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Title: Effects of PEEP-like cyclic stretch on the IL-6 protein production in normal human pulmonary artery endothelial cells in vitro

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Body: Background and Aim: Excessive cyclic stretch is one of the main causes of ventilator-induced lung injury. However, the molecular mechanisms of the injury by cyclic stretch have not yet been fully understood. The aim of the present study was to examine the effects of PEEP-like cyclic stretch on the IL-6 protein production in human pulmonary artery endothelial cells (HPAECs) in vitro. Methods: Normal HPAECs were stretched by a Flexcell[®] FX-4000T[™] Tension System (Flexcell International). The stretching rate was 15 cycles/minute, and the cells were cyclicly stretched from 0 to 5%, 0 to 10%, 0 to 20%, 3 to 20% and 5 to 20%. Stretch from 3 to 20% and from 5 to 20% simulated excessive stretch during mechanical ventilation with PEEP. The cells were stretched for various durations (0, 1, 3, 6 and 12 hours). During the experiments, culture medium was sampled 0, 3, 6 and 12 hours after stretch started. The IL-6 concentration of the samples was determined by ELISA. Results: Excessive stretch (0 to 20%) significantly increased the IL-6 production of the cells stretched for more than 3 hours compared to the unstretched cells (n=5, P<0.05), but moderate stretch (0 to 5% and 0 to 10%) did not. PEEP-like stretch (3 to 20% and 5 to 20%) produced no significant changes in the IL-6 protein production of the cells at any sampling points. Conclusions: The IL-6 protein was produced by excessive cyclic stretch (0 to 20%). However, the IL-6 production was significantly suppressed by PEEP-like cyclic stretch (3 to 20% and 5 to 20%). This result suggests that HPAECs would be protected by PEEP during mechanical ventilation even if the cells are excessively stretched.