99	85.63±24.01	55.79±10.56	251.63±65.64
模型组	10	141.97±24.48^{a}	245.36±24.10^{a}	643.95±145.56^{a}	643.95±145.56^{a}
水飞蓟素组	10	104.68±12.93^{ab}	172.80±21.67^{ab}	81.88±26.80^{ab}	272.31±144.19^{b}
低剂量假木豆组	10	124.85±25.72^{abc}	199.75±22.67^{abc}	152.81±32.22^{abc}	485.33±116.30^{abc}
中剂量假木豆组	10	112.64±21.87^{ab}	190.44±19.43^{ab}	121.99±15.86^{abcd}	423.57±58.83^{abc}
高剂量假木豆组	10	105.28±18.82^{ab}	169.32±25.61^{abd}	103.79±20.93^{abde}	364.95±80.77^{abd}
F		11.240	43.512	42.604	18.328
P		0.000	0.000	0.000	0.000

注:与空白组比较$^{a}P<0.05$;与模型组比较$^{b}P<0.05$;与水飞蓟素组比较$^{c}P<0.05$;与低剂量假木豆组比较$^{d}P<0.05$;与中剂量假木豆组比较$^{e}P<0.05$。

2.3　各组大鼠肝脏大体形态　空白组大鼠肝脏颜色鲜红,形状楔形,肝脏边缘整齐,表面光滑平整。模型组大鼠肝脏颜色晦暗,肝脏表面布有密集的小孔,凹凸不平,有较为严重的肿大或萎缩。水飞蓟素组及各剂量假木豆组大鼠肝脏颜色较为暗淡,表面稍粗糙、布有较多小孔,肝脏变形情况较模型组轻。

2.4　各组大鼠肝组织病理学形态　结果见图 1。空白组大鼠肝组织结构完整,肝小叶结构正常,未见有肝细胞水肿及脂肪变。模型组大鼠肝组织肝纤维化明显,可见严重的肝细胞水肿、脂肪变性以及明显的假小叶形成。水飞蓟素组大鼠肝组织可见较为严重的肝细胞水肿、脂肪变性,未见纤维化,也未见假小叶形成。低剂量假木豆组大鼠肝组织可见严重肝细胞水肿和脂肪变性,肝组织中可见较薄的纤维结缔组织增生,部分分隔肝小叶,有少量假小叶形成。中剂量假木豆组大鼠肝组织中可见严重肝细胞水肿、脂肪变性,有少量肝纤维形成,未见假小叶形成。高剂量假木豆组大鼠肝组织中可见较为严重的肝细胞水肿及脂肪变性,肝纤维化情况轻微,未见有假小叶形成。水飞蓟素组和假木豆中、高剂量组大鼠肝组织病理学形态较模型组有较大的改善,肝纤维形成减轻或消失,没有假小叶形成;低剂量假木豆组大鼠肝组织病理学形态较模型组有轻微改善但不明显。各组大鼠肝细胞水肿、脂肪变性均未得到明显改善。



A:空白组;B:模型组;C:水飞蓟素组;D:高剂量假木豆组;E:中剂量假木豆组;F:低剂量假木豆组。

图 1　6 组大鼠肝组织病理学形态(HE 染色,×100)

Fig. 1　Histopathological morphology of liver tissue of rats in the six groups (HE staining, ×100)

3 讨论

肝纤维化是由多种原因引起的慢性肝损害所致的病理改变,是多种慢性肝病共有的病理变化。肝纤维化是一个动态发展过程,随着纤维化程度的加深,可发展为肝硬化,甚至肝癌。肝纤维化是一种损伤愈合反应,其发生是肝脏中 ECM 增生和降解失衡,造成肝脏内纤维结缔组织异常沉积、肝脏结构和功能改变的结果[7-8]。有研究认为,肝纤维化是一个可逆的过程,阻断肝纤维化是一个有前景的针对肝脏相关疾病的治疗手段[9]。目前临床上尚无有效治疗肝纤维化的药物,故开发治疗肝纤维化的药物仍是新药研发的热点[1],而中药因其低毒性及全局性效果受到广泛关注[10-12]。

IL 属于细胞因子家族,是重要的炎症介质和免疫调节因子,可以调节肝脏炎症及免疫功能,从而影响肝纤维化的发生、发展。IL-1β、IL-6、IL-8 具有明确的促肝纤维化作用,IL-2、IL-10 具有抗肝纤维化作用。肝纤维化发生时,肝脏细胞中 IL-1β 的表达增加,IL-1β 可刺激肝脏发生炎症反应,进一步促进肝纤维化的发展[13-14]。IL-6 参与肝纤维化的多种病理过程,被认为是肝星状细胞(hepatic stellate cell,HSC)活化的标志[15-16]。TNF 是一种与内毒素无关的、可引起肿瘤出血坏死的活性分子[17]。正常情况下,肝组织中不表达或微量表达 TNF-α,当受到损伤因素刺激时,大量的 TNF-α 被释放出来,参与肝损伤过程,促进肝纤维化的发生、发展[18-19]。有研究显示,血清中 TNF-α 水平随患者肝纤维化程度的加重而升高,且与血清中的某些肝纤维化因子水平呈正相关[20],说明 TNF-α 不仅介导炎症反应,还参与了肝纤维化的形成过程,与肝纤维化密切相关。TGF-β_1 是细胞因子 TGF-β 超家族的成员,对细胞的生长、分化和多种生理、病理过程起重要调节作用,在肝纤维化的发生、发展过程中起着关键作用[21]。其通过活化 HSC、促进 ECM 的合成与沉积而致肝纤维化,是肝纤维化重要的始动因子之一。本研究结果显示,与空白组相比,模型组大鼠血清中 IL-1β、IL-6、TNF-α 和 TGF-β_1 水平显著升高,且病理组织学显示模型组大鼠肝脏损伤严重,有明显的纤维化特征,说明 CCl_4 诱导的肝纤维化模型制备成功。本研究发现,与模型组比较,假木豆高、中剂量组大鼠血清中 IL-1β、IL-6、TNF-α 和 TGF-β_1 水平显著降低;低剂量假木豆组大鼠血清中 IL-6、TNF-α 和 TGF-β_1 水平显著降低,IL-1β 水平无明显变化。该结果提示假木豆提取物对大鼠肝纤维化有保护作用,其作用机制可能与降低血清中 IL-1β、IL-6、TNF-α 及 TGF-β_1 水平有关。高剂量假木豆组与水飞蓟素组大鼠血清中 IL-1β、IL-6、TNF-α 和 TGF-β_1 比较差异无统计学意义,说明高剂量假木豆对肝纤维化大鼠血清中 IL-1β、IL-6、TNF-α 和 TGF-β_1 的作用与水飞蓟素相当。不同剂量假木豆组大鼠血清中 IL-1β、IL-6、TNF-α 及 TGF-β_1 水平随假木豆水提物剂量的升高呈下降趋势,但只有血清中 TNF-α 水平在 3 个剂量组之间比较差异有统计学意义,而血清中 IL-1β 水平在 3 个剂量组之间比较差异无统计学意义。因此,假木豆提取物与上述各指标的量效关系需进一步研究来验证。

综上所述,假木豆提取物具有抗肝纤维化作用,其作用机制可能与降低血清中 IL-1β、IL-6、TNF-α 及 TGF-β_1 水平有关。

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