·论著·

中药清燥润肺化浊行血汤对PM2.5致肺损伤小鼠的干预作用

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【摘要】 目的 观察中药清燥润肺化浊行血汤对PM2.5致肺损伤小鼠炎症反应及组织病理学的影响，探讨中药防治雾霾致肺损伤的可能机制。**方法** 选择50只健康清洁级雄性C57BL/6小鼠，按随机数字表法分为对照组、模型组和低、中、高剂量清燥润肺化浊行血汤干预组5组，每组10只。采用经鼻腔滴注PM2.5悬液40 mg/kg的方法建立PM2.5致肺损伤小鼠模型，对照组滴注等量生理盐水。低、中、高剂量清燥润肺化浊行血汤干预组于制模后次日灌胃15、25、50 mL·kg^{-1}·d^{-1}清燥润肺化浊行血汤（组成：鸭梨75 g、川贝母10 g、百部8 g、半夏8 g、桔梗6 g、紫苑10 g、杏仁5 g、百合6 g、红景天4 g、荷叶3 g、路路通6 g、赤芍5 g、决明子6 g），共21 d；对照组和模型组给予等量生理盐水。于干预21 d后处死小鼠，取左侧肺脏进行支气管肺泡灌洗，收集支气管肺泡灌洗液（BALF），测定BALF中酸性磷酸酶（ACP）、碱性磷酸酶（AKP）、乳酸脱氢酶（LDH）及白蛋白（ALB）的含量；取右侧肺组织，苏木素－伊红（HE）染色后光镜下观察肺组织病理学改变。**结果** 经鼻腔滴注PM2.5悬液后，模型组BALF中ACP、AKP、LDH、ALB均较对照组明显升高〔ACP（U/L）：3.9±0.4比1.7±0.3，AKP（U/L）：9.0±1.5比4.8±0.3，LDH（U/L）：416.7±44.4比112.5±20.3，ALB（mg/L）：198.7±32.4比65.8±21.3，均$P<0.05$〕；光镜下显示，肺组织可见PM2.5颗粒聚积，肺泡间隔明显增厚，肺泡腔及肺间质炎性细胞浸润明显。经清燥润肺化浊行血汤干预后，BALF中生化指标明显改善，并呈剂量依赖性，高剂量清燥润肺化浊行血汤干预组各项指标明显低于模型组〔ACP（U/L）：2.1±0.8比3.9±0.4，AKP（U/L）：5.3±1.4比9.0±1.5，LDH（U/L）：146.6±29.8比416.7±44.4，ALB（mg/L）：88.5±26.7比198.7±32.4，均$P<0.05$〕；光镜下显示，肺组织病理学改变随干预剂量增加逐渐减轻。**结论** 清燥润肺化浊行血汤能减轻PM2.5引起的肺部炎症反应及组织损伤，且呈一定量效关系，其机制可能与其调节炎性介质免疫应答有关，为中药治疗PM2.5致肺损伤提供了依据。

【关键词】 清燥润肺化浊行血汤； PM2.5； 肺损伤； 炎症反应

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Intervention effect of Qingzao Runfei Huazhuo Xingxue decoction on PM2.5-induced pulmonary injury in mice

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【Abstract】 Objective To study the effects of Qingzao Runfei Huazhuo Xingxue decoction (QRHXD) on inflammatory reaction and histopathology in mice with PM2.5-induced pulmonary injury, and to approach the possible mechanism of prevention and treatment of traditional Chinese medicine on lung injury induced by haze. **Methods** Fifty healthy male C57BL/6 mice were randomly divided into five groups (n = 10): namely control, PM2.5, PM2.5 + low-, moderate-, and high-dose groups. The PM2.5 suspensions at a dosage of 40 mg/kg was respectively given to mice by the nasal instillation for reproduction of mouse model of lung injury induced by PM2.5, and the mice in control group were given the same volume of normal saline. The mice in PM2.5 + low-, moderate-, and high-dose QRHXD groups were given 15, 25, 50 mL·kg^{-1}·d^{-1} QRHXD by oral perfusion daily for consecutive 21 days at the next day of model reproduction (the QRHXD included: Pear 75 g, *Bulbus Fritillariae Cirrhosae* 10 g, *Radix Stemonae* 8 g, *Rhizoma Pinelliae* 8 g, *Radix Platycodi* 6 g, Aster 10 g, Almond 5 g, Lily 6 g, Rhodiola 4 g, Lotus 3 g, *Fructrs Liquidambaris* 6 g, *Radix Paeoniae Rubra* 5 g, Semen Cassiae 6 g). The mice in control and PM2.5 groups were given equivalent volume of normal saline respectively. After treatment for 21 days, the mice were sacrificed, and the left lung was harvested for bronchoalveolar lavage, and the bronchoalveolar lavage fluid (BALF) was collected for determination of levels of acid phosphatase (ACP), alkaline phosphatase (AKP), lactic dehydrogenase (LDH), and

albumin (ALB). The right lung was harvested for histopathology observation under light microscope using hematoxylin and eosine (HE) staining. **Results** After intranasal instillation of PM2.5 suspension, the levels of ACP, AKP, LDH, and ALB in PM2.5 group were significantly higher than those in control group [ACP (U/L): 3.9±0.4 vs. 1.7±0.3, AKP (U/L): 9.0±1.5 vs. 4.8±0.3, LDH (U/L): 416.7±44.4 vs. 112.5±20.3, ALB (mg/L): 198.7±32.4 vs. 65.8±21.3, all $P < 0.05$]. Under light microscope, the PM2.5 particles were collected, the alveolar septa were thickened, and the inflammatory cells in the alveolar cavity and pulmonary interstitium were found. On the contrary, after administration of QRHXD, a significant reduction of biochemical indexes was found, which showed a dose-dependent manner. The parameters of PM2.5 + high-dose QRHXD group were significantly lower than those in PM2.5 group [ACP (U/L): 2.1±0.8 vs. 3.9±0.4, AKP (U/L): 5.3±1.4 vs. 9.0±1.5, LDH (U/L): 146.6±29.8 vs. 416.7±44.4, ALB (mg/L): 88.5±26.7 vs. 198.7±32.4, all $P < 0.05$]. At the same time, the pathological changes in lung tissue were better with the increase of the dose. **Conclusions** QRHXD can reduce the pulmonary inflammatory response and tissue damage caused by PM2.5, with the increase concentration of Chinese medicine, and the effect is more obvious. This may be related to the immune response of the human body to regulate inflammatory mediators, which provide basis for the treatment of pulmonary injury induced by PM2.5.

【Key words】 Qingzao Runfei Huazhuo Xingxue decoction; PM2.5; Lung injury; Inflammatory reaction
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有研究表明，长期暴露于被高浓度细颗粒物 PM2.5 污染的空气中，会增加肺部疾病的发生风险，如慢性阻塞性肺疾病（COPD）[1-3]。流行病学研究表明，空气污染细颗粒物与呼吸系统疾病的发病率和病死率明显相关[4]。Zanobetti 等[5]研究发现，PM2.5 平均每天升高 10 μg/m^3，冠心病发病率升高 1.89%，心肌梗死发病率升高 2.25%，呼吸系统疾病危险度升高 2.07%；Dominici 等[6]研究也表明，PM2.5 每天升高 10 μg/m^3，呼吸系统疾病发病率升高 8.0%。因此，防治雾霾导致的呼吸系统相关疾病是医务工作者亟待解决的重大问题。本课题组前期研究表明，中药清燥润肺化浊行血汤对 PM2.5 致呼吸系统疾病小鼠疗效确切[7]，但具体机制尚未明确。本研究通过观察清燥润肺化浊行血汤对经鼻腔滴注 PM2.5 诱导肺损伤小鼠肺组织的保护作用，探讨其可能的作用机制。

1　材料与方法

1.1　PM2.5 悬液制备：2016 年 3 月 1 日至 2016 年 4 月 1 日用 Thermo Anderson 采样器采集烟台市环境空气质量监测一号站、二号站、三号站大气 PM2.5，将采有 PM2.5 的滤膜浸入三蒸水中洗脱细颗粒物，过滤后将滤液经真空干燥机干燥、蒸发，用生理盐水配制成 4 mg/μL PM2.5 悬液，高压灭菌后 4℃保存。

1.2　实验动物及分组：健康清洁级雄性 C57BL/6 小鼠 50 只，周龄 4～5 周，体重 25～30 g，购自苏州爱尔麦特科技有限公司，动物许可证号：SCXK（苏）2014-0007。于 SPF 实验室适应性喂养 1 周后，按随机数字表法分为对照组、模型组和低、中、高剂量清燥润肺化浊行血汤干预组 5 组，每组 10 只。

1.3　模型制备：分别于饲养 7、14、21 d 经鼻腔滴注 PM2.5 悬液 40 mg/kg 制模；对照组滴注等量生理盐水。低、中、高剂量清燥润肺化浊行血汤干预组于制模后次日灌胃 15、25、50 mL·kg^{-1}·d^{-1} 清燥润肺化浊行血汤（组成：鸭梨 75 g、川贝母 10 g、百部 8 g、半夏 8 g、桔梗 6 g、紫苑 10 g、杏仁 5 g、百合 6 g、红景天 4 g、荷叶 3 g、路路通 6 g、赤芍 5 g、决明子 6 g，北京康仁堂药业有限公司全成分中药配方颗粒），共 21 d；对照组和模型组给予等量生理盐水。

1.4　观察指标及方法

1.4.1　支气管肺泡灌洗液（BALF）的制备：腹腔注射 1% 戊巴比妥麻醉后引颈脱臼处死小鼠，开胸取双侧肺组织，在主气管上插入导管至下端分叉处，注入 2 mL 磷酸盐缓冲液（PBS）灌洗左肺，反复冲洗 3 次，回收 BALF，4 ℃离心取上清液，20 ℃保存。

1.4.2　BALF 中生化指标的测定：对 BALF 进行成分分析，测定 BALF 中酸性磷酸酶（ACP）、碱性磷酸酶（AKP）、乳酸脱氢酶（LDH）及白蛋白（ALB）的含量，严格按试剂盒说明书要求操作。

1.4.3　肺组织病理学观察：取右侧肺组织，40 g/L 多聚甲醛固定 48 h 后石蜡包埋，制作厚度为 5 μm 的切片，苏木素－伊红（HE）染色后，光镜下观察肺组织病理学改变。

1.5　统计学方法：采用 SPSS 17.0 软件对数据进行统计学处理，计量资料以均数 ± 标准差（$\bar{x}\pm s$）表示，两组间比较采用 t 检验，多组间比较采用单因素方差分析。以 $P<0.05$ 表示差异有统计学意义。

2　结　果

2.1　BALF 中生化指标的变化（表 1）：经鼻腔滴注 PM2.5 诱导小鼠肺损伤后，BALF 中 ACP、AKP、LDH、ALB 水平均显著高于对照组；清燥润肺化浊行血汤干预组 ACP、AKP、LDH、ALB 均较模型组明显下降，且呈剂量依赖性（均 $P<0.05$）。

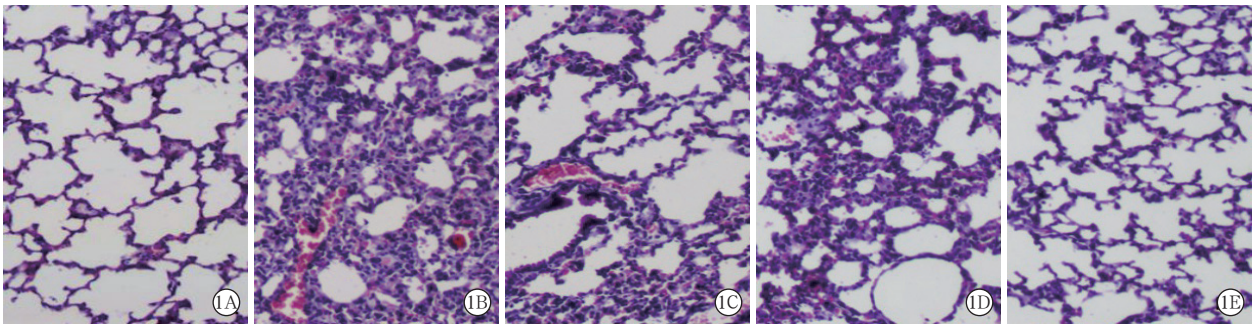

图 1 光镜下观察各组小鼠肺组织病理学改变 对照组（A）肺泡结构清晰完整，肺泡间隔无水肿，肺泡腔内有微量炎性细胞浸润，未见颗粒物聚积；PM2.5 致肺损伤模型组（B）肺组织可见 PM2.5 颗粒聚积，肺泡间隔明显增厚，肺泡腔及肺间质炎性细胞浸润明显，以中性粒细胞、淋巴细胞为主，伴有肺泡腔缩小；低、中、高剂量清燥润肺化浊行血汤干预组（C～E）肺泡间隔增厚及炎性细胞浸润程度均较模型组改善，且随干预剂量增加，炎症反应逐渐减轻 HE 中倍放大

表 1 不同剂量清燥润肺化浊行血汤对 PM2.5 致肺损伤小鼠 BALF 中生化指标的影响（$\bar{x}\pm s$）

组别	动物数（只）	ACP（U/L）	AKP（U/L）	LDH（U/L）	ALB（mg/L）
对照组	10	1.7±0.3	4.8±0.3	112.5±20.3	65.8±21.3
模型组	10	3.9±0.4^{a}	9.0±1.5^{a}	416.7±44.4^{a}	198.7±32.4^{a}
低剂量组	10	3.2±0.7^{a}	7.9±2.2^{a}	325.4±36.2^{ab}	148.4±26.1^{a}
中剂量组	10	2.4±1.0^{abc}	6.2±2.3^{abc}	220.2±20.2^{abc}	124.2±30.2^{ab}
高剂量组	10	2.1±0.8^{bc}	5.3±1.4^{bc}	146.6±29.8^{bc}	88.5±26.7^{bc}

注：BALF 为支气管肺泡灌洗液，ACP 为酸性磷酸酶，AKP 为碱性磷酸酶，LDH 为乳酸脱氢酶，ALB 为白蛋白；与对照组比较，$^{a}P<0.05$；与模型组比较，$^{b}P<0.05$；与低剂量组比较，$^{c}P<0.05$

2.2 肺组织病理学改变（图 1）：对照组小鼠肺组织无明显病理学改变；模型组可见明显肺损伤表现，肺组织可见 PM2.5 颗粒聚积，且炎性细胞浸润明显；与模型组比较，低、中、高剂量清燥润肺化浊行血汤干预组肺损伤表现明显减轻，炎性细胞浸润明显改善，且呈剂量依赖性。

3 讨 论

近年来，雾霾的高频率大范围出现导致肺部疾病发生率呈直线上升趋势。大量流行病学数据表明，短期 PM2.5 暴露与肺损伤的发生有直接关系[8-10]。本研究采用经鼻腔滴注 PM2.5 混悬液制备肺损伤小鼠模型，通过数据统计及病理学观察发现，短期 PM2.5 暴露确实可以增加肺部的炎症反应，再次证实了雾霾对呼吸系统的负面影响。

肺脏是大气 PM2.5 损害的首要器官，可能与下列机制有关：① 雾霾刺激会导致肺组织中免疫细胞激活，诱导氧化应激及炎症信号通路，导致亚细胞结构和功能损伤[11-12]；② 雾霾中的有毒成分可以通过诱导细胞凋亡[13]或自噬[14]等机制使细胞死亡，并引起局部炎症，甚至导致肺泡萎缩、细支气管重塑及肺组织纤维化[15]；③ 长期的 PM2.5 暴露可改变肺脏正常的免疫功能，使免疫球蛋白 IgE 表达升高等[16]，从而促进 COPD[17]、慢性支气管炎[18]及哮喘[19]等疾病的发生发展；④ PM2.5 还可通过影响转铁蛋白对 Fe^{3+} 的运输[20]、改变 Toll 样受体（TLR）的表达[21]及干扰细胞内微管系统等多种机制影响肺巨噬细胞的抗菌作用，损伤肺的非特异和特异性免疫，导致感染性疾病的发生及恶化[22]。

目前，西医针对雾霾致肺损伤采取的防治措施主要是佩戴防尘防毒口罩以及安装空气净化器来预防呼吸道的不适反应；一旦导致肺损伤主要应用抗菌药物治疗，但效果不明显或副作用较多[9,23]。本课题组在王新陆教授“血浊污肺”理论[24]的基础上，自拟康肺散结汤联合化浊行血汤组成清燥润肺化浊行血汤，用于治疗雾霾导致的肺损伤，能明显改善呼吸困难、咳嗽痰多及恶心、呕吐、眩晕等症状。前期研究表明，此方能显著改善 PM2.5 所致呼吸系统的炎性损伤，改善病理炎症[7,25]，但其机制尚未明确。本研究对清燥润肺化浊行血汤的起效机制进行深入研究，使其更适合推广应用。

中医认为，呼吸系统疾病的主要发病机制为机体正气不足，虚邪贼风乘虚而入，导致气血阴阳、脏腑功能失调，故增强正气、防御外邪入侵可以“防病于未然、既病防变”[26-27]。《灵枢·邪客》曰：“宗气积于胸中，出于喉咙，以贯心脉，而行呼吸焉。”《灵枢·营卫生会》曰：“中焦亦并胃中，出上焦之后，此所受气者，泌糟粕，蒸津液，化其精微，上注于肺脉乃化而为血。”可见血的生成与肺有密切关系。《灵枢·阴阳清浊》曰：“受气者清。”污浊之邪气弥漫空中，随天之清气同入于肺中，致宗气浊而不清，“贯心脉后”浊邪随而入血，沉积血中，是为血浊。王新陆教授提出，血浊是现代诸多疾病的病理枢纽，血浊日久可损伤人体正气，并能化生痰浊、瘀血、热毒，亦可化燥、伤津、损络，血浊内生而污肺[24,28-29]。

吸入雾霾中细颗粒物后直接作用于呼吸道，引起肺部细胞通透性改变，激活各种蛋白及酶类，导致炎症反应的发生。ACP、AKP 、LDH 是反映细胞炎性损伤的早期敏感指标，是细胞毒性标志物；ALB 是反映肺泡气血屏障完整性的指标。本研究显示，清燥润肺化浊行血汤能显著降低 PM2.5 致肺损伤小鼠 BALF 中 ACP、AKP、LDH、ALB 含量，且剂量越高，指标下降越明显，表明清燥润肺化浊行血汤能减轻雾霾导致的肺损伤；光镜下显示，肺组织病理学改变随干预剂量增加而逐渐减轻，表明清燥润肺化浊行血汤能减轻 PM2.5 引起的肺部炎症反应。

综上，清燥润肺化浊行血汤对 PM2.5 导致的肺组织损伤及炎症反应有明显的改善作用，且呈一定量效关系，其机制可能与调节炎性因子免疫应答，促进免疫细胞激活，减轻氧化应激反应有关，为 PM2.5 引起的呼吸系统疾病提供了一种新的诊疗思路。

参考文献

[1] Calverley PM, Walker P. Chronic obstructive pulmonary disease [J]. Lancet, 2003, 362 (9389): 1053–1061.

[2] Pauwels R. Global initiative for chronic obstructive lung diseases (GOLD): time to act [J]. Eur Respir J, 2001, 18 (6): 901–902.

[3] Buist AS, McBurnie MA, Vollmer WM, et al. International variation in the prevalence of COPD (the BOLD Study): a population–based prevalence study [J]. Lancet, 2007, 370 (9589): 741–750. DOI: 10.1016/S0140–6736(07)61377–4.

[4] Brunekreef B, Holgate ST. Air pollution and health [J]. Lancet, 2002, 360 (9341): 1233–1242. DOI: 10.1016/S0140–6736(02)11274–8.

[5] Zanobetti A, Franklin M, Koutrakis P, et al. Fine particulate air pollution and its components in association with cause–specific emergency admissions [J]. Environ Health, 2009, 8: 58. DOI: 10.1186/1476–069X–8–58.

[6] Dominici F, Peng RD, Bell ML, et al. Fine particulate air pollution and hospital admission for cardiovascular and respiratory diseases [J]. JAMA, 2006, 295 (10): 1127–1134. DOI: 10.1001/jama.295.10.1127.

[7] 张金波，孙丽，李史清，等. 中药清燥润肺化浊行血汤对 PM2.5 致小鼠呼吸系统疾病的治疗作用 [J]. 中华危重病急救医学，2016, 28 (10): 916–920. DOI: 10.3760/cma.j.issn.2095–4352.2016.10.011.
Zhang JB, Sun L, Li SQ, et al. Therapeutic effect of Qingzao Runfei Huazhuo Xingxue decoction on PM2.5–induced respiratory disease in mice [J]. Chin Crit Care Med, 2016, 28 (10): 916–920. DOI: 10.3760/cma.j.issn.2095–4352.2016.10.011.

[8] Li P, Xin J, Wang Y, et al. The acute effects of fine particles on respiratory mortality and morbidity in Beijing, 2004–2009 [J]. Environ Sci Pollut Res Int, 2013, 20 (9): 6433–6444. DOI: 10.1007/s11356–013–1688–8.

[9] Tsai SS, Chiu HF, Liou SH, et al. Short–term effects of fine particulate air pollution on hospital admissions for respiratory diseases: a case–crossover study in a tropical city [J]. J Toxicol Environ Health A, 2014, 77 (18): 1091–1101. DOI: 10.1080/15287394.2014.922388.

[10] Strak M, Janssen NA, Godri KJ, et al. Respiratory health effects of airborne particulate matter: the role of particle size, composition, and oxidative potential–the RAPTES project [J]. Environ Health Perspect, 2012, 120 (8): 1183–1189. DOI: 10.1289/ehp.1104389.

[11] Gualtieri M, Ovrevik J, Mollerup S, et al. Airborne urban particles (Milan winter–PM2.5) cause mitotic arrest and cell death: Effects on DNA, mitochondria, AhR binding and spindle organization [J]. Mutat Res, 2011, 713 (1–2): 18–31. DOI: 10.1016/j.mrfmmm.2011.05.011.

[12] Huang Q, Zhang J, Peng S, et al. Effects of water soluble PM2.5 extracts exposure on human lung epithelial cells (A549): A proteomic study [J]. J Appl Toxicol, 2014, 34 (6): 675–687. DOI: 10.1002/jat.2910.

[13] Longhin E, Holme JA, Gutzkow KB, et al. Cell cycle alterations induced by urban PM2.5 in bronchial epithelial cells: characterization of the process and possible mechanisms involved [J]. Part Fibre Toxicol, 2013, 10 : 63. DOI: 10.1186/1743–8977–10–63.

[14] Deng X, Zhang F, Rui W, et al. PM2.5–induced oxidative stress triggers autophagy in human lung epithelial A549 cells [J]. Toxicol In Vitro, 2013, 27 (6): 1762–1770. DOI: 10.1016/j.tiv.2013.05.004.

[15] Riva DR, Magalhães CB, Lopes AA, et al. Low dose of fine particulate matter (PM2.5) can induce acute oxidative stress, inflammation and pulmonary impairment in healthy mice [J]. Inhal Toxicol, 2011, 23 (5): 257–267. DOI: 10.3109/08958378.2011.566290.

[16] Ogino K, Zhang R, Takahashi H, et al. Allergic airway inflammation by nasal inoculation of particulate matter (PM2.5) in NC/Nga mice [J]. PLoS One, 2014, 9 (3): e92710. DOI: 10.1371/journal.pone.0092710.

[17] Tsai SS, Chang CC, Yang CY. Fine particulate air pollution and hospital admissions for chronic obstructive pulmonary disease: a case–crossover study in Taipei [J]. Int J Environ Res Public Health, 2013, 10 (11): 6015–6026. DOI: 10.3390/ijerph10116015.

[18] Nachman KE, Parker JD. Exposures to fine particulate air pollution and respiratory outcomes in adults using two national datasets: a cross–sectional study [J]. Environ Health, 2012, 11: 25. DOI: 10.1186/1476–069X–11–25.

[19] Jung KH, Hsu SI, Yan B, et al. Childhood exposure to fine particulate matter and black carbon and the development of new wheeze between ages 5 and 7 in an urban prospective cohort [J]. Environ Int, 2012, 45 : 44–50. DOI: 10.1016/j.envint.2012.03.012.

[20] Corsini E, Budello S, Marabini L, et al. Comparison of wood smoke PM2.5 obtained from the combustion of FIR and beech pellets on inflammation and DNA damage in A549 and THP–1 human cell lines [J]. Arch Toxicol, 2013, 87 (12): 2187–2199. DOI: 10.1007/s00204–013–1071–z.

[21] Zhao C, Liao J, Chu W, et al. Involvement of TLR2 and TLR4 and Th1/Th2 shift in inflammatory responses induced by fine ambient particulate matter in mice [J]. Inhal Toxicol, 2012, 24 (13): 918–927. DOI: 10.3109/08958378.2012.731093.

[22] Jedrychowski WA, Perera FP, Spengler JD, et al. Intrauterine exposure to fine particulate matter as a risk factor for increased susceptibility to acute broncho–pulmonary infections in early childhood [J]. Int J Hyg Environ Health, 2013, 216 (4): 395–401. DOI: 10.1016/j.ijheh.2012.12.014.

[23] Mosbah AA, Abdellatif NA, Sorour EI, et al. Serum SP–D levels as a biomarker of lung injury in children suffering of bronchopneumonia [J]. J Egypt Soc Parasitol, 2012, 42 (1): 25–32.

[24] 王新陆. 关于血浊理论在现代疾病谱系中作用与地位的探讨 [J]. 天津中医药，2011, 28 (5): 355–357.
Wang XL. The study on effect and position of turbid blood theory in modern spectrum of disease [J]. Tianjin J TCM, 2011, 28 (5): 355–357.

[25] 张金波，慕岳峻. 中药在维持化疗中作用研究的新进展 [J]. 中国中西医结合急救杂志，2015, 22 (6): 669–672. DOI: 10.3969/j.issn.1008–9691.2015.06.033.
Zhang JB, Mu YJ. New progress in the study of the importance of maintenance chemotherapy [J]. Chin J TCM WM Crit Care, 2015, 22 (6): 669–672. DOI: 10.3969/j.issn.1008–9691.2015.06.033.

[26] 来薛，蔡哲，俞庆声，等. 固本止咳中药对慢性阻塞性肺疾病小鼠肺功能及呼吸道分泌 sIgA 的影响 [J]. 中华中医药杂志，2012, 27 (12): 3064–3067.
Lai X, Cai Z, Yu QS, et al. Effect of Gubenzhike recipe on pulmonary function and sIgA in respiratory tract of COPD mice [J]. CJTCMP, 2012, 27 (12): 3064–3067.

[27] Hwang BF, Chen YH, Lin YT, et al. Relationship between exposure to fine particulates and ozone and reduced lung function in children [J]. Environ Res, 2015, 137: 382–390. DOI: 10.1016/j.envres.2015.01.009.

[28] 王新陆. 论“血浊”与“治未病” [J]. 天津中医药，2008, 25 (3): 177–180.
Wang XL. Discussion on blood turbid and treating disease before attack [J]. Tianjin J TCM, 2008, 25 (3): 177–180.

[29] 王新陆. 血浊证的辨证治疗 [J]. 山东中医杂志，2007, 26 (1): 3–5. DOI: 10.3969/j.issn.0257–358X.2007.01.001.
Wang XL. Diagnosis and treatment of syndrome of turbid blood [J]. Shandong J TCM, 2007, 26 (1): 3–5. DOI: 10.3969/j.issn.0257–358X.2007.01.001.

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