基础研究 论 著

甘露清瘟方对小鼠急性肺损伤的治疗作用及其机制

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[摘要] **目的** 探究甘露清瘟方对脂多糖(LPS)诱导的小鼠急性肺损伤/急性呼吸窘迫综合征(ALI/ARDS)的治疗作用及其机制。**方法** 50只ICR小鼠随机分为对照组、模型组及甘露清瘟低(7.10 g/kg)、中(15.21 g/kg)和高(30.42 g/kg)剂量组，每组10只。第1—3天，各甘露清瘟组小鼠分别灌胃给予相应剂量的甘露清瘟方，对照组、模型组灌胃给予等体积生理盐水，1次/d；第4天，模型组与各甘露清瘟组小鼠腹腔注射LPS(20 mg/kg)构建ALI/ARDS模型，对照组腹腔注射等体积PBS；24 h后观察小鼠存活率，采集血清和肺组织，检测肺组织湿干重比(W/D)并行肺组织HE染色，ELISA法检测血清肿瘤坏死因子(TNF)-α、γ干扰素(IFN-γ)、白细胞介素(IL)-4、IL-10、IL-12水平及肺组织TNF-α、IFN-γ、IL-1β、IL-4、IL-6、IL-10、IL-12水平，Western blotting检测肺组织NOD样受体热蛋白结构域相关蛋白3(NLRP3)、膜穿孔蛋白Gasdermin D(GSDMD)、胱天蛋白酶-1(Caspase-1)、凋亡相关斑点样蛋白(ASC)表达水平。**结果** 各组小鼠存活率比较差异无统计学意义($P>0.05$)。ELISA法和Western blotting检测结果显示，与对照组比较，模型组小鼠肺组织W/D、IFN-γ、TNF-α、IL-4、IL-12、IL-1β、IL-6、NLRP3、ASC、Caspase-1、GSDMD和血清IFN-γ、TNF-α、IL-4、IL-12水平明显升高($P<0.05$)，肺组织和血清IL-10水平明显降低($P<0.05$)；与模型组比较，甘露清瘟低、中、高剂量组小鼠肺组织W/D、IFN-γ、TNF-α、IL-1β、IL-4、IL-6、IL-12、NLRP3、ASC、Caspase-1、GSDMD和血清IFN-γ、TNF-α、IL-4、IL-12水平均明显降低($P<0.05$)，肺组织IL-10水平明显升高($P<0.05$)，甘露清瘟高剂量组血清IL-10水平明显升高($P<0.05$)；与甘露清瘟低剂量组比较，甘露清瘟高剂量组小鼠肺组织IFN-γ、IL-6、NLRP3、ASC、Caspase-1、GSDMD和血清TNF-α水平明显降低($P<0.05$)，肺组织和血清IL-10水平明显升高($P<0.05$)。小鼠肺组织HE染色结果显示，对照组肺结构清晰、正常；模型组部分肺间质充血、出血，部分细支气管周围炎性细胞浸润；甘露清瘟低、中、高剂量组肺间质充血、出血现象减少，炎性细胞浸润程度减轻。**结论** 甘露清瘟方可能通过抑制NLRP3/Caspase-1/GSDMD通路，抑制细胞焦亡，延缓小鼠AIL/ARDS的发生。

[关键词] 甘露清瘟；脂多糖；急性肺损伤；急性呼吸窘迫综合征；细胞焦亡

Therapeutic effects and underlying mechanisms of Ganluqingwen formula on acute lung injury in mice

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[**Abstract**] **Objective** To investigate the therapeutic effects and underlying mechanisms of Ganluqingwen formula on

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lipopolysaccharide (LPS)-induced acute lung injury/acute respiratory distress syndrome (ALI/ARDS) in mice. **Methods** Fifty ICR mice were randomly divided into five groups: control, model, and Ganluqingwen formula (GLQW) low dose (7.10 g/kg), medium dose (15.21 g/kg), and high dose (30.42 g/kg) groups, with 10 mice per group. On days 1-3, mice in GLQW groups were daily gavaged with the corresponding dose of GLQW, while control and model groups received equal volumes of saline. On day 4, ALI/ARDS was induced in model and GLQW groups using intraperitoneal injection of LPS (20 mg/kg), while control group received an equal volume of PBS. At 24 h post-treatment, survival rate, wet-to-dry weight ratio (W/D) and lung histological changes (HE staining) were observed. Serum levels of tumor necrosis factor (TNF)-α, interferon gamma (IFN-γ), interleukin (IL)-4, IL-10, IL-12, as well as lung tissue levels of TNF-α, IFN-γ, IL-1β, IL-4, IL-6, IL-10 were measured by ELISA. Western blotting was used to determine the expression levels of NOD-like receptor thermal protein domain associated protein 3 (NLRP3), cystatinase-1 (Caspase-1), apoptosis-associated speck-like protein (ASC), and membrane perforating protein Gasdermin D (GSDMD) in lung tissue. **Results** No significant differences in survival rates were observed among the groups (P>0.05). Compared with control group, ELISA and Western blotting results showed that lung tissue W/D, IFN-γ, TNF-α, IL-4, IL-12, IL-1β, IL-6, NLRP3, ASC, and Caspase-1, GSDMD and serum IFN-γ, TNF-α, IL-4, IL-12 levels were significantly higher (P<0.05), and IL-10 levels in lung tissue and serum were significantly lower in mice of model group (P<0.05). Compared with model group, lung tissue W/D, IFN-γ, TNF-α, IL-1β, IL-4, IL-6, IL-12, NLRP3, ASC, Caspase-1, and GSDMD, and serum IFN-γ, TNF-α, IL-4, and IL-12 levels were significantly lower (P<0.05), and lung tissue IL-10 levels were significantly higher (P<0.05) in GLQW low, medium, and high dose groups, with high-dose group showing significantly higher level in serum IL-10 (P<0.05). Compared with GLQW low-dose group, the lung tissue levels of IFN-γ, IL-6, NLRP3, ASC, Caspase-1, and GSDMD, and serum TNF-α were significantly lower (P<0.05), and lung and serum IL-10 levels were significantly higher in GLQW high-dose group (P<0.05). HE staining results showed that lung structure was clear and normal in control group; part of the lung interstitium was congested and hemorrhagic, and some of the fine bronchial periphery was infiltrated with inflammatory cells in model group; the phenomena of lung interstitial congestion and hemorrhage were reduced, and the degree of infiltration of inflammatory cells was alleviated in GLQW low-, medium-, and high-dose groups. **Conclusion** Ganluqingwen formula can delay the development of ALI/ARDS in mice by inhibiting NLRP3/Caspase-1/GSDMD pathway, thereby suppressing cellular pyroptosis.

[**Key words**] Ganluqingwen; lipopolysaccharide; acute lung injury; acute respiratory distress syndrome; pyroptosis

急性肺损伤(acute lung injury，ALI)/急性呼吸窘迫综合征(acute respiratory distress syndrome，ARDS)是多种直接或间接原因如肺炎、创伤、中毒、误吸、胰腺炎、输血等[1]引起的以进行性呼吸困难、持续性顽固低氧血症、弥漫性肺水肿为临床表现的综合征[2-3]。ALI/ARDS发病率较高，30 d病死率达21.5%~27.2%，严重威胁人类健康[4-7]；目前其临床治疗以支持治疗为主，但随着抗生素、糖皮质激素等药物的使用及长时间的机械通气治疗，可能引起多种并发症及后遗症，如住院周期延长、呼吸机相关感染和患者死亡风险增加[8]；机械通气虽然能帮助塌陷的肺泡膨胀复张，改善氧合，纠正低氧血症[9]，但不当的正压和潮气量可导致机械性损伤及肺泡表面活性物质减少[10]，亦可导致毛细血管通透性增加，使更多炎性因子释放至间质[11]。因此，ALI/ARDS治疗药物的开发与应用已成为近年来的研究热点。

甘露清瘟方(以下简称甘露清瘟)是李风森教授治疗呼吸系统感染性疾病的经验方，由普济消毒饮加减化裁而成，具有清热解毒、清肺化痰之功，在病毒性感冒、冠状病毒感染、社区获得性肺炎等疾病的临床治疗中取得一定疗效。本研究采用脂多糖(lipopolysaccharide，LPS)诱导的小鼠ALI/ARDS模型，探讨甘露清瘟对ALI/ARDS的治疗作用及与肺组织细胞焦亡相关的作用机制，旨在为甘露清瘟颗粒制剂的研发提供理论依据。

1 材料与方法

1.1 实验动物及分组 10周龄SPF级ICR小鼠50只，雌雄各半，体重(20±2) g，购自新疆医科大学动物实验中心并于该中心饲养[实验动物生产许可证号：SCXK(新)2023-0001；使用许可证号：SYXK(新)2023-0004]，环境温度(23±3) ℃，相对湿度50%±10%，自由进食水。将小鼠随机分为对照组、模型组及甘露清瘟低、中、高剂量组，每组10只。本研究经新疆医科大学实验动物伦理委员会审批(IACUC-JT-20230831-52)。实验过程符合国家和单位有关实验动物的管理和使用规定。

1.2 药物 甘露清瘟方组成包括茵陈、青蒿各15 g，连翘12 g，黄芩、射干、伊贝母、陈皮、豆蔻、石菖蒲、燀桃仁各9 g，藿香、法半夏各6 g，购自新疆医科大学附属中医医院。

1.3 主要试剂 LPS(L2630)购自美国Sigma公司；白细胞介素(IL)-1β(E-EL-M003)、IL-4（E-EL-M0043）、IL-6(E-EL-M0044)、IL-I0(E-EL-M0046)、IL-12(E-EL-M3062)、肿瘤坏死因子-α(TNF-α；E-EL-M3063)、γ干扰素(IFN-γ；E-EL-M0048)ELISA检测试剂盒购自

武汉伊莱瑞特生物科技有限公司；苏木精-伊红(HE)染色试剂盒(G1120)、高效RIPA裂解液(R0010)、5×蛋白上样缓冲液(P1040)购自北京索莱宝科技有限公司；BCA蛋白浓度测定试剂盒(BL521A)、特超敏ECL化学发光底物(BL520B)购自中国兰杰柯科技有限公司；抗NOD样受体热蛋白结构域相关蛋白3(NOD-like receptor thermal protein domain associated protein 3，NLRP3；19771-1-AP)、凋亡相关斑点样蛋白(apoptosis-associated speck-like protein，ASC；10500-1-AP)、GAPDH抗体(10494-1-AP)购自武汉三鹰生物技术有限公司；抗胱天蛋白酶-1(cysteinyl aspartate specific proteinase，Caspase-1；ab286125)抗体、辣根过氧化物酶(horseradish peroxidase，HRP)标记的山羊抗兔IgG二抗(ab205718)购自英国Abcam公司；抗膜穿孔蛋白Gasdermin D(GSDMD)抗体(AF4012)购自江苏亲科生物研究中心有限公司。

1.4 方法

1.4.1 药物制备 取甘露清瘟草药2剂，加1170 g水浸泡30 min，大火煮沸后，转小火煮30 min，无纺布过滤药渣；药渣加936 g水，再次煎煮、过滤；将两次药液混匀，继以浓缩、低温烘干，最终获得约40 g干浸膏粉，密封、冷藏，造模前以0.9%氯化钠注射液溶解。

1.4.2 ALI/ARDS小鼠模型的建立 第1—3天，各甘露清瘟组小鼠灌胃给予甘露清瘟，1次/d，其中低剂量组7.10 g/kg、中剂量组15.21 g/kg、高剂量组30.42 g/kg[1付中药为117 g(相当于20 g干浸膏粉)，中剂量：117 g/70 kg×9.1=15.21 g/kg；低剂量：15.21 g/kg×0.5=7.10 g/kg；高剂量：15.21 g/kg×2=30.42 g/kg]；对照组、模型组灌胃给予等体积生理盐水，1次/d。第4天，模型组与各甘露清瘟组腹腔注射20 mg/kg LPS(半数致死剂量)建立ALI/ARDS小鼠模型，对照组腹腔注射等体积PBS。

1.4.3 血清和肺组织采集 腹腔注射LPS(对照组腹腔注射等体积PBS)24 h后腹主动脉取血处死小鼠，离心取血清用于ELISA检测；迅速收集肺组织，右肺上叶用于检测肺组织湿/干重比(W/D)，右肺中叶以4%多聚甲醛溶液固定用于病理检测，右肺下叶制作组织匀浆后用于ELISA检测，左肺组织剪碎后以RIPA裂解制备蛋白母液用于Western blotting检测。

1.4.4 小鼠肺组织W/D检测 PBS冲洗右肺上叶血液，称量肺湿重(W)，然后在60 ℃烘箱烘烤48 h，称量肺干重(D)，计算W/D。

1.4.5 HE染色观察小鼠肺组织病理学改变 右肺中叶固定、脱水、石蜡包埋、切片后行HE染色，光学显微下观察肺组织病理学改变。

1.4.6 ELISA法检测小鼠血清和肺组织炎症指标 按照说明书步骤，取血清检测IFN-γ、TNF-α、IL-4、IL-10、IL-12水平。取右肺下叶组织匀浆检测IFN-γ、TNF-α、IL-4、IL-10、IL-12、IL-1β、IL-6水平。

1.4.7 Western blotting检测小鼠肺组织NLRP3、ASC、Caspase-1、GSDMD蛋白表达水平 蛋白母液BCA定量后，将蛋白母液、RIPA、5×上样缓冲液按照BCA定量计算结果混匀，使各组蛋白浓度达到20 μg/10 μl，煮样。上样20 μg，行SDS-PAGE凝胶电泳及转膜。5%脱脂奶粉室温封闭2 h，分别加入抗NLRP3(1∶1000)、ASC(1∶1000)、Caspase-1(1∶1000)、GSDMD(1∶1000)、GAPDH(1∶5000)抗体，4 ℃摇床过夜。洗膜3次，加入二抗HRP标记的山羊抗兔IgG(1∶10 000)室温孵育50 min；再次洗膜，使用超敏ECL发光底物试剂，A液、B液等量混匀，将显色液均匀滴洒在PVDF膜上，保持1 min，使用化学发光成像系统曝光，并保存图像。

1.5 统计学处理 采用GraphPad Prism 8.2软件进行统计分析。计量资料均符合正态分布，以$\bar{x}\pm s$表示，多组间比较采用单因素方差分析，进一步两两比较采用LSD-*t*检验。计数资料以例(%)表示，组间比较采用Fisher确切概率法。*P*<0.05为差异有统计学意义。

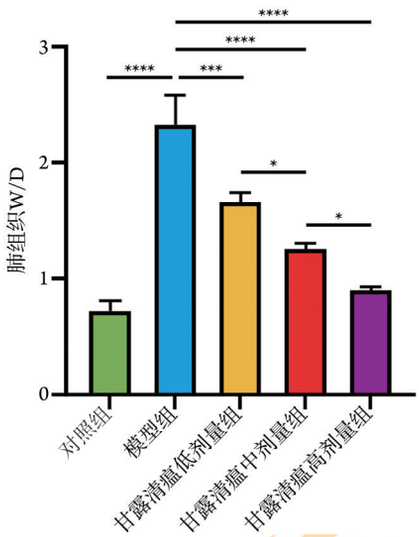
2 结 果

2.1 甘露清瘟对小鼠存活率的影响 造模后24 h，对照组小鼠存活10只(100.0%)，模型组存活5只(50.0%)，甘露清瘟低剂量组和中剂量组均存活6只(60.0%)，甘露清瘟高剂量组存活8只(80.0%)。各组小鼠存活率比较差异无统计学意义(*P*>0.05)。

2.2 甘露清瘟对小鼠肺组织W/D的影响 与对照组比较，模型组小鼠肺组织W/D明显增高(*P*<0.0001)；与模型组比较，甘露清瘟低、中、高剂量组小鼠W/D均明显降低(*P*<0.001或*P*<0.0001)；甘露清瘟中剂量组W/D明显低于低剂量组(*P*<0.05)，高剂量组W/D明显低于中剂量组(*P*<0.05)(图1)。

2.3 甘露清瘟对小鼠血清炎症指标的影响 ELISA法检测结果显示，与对照组比较，模型组小鼠血清IFN-γ、TNF-α、IL-4、IL-12水平明显升高(*P*<0.0001)，IL-10水平明显降低(*P*<0.0001)；与模型组比较，甘露清瘟低、中、高剂量组血清IFN-γ、TNF-α、IL-4、IL-12水平明显降低(*P*<0.001)，甘露清瘟高剂量组IL-10水平明显升高(*P*<0.05)；与甘露清瘟低剂量组比较，甘露清瘟高剂量组小鼠血清IL-10水平明显升高(*P*<0.05)；与甘露清瘟低、中剂量组比较，甘露清瘟高剂量组小鼠血清TNF-α水平明显降低(*P*<0.001或*P*<0.0001)，IL-10水平明显升高(*P*<0.05或*P*<0.01)(图2)。

2.4 甘露清瘟对小鼠肺组织炎症指标的影响 ELISA



*P<0.05，***P<0.001，****P<0.0001

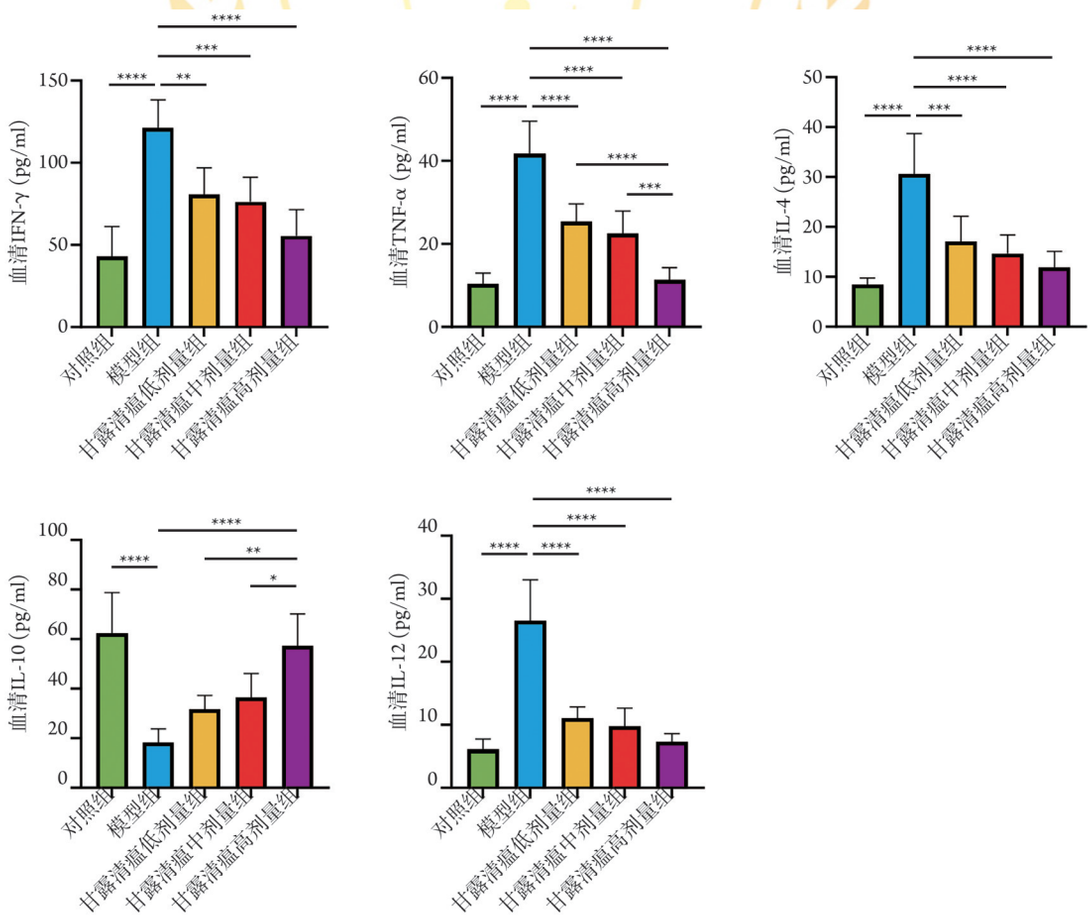
图 1 甘露清瘟方对各组小鼠肺组织湿干重比(W/D)的影响

Fig.1 Effect of Ganluqingwen formula on lung wet to dry weight ratio (W/D) of mice in each group

法检测结果显示，与对照组比较，模型组小鼠肺组织IFN-γ、TNF-α、IL-4、IL-12、IL-1β、IL-6水平明显升高(*P*<0.0001)，IL-10水平明显降低(*P*<0.0001)；与模型组比较，甘露清瘟低、中、高剂量组小鼠肺组织IFN-γ、TNF-α、IL-1β、IL-4、IL-6、IL-12水平明显降低(*P*<0.05)，IL-10水平明显升高(*P*<0.05)；与甘露清瘟低剂量组比较，甘露清瘟高剂量组小鼠肺组织IFN-γ、IL-6水平明显降低(*P*<0.0001或*P*<0.01)，IL-10水平明显升高(*P*<0.05)(图3)。

2.5 甘露清瘟对小鼠肺组织病理学改变的影响 HE染色结果显示，对照组小鼠肺组织结构清晰，肺泡壁薄，肺泡大小、数量正常，排列有序，肺泡腔内、支气管内无炎性渗出物，偶见少量炎性细胞浸润；与对照组比较，模型组小鼠部分肺间质充血、出血，部分肺泡壁明显增厚，部分肺泡腔内有炎性渗出物，部分细支气管周围较多炎性细胞浸润；与模型组比较，甘露清瘟低、中和高剂量组增厚的肺泡壁范围均减少，肺间质充血、出血现象减少，炎性细胞浸润程度减轻；与甘露清瘟低剂量组比较，甘露清瘟中剂量组差异不明显，甘露清瘟高剂量组炎性细胞浸润程度稍减轻(图4)。

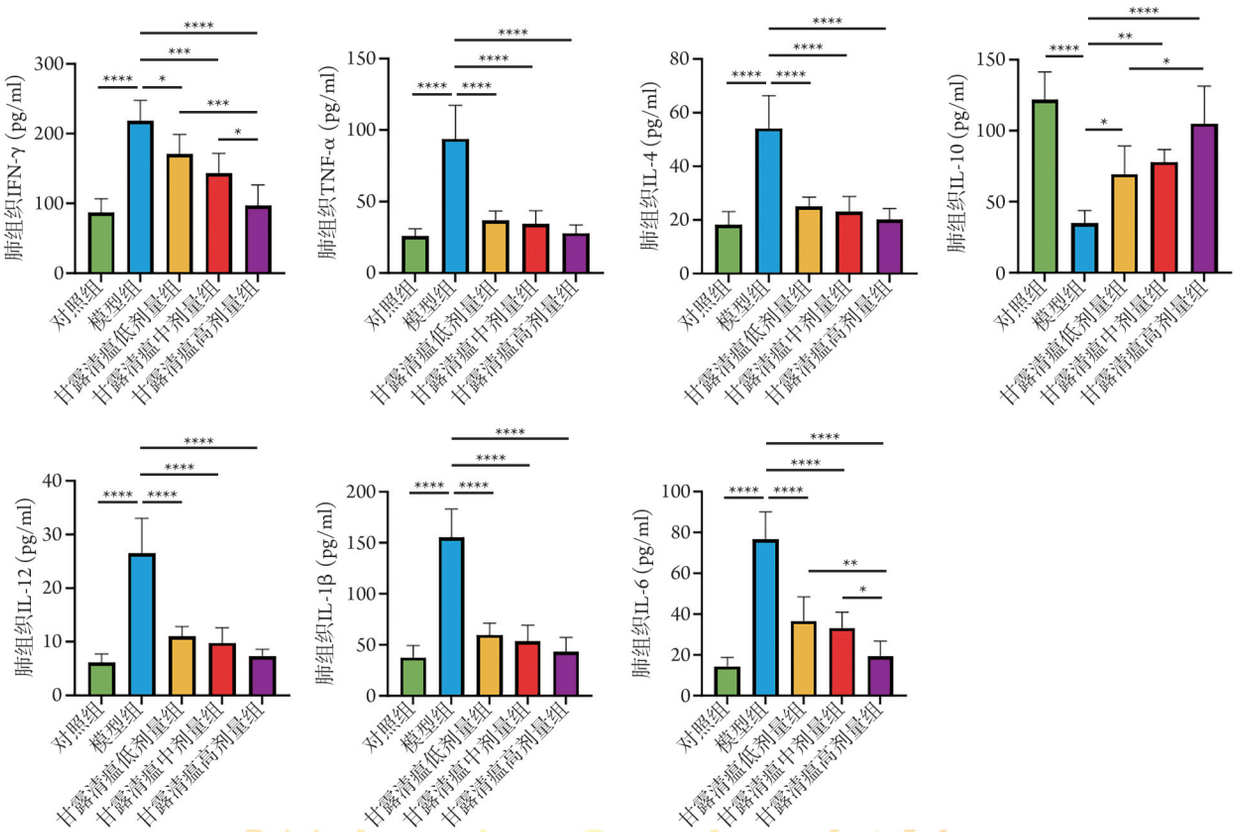
2.6 甘露清瘟对小鼠肺组织NLRP3、GSDMD、



IFN-γ.γ干扰素；TNF-α.肿瘤坏死因子-α；IL.白细胞介素；*P<0.05，**P<0.01，***P<0.001，****P<0.0001

图 2 甘露清瘟方对小鼠血清炎症指标的影响

Fig. 2 Effects of Ganluqingwen formula on the serum levels of inflammatory indicators in mice of each group



IFN-γ.γ干扰素；TNF-α.肿瘤坏死因子-α；IL.白细胞介素；*P<0.05，**P<0.01，***P<0.001，****P<0.0001

图3 甘露清瘟方对小鼠肺组织炎症指标的影响

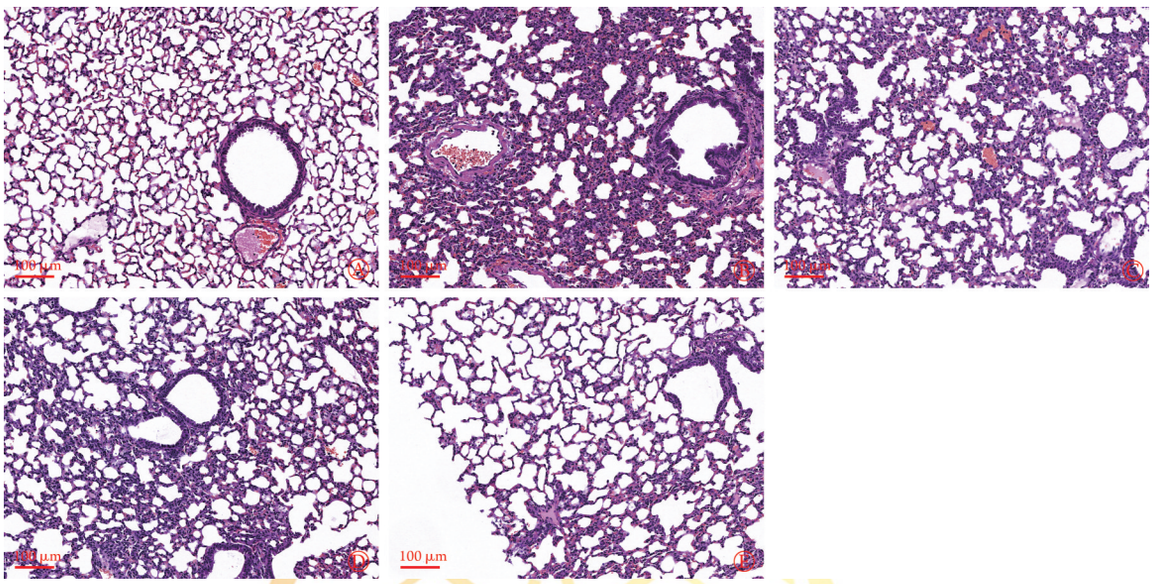
Fig. 3 Effects of Ganluqingwen formula on the levels of inflammatory indicators in lung tissues of mice in each group

Caspase-1、ASC蛋白表达的影响 Western blotting检测结果显示，与对照组比较，模型组小鼠肺组织NLRP3、GSDMD、Caspase-1、ASC蛋白表达水平明显增高(P<0.0001)；与模型组比较，甘露清瘟低、中、高剂量组小鼠肺组织NLRP3、GSDMD、Caspase-1、ASC蛋白表达水平明显降低(P<0.05)；与甘露清瘟低剂量组比较，甘露清瘟中剂量组小鼠肺组织ASC、Caspase-1、GSDMD蛋白表达水平明显降低(P<0.05)；与甘露清瘟中剂量组比较，甘露清瘟高剂量组小鼠肺组织Caspase-1蛋白表达水平明显降低(P<0.0001)(图5)。

3 讨 论

ALI/ARDS是以肺部炎症反应为病理特征，主要表现为肺水肿和呼吸衰竭的严重肺部疾患[12]，具有肺顺应性下降、肺内分流增加、无效腔容量增加、通气-血流比失衡、毛细血管通透性增加、肺间质水肿等病理生理特征。因过度的炎症反应导致大量中性粒细胞、巨噬细胞浸润，进而导致弥漫性肺泡损伤、肺泡毛细血管和上皮细胞功能障碍甚至坏死、透明膜形成，最终形成毛细血管内血栓。ALI/ARDS的治疗以抗感染、抗炎及保护性肺通气为主，其他治疗还包括严格的液体管理以减轻肺水肿、给予吸入性肺血管扩张药物改善分流，必要时可给予体外膜肺氧合(extracorporeal membrane oxygenation，ECMO)提供体外呼吸、循环支持；糖皮质激素的使用仍存在争议，虽然可发挥一定的抗炎作用，但不能降低病死率且存在一定的不良反应[13-20]。促炎因子的大量堆积可诱导上皮细胞和血管内皮细胞受损，使毛细血管通透性增加，炎性细胞浸润，导致肺组织损伤加重[3,21-22]。因此，抑制炎性介质的产生和释放是预防和治疗ALI/ARDS的关键节点。本研究中，模型组小鼠肺组织病理切片表现出弥漫性肺泡及肺间质水肿，其间充斥大量炎性细胞，亦可见淤血及微小血栓形成，与ALI/ARDS的病理表现一致；而甘露清瘟用药组肺组织炎性浸润减轻，间质水肿减少，提示甘露清瘟可减轻肺组织的炎症反应和肺水肿。

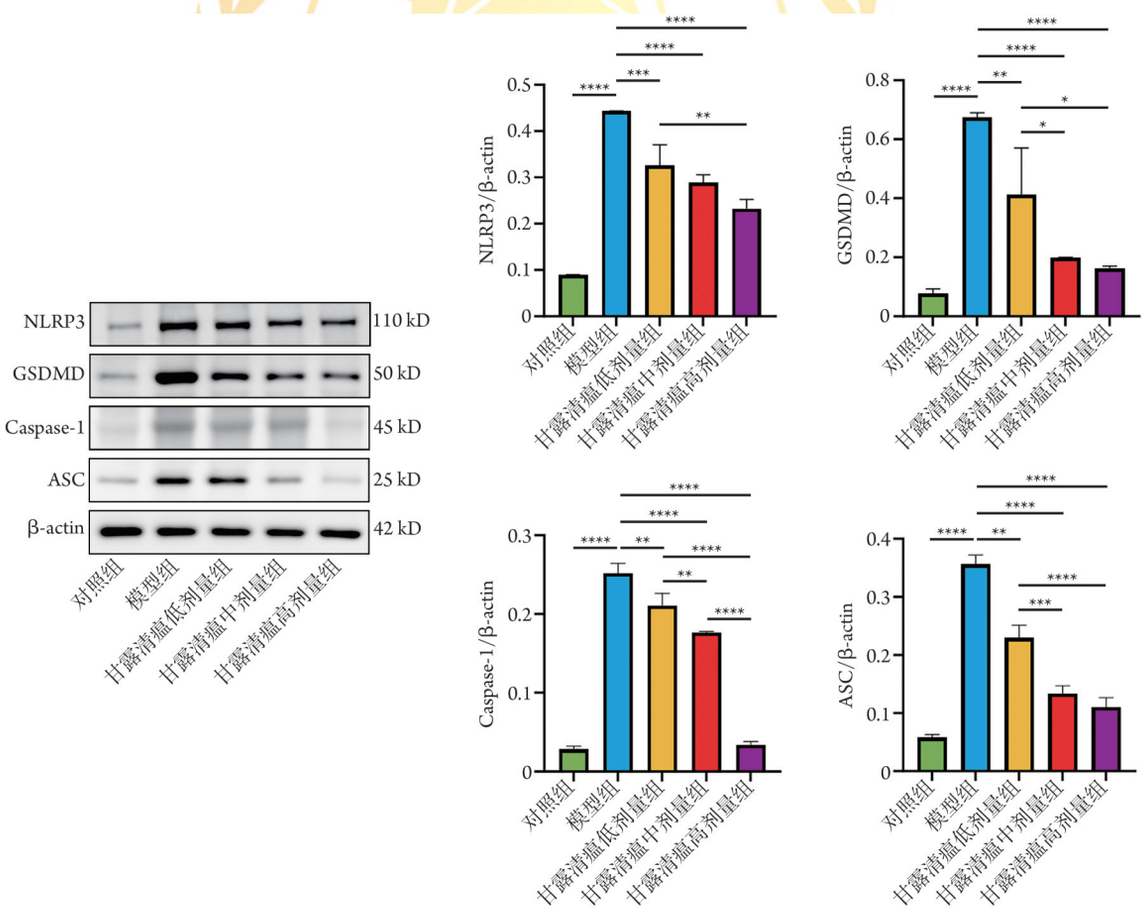
细胞焦亡是程序性细胞死亡的一种，其形态学表现为细胞肿胀甚至细胞膜破裂，染色质浓缩碎裂，DNA断裂，但细胞核结构完整，细胞内的炎性介质释放至细胞外，引发炎症反应[23-27]。细胞焦亡的分子机制可分为经典炎性途径和非经典炎性途径。经典炎性途径称为Caspase-1依赖性途径，即固有免疫细胞膜表面的膜识别受体(PRR)在感受到损伤相关分



A. 对照组；B. 模型组；C. 甘露清瘟低剂量组；D. 甘露清瘟中剂量组；E. 甘露清瘟高剂量组

图4 甘露清瘟方对小鼠肺组织病理学改变的影响

Fig.4 Effects of Ganluqingwen formula on lung histopathologic changes in mice of each group



NLRP3. NOD样受体热蛋白结构域相关蛋白3；GSDMD. 膜穿孔蛋白Gasdermin D；ASC. 凋亡相关斑点样蛋白；Caspase-1. 胱天蛋白酶-1；*P<0.05，**P<0.01，***P<0.001，****P<0.0001

图5 甘露清瘟方对小鼠肺组织中NLRP3、GSDMD、ASC、Caspase-1蛋白表达的影响

Fig.5 Effects of Ganluqingwen formula on expression of NLRP3, GSDMD, ASC and Caspase-1 proteins in mouse lung tissue

子模式(DAMP)或病原相关分子模式(PAMP)的刺激后，募集胞质中多种成分组装成炎性小体[28-30]。PPR包括Toll样受体(TLRs)、NOD样受体(NOD-like receptors，NLRs)、黑色素瘤缺乏因子样受体2

(absent in melanoma 2，AIM2)[31-33]，但仅NLRs能够直接组装成炎性小体[34]。当PRR感受到PAMP、DAMP刺激后，可通过激活核因子κB(nuclear factor kappa-B，NF-κB)导致NLRP3表达上调并激活，NLRP受体寡聚化，引起Pyrin结构域聚集，并通过Pyrin-Pyrin模式结合，招募ASC蛋白并形成复合物；ASC的CARD结构域随后募集pro-Caspase-1[35-37]。Pro-Caspase-1裂解释放Caspase-1，Caspase-1切割由肽键连接的GSDMD蛋白的N端效应结构域和C端抑制结构域，N端可插入细胞膜，在细胞膜表面形成“孔道”；Caspase-1切割IL-1β前体(pro-IL-1β)、pro-IL-18，使其变为具有活性的IL-1β、IL-18[38]；此时细胞内容物流出[39-40]，从而导致免疫反应的发生。非经典免疫途径亦称为非Caspase-1依赖途径，当其识别细菌细胞壁的LPS成分后，直接激活Caspase-4/-5/-11，不需组装炎性小体；与经典途径相似，活化的Caspase-4/-5/-11直接切割GSDMD[30,41]，使细胞内容物流出，进一步促进NLRP3炎性小体的组装[42-44]，导致焦亡的发生。细胞焦亡是由NLRP3介导的炎性程序性细胞死亡过程，NLRP3是炎症发生的关键调控因子[45-46]。因此，抑制NLRP3是抑制细胞焦亡发生的重要途径之一[47]。在细胞焦亡发生时，LPS通过TLRs识别并激活NF-κB信号通路，上调NLRP3的表达，为炎性小体的激活提供起始点[41]。NLRP3、ASC、Caspase-1组装成炎性小体，促进IL-1β、IL-18的释放。LPS亦能直接激活Caspase-4/-5/-11，进而切割GSDMD，诱导细胞焦亡的发生。NLRP3炎性小体在下游炎性细胞释放、募集更多的促炎因子及加重炎症反应的过程中发挥了重要作用[48]。大量促炎因子的释放导致细胞因子风暴，其中包括促炎因子TNF-α、IFN-γ、IL-4、IL-6、IL-12，以及抗炎因子IL-10的大量堆积[49-54]，因此，NLRP3可作为抑制炎症风暴和治疗ALI/ARDS的潜在靶点[55]。

综上所述，本研究结果显示，甘露清瘟可降低ALI/ARDS小鼠血清IFN-γ、TNF-α、IL-4、IL-12水平，上调IL-10水平；降低ALI/ARDS小鼠肺组织FN-γ、TNF-α、IIL-1β、IL-4、IL-6、IL-12水平，上调IL-10水平；下调小鼠肺组织NLRP3、ASC、Caspase-1、GSDMD蛋白的表达。总之，甘露清瘟可通过抑制炎性小体NLRP3的激活而抑制NLRP3/Caspase-1/GSDMD通路所致的细胞焦亡，减少炎性因子释放，减轻肺组织炎性细胞浸润，减轻急性肺损伤。本研究仍存在一定的局限性，如未对NLRP3、Caspase-1、GSDMD进行共定位，对通路蛋白之间的关系验证存在些许不足，有待后续研究进一步论证。

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