以動物模式評估巴拉刈急性肺傷害之治療及慢性肺纖維化之機轉

Evaluation of Acute Treatment and Chronic Fibrosis Mechanism in a Rat Model of Paraquat-Induced Lung Injury

中文摘要

我們以巴拉刈(paraquat)中毒引起急性肺傷害的老鼠模式，研究 methylprednisolone 治療對其血液氣體交換、壓力體積曲線、支氣管肺泡灌洗液發炎細胞數目、灌洗液蛋白質量、肺臟 surfactant 容量、及肺組織的影響。23 隻成年公 Sprague-Dawley 老鼠接受腹腔內注射 paraquat(35 毫克/公斤體重)後，被隨機分成三組：(1)對照組:沒有接受更進一步的處理；(2)一劑量的 methylprednisolone 組:腹腔內注射巴拉刈時，同時接受一次腹腔內注射 methylprednisolone(30 毫克/公斤體重)；(3)三劑量的 methylprednisolone 組:腹腔內注射巴拉刈時，同時接受一次和每日的腹腔內注射 methlprednisolone 注射(30 毫克/公斤體重)。在 paraquat 注射之後的第 3 天，老鼠接受 90 分鐘的機械性呼吸後、作靜態壓力曲線、支氣管肺泡灌洗、及肺組織學檢查。當與控制組和一劑量 methylprednisolone 組相比較時，三劑量 methylprednisolone 組的肺臟 surfactant 容量有顯著地增加。Methylprednisolone 治療可以增加血液氧化程度，其數值在三劑量 methylprednisolone 組於機械呼吸 90 分鐘時最高。支氣管肺泡灌洗液的發炎性細胞數目和肺傷害分數隨著 methylprednisolone 劑量的增加而減少。Transforming grwoth factor-beta1(TGF-beta1)會促進受傷害器官產生纖維變性。在心臟和腎臟的細胞裡，angiotensin II (Ang II)會誘導 TGF-beta1 產生，但在肺組織裡 Ang II 對 TGF-beta1 的調節仍然所知不多。因此我們評估 TGF-beat1 在 paraquat 中毒引起的肺纖維變性裡所扮演的角色和它與 Ang II 的關係。成年的公 Sprague-Dawley 老鼠接受腹腔內注射 paraquat(20 毫克/公斤體重)，或者在對照組腹腔內注射等量生理食鹽水。在 paraquat 處理之後的第一、三、七、及二十一天時，我們測量肺組織 TGF-beta1 和 collagen 基因的表現，TGF-beat1 蛋白質，angiotensin converting enzyme (ACE) 的活性、Ang II 及 hydroxyproline 的量。肺組織 TGF-beta1 mRNA 的表現在 paraquat 處理之後逐漸增加，在第七天時達最高。肺組織 TGF-beta1 mRNA 及 TGF-beta1 蛋白質量先增加，再伴隨 collagen type I mRNA 的表現和 hydroxyproline 量增加。c-myc mRNA 的表現模式與肺組織 TGF-beta1 蛋白質的量成相反關係。肺組織 ACE 活性在 paraquat 處理之後減少，減少量在第七天時最大。肺組織 Ang II 量在給予 paraquat 後立即減少，而且其數值與 TGF-beta1 沒有關係存在。

結論：我們發現高劑量的 methylprednisolone 治療可以增加肺臟 surfactant 的量，並且改善急性 paraquat 肺傷害的組織變化，但是並不能改進整個機械呼

吸期的肺臟氣體交換。在 paraquat 引起的慢性肺纖維化時，TGF-beta1 是被向上調節地，但這種變化並不倚賴 renin-angiotensin 系統。

英文摘要

We examined the effects of methylprednisolone on gas exchange, pressure-volume curve, lavage fluid inflammatory cell counts, protein content, surfactant pool size, and lung histology in a rat model of paraquat-induced lung injury. Twenty-three adult male Sprague-Dawley rats received intraperitoneal paraquat injection (35 mg/kg) and were randomly divided into three groups: (1) control group received no further treatment; (2) 1-dose methylprednisolone group received a concomitant intraperitoneal methylprednisolone injection (30 mg/kg); (3) 3-dose methylprednisolone group received a concomitant and daily intraperitoneal methylprednisolone injection (30 mg/kg) for 3 doses. Three days after paraquat injection, the rat was ventilated for 90 min, a static pressure-volume curve and bronchoalveolar lavage was performed, and postmortem histology was examined. Surfactant pool size of the 3-dose methylprednisolone group was significantly increased when compared with the control and 1-dose methylprednisolone groups. Methylprednisolone treatment increased oxygenation and the value was statistically significant for 3-dose methylprednisolone group at 90 min of ventilation. Inflammatory cell counts in bronchoalveolar lavage fluid and lung injury score were decreased as the methylprednisolone dose increased.

Transforming growth factor-beta1 (TGF-beta1) contributes to the fibrosis of injured organs. Angiotensin-II (Ang II) is an inducer of TGF-beta1 in cells of the heart and kidneys, and the regulation of TGF-beta1 by Ang II has not yet been confirmed in lung tissue. We evaluated the role of TGF-beta1 and its relationship with Ang II in paraquat-induced lung fibrosis. Adult male Sprague-Dawley rats were treated intraperitoneally with paraquat (20 mg/kg) or saline in the control group. On days 1, 3, 7, and 21 after paraquat treatment, TGF-beta1 and collagen expressions, TGF-beta1 protein, angiotensin-converting enzyme (ACE) activity, Ang II, and hydroxyproline contents were measured in lung tissue. Lung TGF-beta1 mRNA expression progressively increased and reached a peak on day 7 after paraquat treatment. Increases in TGF-beta1 mRNA expression and TGF-beta1 levels preceded the onset of increased collagen I mRNA expression and hydroxyproline contents. c-myc mRNA expressions were inversely correlated with TGF-beta1 protein levels in paraquat-treated lungs. Lung ACE activity decreased after paraquat administration and the decrement was maximal on day 7. Lung Ang II concentrations immediately decreased after paraquat administration and the values were not related to TGF-beta1 levels.

We conclude that high-dose methylprednisolone treatment increased surfactant pool size and improved lung histology in acute stage of paraquat-injured lungs but this augmentation could not significantly improve oxygenation throughout the ventilation period and that TGF-beta1 is upregulated and contribute to the paraquat-induced lung fibrosis and this effect is independent of the renin-angiotensin system.