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## Terbutaline lessens protein fluxes across the alveolo-capillary barrier during high-volume ventilation

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**Abstract Objective:** To evaluate whether a  $\beta_2$ -adrenergic agonist may reduce acute alveolo-capillary barrier alterations during high-volume ventilation. **Design:** Experimental study. **Setting:** Animal research laboratory. **Subjects:** A total of 48 male Wistar rats. **Interventions:** A zone of alveolar flooding was produced by liquid instillation in a distal airway. Proteins in the instilled solution were traced with  $^{99m}\text{Tc}$ -albumin.  $^{111}\text{In}$ , which binds to transferrin, was injected into the systemic circulation. Terbutaline was administered in the instilled solution or intra-peritoneally. Conventional ventilation was applied for 30 min followed by different ventilation strategies for 90 min: conventional ventilation, high-volume ventilation with or without 6 cmH<sub>2</sub>O PEEP. **Measurements and main**

**results:** Protein fluxes across the alveolar and microvascular barriers were evaluated by scintigraphy. High-volume ventilation resulted in immediate leakage of  $^{99m}\text{Tc}$ -albumin from alveolar spaces and increased pulmonary uptake of systemic  $^{111}\text{In}$ -transferrin. Terbutaline in the instilled solution and PEEP lessened alveolar  $^{99m}\text{Tc}$ -albumin leakage and pulmonary  $^{111}\text{In}$ -transferrin uptake due to high-volume ventilation, whereas terbutaline given intra-peritoneally only lessened  $^{111}\text{In}$ -transferrin uptake. Terbutaline in the instilled solution also lessened the increase in lung wet-to-dry weight ratio due to high-volume ventilation. **Conclusions:** Terbutaline reduces protein fluxes across the alveolar epithelial and pulmonary microvascular barriers during high-volume ventilation in vivo. The route of administration may be important.

**Keywords** Pulmonary edema · Intermittent positive pressure ventilation · Beta-adrenergic agonists · Radionuclide imaging

### Introduction

Acute respiratory distress syndrome (ARDS), the most severe form of acute lung injury, is characterized by a permeability-type pulmonary edema resulting in severe life-threatening hypoxemia [1]. Experimental studies suggested that unnecessary lung tissue stress could have

played a role in the poor prognosis of this diseased state [2]. As a matter of fact, reducing the mechanical stress applied to the lungs by lowering tidal volume from 12 to 6 ml/kg improved the survival of ARDS patients by 22% [3]. However, the overall prognosis of acute lung injury remains poor, with mortality rates ranging from 15% to 60% depending on patient age [4].

Pharmacological therapies have been tested for many years in ARDS patients, but none of them have improved survival [5]. A randomized placebo-controlled trial showed that prophylactic inhalation of salmeterol, a long-acting  $\beta_2$ -agonist, decreased by 50% the incidence of high-altitude pulmonary edema in volunteers known to be at risk [6]. A randomized trial recently suggested that salbutamol infusion might lessen plateau airway pressure and decrease extravascular lung water in ARDS patients [7]. The protective effect of  $\beta_2$ -adrenergic agonists may be related in part to increased alveolar liquid clearance [8], but perhaps also to the lessening of microvascular and/or alveolar permeability alterations that accompany permeability-type pulmonary edema [9]. Beta-adrenergic agonist infusion reduced endothelial permeability in a model of acid-induced lung injury [10] and pulmonary edema in an ovine model of burn and smoke inhalation [11], indicating that it decreased transvascular fluid flux. Basran et al. [12] found that terbutaline infusion reduced accumulation of plasma protein in the lungs of ARDS patients, which suggested that it might have lessened microvascular permeability alterations.

Whether  $\beta_2$ -adrenergic agonists may similarly lessen the increase in alveolar permeability produced by mechanical stress is unknown. Parker et al. [13] showed that isoproterenol attenuated microvascular permeability increase due to high transmural microvascular pressures in isolated rat lungs. However, improving permeability on one side of the barrier may be offset by an absence of improvement on the other side, because the responses of the epithelial and endothelial layers sometimes differ [14]. Therefore, we used a previously validated model based on scintigraphic imaging [15] to non-invasively study the effect of a  $\beta_2$ -adrenergic agonist on protein fluxes across the alveolo-capillary barrier during high-volume ventilation.

## Material and methods

### Animals

All experiments were conducted in compliance with the recommendations for laboratory animal research of the European Union and the French Ministry of Agriculture. Male Wistar rats ( $n = 48$ , weighing 295–345 g, Elevage Janvier, Le Genest Saint Isle, France) were anesthetized by intra-peritoneal injection of 50 mg/kg pentobarbital (Sigma, St Quentin Fallavier, France). They were tracheotomized and a cannula (Braun, Melsungen, Germany) was tightly secured to the trachea to avoid gas leaks.

### Ventilation modalities

Rats were paralyzed by injection of 15 mg/kg succinylcholine (Sigma) and ventilated using a Harvard rodent

volume ventilator (Ealing, Courtaboeuf, France). Additional 10 mg/kg pentobarbital injections were performed every 30 min. Rats not injected with succinylcholine had no nociceptive reaction with this protocol. Conventional ventilation (CV; 8 ml/kg  $V_T$ , PEEP 2 cmH<sub>2</sub>O, respiratory rate 70/min) was applied for 30 min, followed by different ventilation modalities for 90 min. Several ventilation modalities were tested: CV, high-volume ventilation (HV) at 30 cmH<sub>2</sub>O end-inspiratory pressure (Pei) and no PEEP (HV1) and HV with PEEP set at 6 cmH<sub>2</sub>O (HV1P) and  $V_T$  adjusted to be about the same (25 ml/kg) as during HV1. This latter ventilation modality resulted in a slightly higher Pei (Table 1). Pei during HV was very close to plateau pressure considering the low respiratory rate and the shape of the ventilator waveform, which was nearly sinusoidal [16]. A less injurious strategy was used when terbutaline was given intra-peritoneally (IP; see below), in order to increase sensitivity. Pei was set at 25 cmH<sub>2</sub>O (HV2), which was shown to increase permeability [15], respiratory rate being set the same as during 30 cmH<sub>2</sub>O Pei ventilation. During these experiments a catheter was placed in a carotid artery and arterial pressure was measured using a Statham pressure transducer and registered at 15-min intervals.

### Scintigraphic imaging

Scintigraphic imaging was performed as previously described [15]. Briefly, continuous planar thoracic acquisitions were performed ( $\gamma$  Imager, Biospace, Paris, France) that lasted 120 min without interruption. Dynamic series of images of 150 s each were computed from the registered scintigraphy data.

### Measurement of microvascular and epithelial permeability

Pulmonary microvascular permeability was estimated by the rate of <sup>111</sup>In-transferrin accumulation in lungs relative to heart. About 400  $\mu$ Ci of <sup>111</sup>In chloride (indium strongly binds to transferrin in vivo) were injected in the dorsal penile vein. Regions of interest (ROI) were delineated on the cardiac cavities using the early images and on the zones corresponding to the instilled and contralateral lung (ROI<sub>E</sub> and ROI<sub>CL</sub>). <sup>111</sup>In-transferrin accumulation rate was deduced from the normalized lung to heart activity ratio [17]. <sup>111</sup>In-transferrin accumulation slope was calculated by linear regression.

Alveolar epithelial permeability to albumin was obtained from the rate at which <sup>99m</sup>Tc-labeled albumin leaked from the lungs. <sup>99m</sup>Tc-labeled albumin was prepared using a commercial kit (Vasculocis; Cis Bio International, Gif sur Yvette, France). Two hundred and fifty microliters of an  $\times 2$  hypertonic mannitol solution

**Table 1** Parameters (mean values  $\pm$  SD) at the beginning of the test ventilation period (after 30 min of conventional ventilation, CV)

Ventilation modality	<i>n</i>	$V_T$ (ml/kg)	RR (stroke/min)	PEEP (cmH <sub>2</sub> O)	Pei (cm H <sub>2</sub> O)	HR (bpm)	MAP (mmHg)
CV	4	8	70	2	13.8 $\pm$ 1.38	–	–
CV + T	4	8	70	2	12.0 $\pm$ 0.71	–	–
HV1	7	25.3 $\pm$ 2.93	25.7 $\pm$ 6.16	0	30	–	–
HV1 + Terbu IT	7	27.3 $\pm$ 3.16	22.0 $\pm$ 2.58	0	30	–	–
HV1P	7	25	24.1 $\pm$ 0.38	6	37.1 $\pm$ 1.67	–	–
HV1P + Terbu IT	7	25	24.5 $\pm$ 1.28	6	34.3 $\pm$ 1.69	–	–
HV2	6	21.7 $\pm$ 1.29	22.0 $\pm$ 0.63	0	25	267 $\pm$ 25.1	103 $\pm$ 7.8
HV2 + Terbu IP	6	22.0 $\pm$ 1.42	22.1 $\pm$ 0.74	0	25	345 $\pm$ 20.4**	66 $\pm$ 4.6**

HR, heart rate; HV1, ventilation at 30 cmH<sub>2</sub>O Pei and no PEEP; HV1P, ventilation at 30 cmH<sub>2</sub>O Pei and 6 cmH<sub>2</sub>O PEEP; HV2, ventilation at 25 cmH<sub>2</sub>O Pei and no PEEP; IP, intra-peritoneal; IT, intra-tracheal; MAP, mean arterial pressure; Pei, end-inspiratory pressure; RR, respiratory rate; Terbu, terbutaline; \*\*  $p < 0.01$

(120 mg/ml), supplemented with bovine serum albumin (80 mg/ml), sodium transport inhibitors and 600  $\mu$ Ci <sup>99m</sup>Tc-albumin [15], were instilled in one lung so that pulmonary microvascular permeability could be compared between the instilled and the uninstilled lung. The solution was slowly instilled in a distal airway after a short period of ventilation with FiO<sub>2</sub> = 1.  $\beta$ -2-Adrenergic stimulation was done adding 10<sup>-4</sup>M (approximately 0.02 mg/kg body weight) terbutaline to the instillate (intra-tracheally, IT), or injecting 0.5 mg/kg IP, immediately before the initial CV period. <sup>99m</sup>Tc activity in a ROI drawn over the thorax (ROI<sub>T</sub>) was integrated over 150-s steps and divided by initial activity. We have shown [15] that the initial decay slope, measured during the first 6 min of change in lung <sup>99m</sup>Tc-albumin content, was proportional to alveolar albumin permeability–surface area product (PS<sub>A</sub>).

#### Wet-to-dry lung ratios

After the rats were killed with an overdose of pentobarbital, the trachea was clamped. The thorax was opened and the main bronchi tied. The lungs were weighed and placed at  $-20^\circ\text{C}$  for 30 days to allow radioactivity to decay. They were then again weighed to verify the absence of significant weight loss and weighed dry after being left at  $80^\circ\text{C}$  for 7 days.

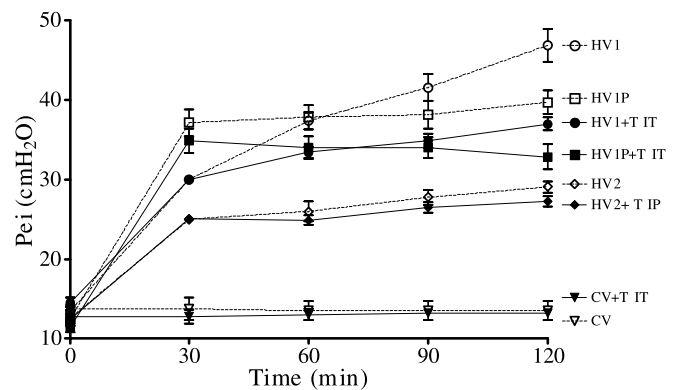
#### Statistical analysis

Results are expressed as mean  $\pm$  SEM. Comparisons between groups were made by one or two-way ANOVA and the Bonferroni post-hoc test, or by the Kruskal–Wallis test and the Dunn post-hoc test. A probability value of  $p < 0.05$  or less was considered significant (Prism; GraphPad, San Diego, USA).

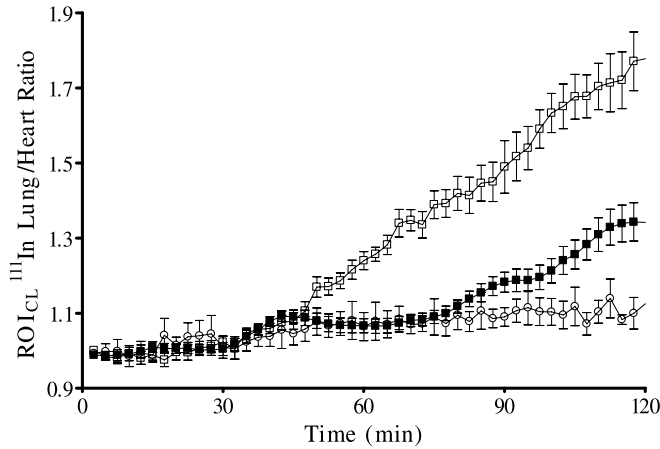
## Results

### End-inspiratory pressure during ventilation

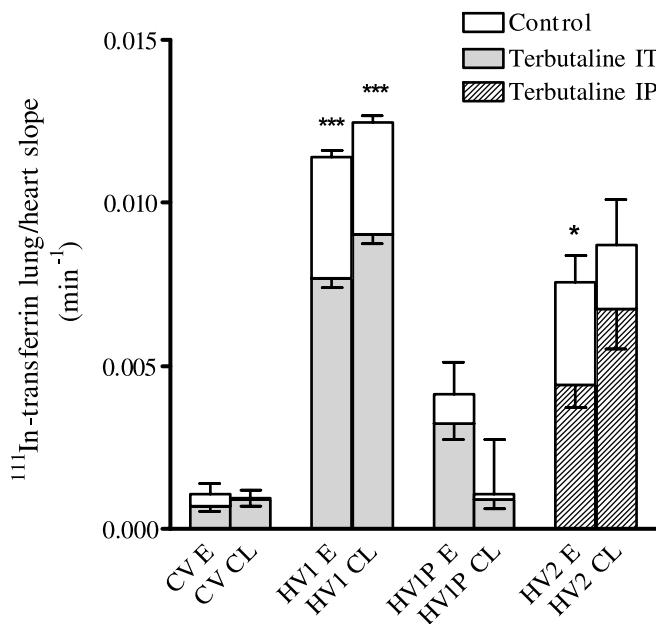
The Pei values at t30 (end of the baseline CV period) and later are shown in Fig. 1. Pei remained steady in the CV groups. It increased when 30 cmH<sub>2</sub>O Pei ventilation was initiated, but significantly less in the presence of terbutaline ( $p < 0.001$  at 90 and 120 min). Pei remained steady in the presence of PEEP, and even decreased to be significantly lower at 120 min ( $p < 0.001$ ) when terbutaline was given IT. After an initial increase, Pei remained steady during HV at 25 cmH<sub>2</sub>O Pei whether terbutaline was given IP or not.



**Fig. 1** Changes in end-inspiratory pressure (*Pei*) during ventilation with terbutaline (*T*) given in the instillate (intra-tracheally, *IT*) or intra-peritoneally (*IP*) or without terbutaline. *Pei* was similar in all groups during the 30-min conventional ventilation (*CV*) period and stayed low in animals ventilated with this modality. *Pei* increased sharply in rats ventilated with a high volume at 30 cmH<sub>2</sub>O *Pei* (*HV1*), either in the absence or the presence of PEEP (*HV2P*), but not at 25 cmH<sub>2</sub>O *Pei* and no PEEP (*HV2*). Intra-tracheal terbutaline significantly decreased *Pei* in animals ventilated at 30 cmH<sub>2</sub>O *Pei* ( $p < 0.001$  at 90 and 120 min)



**Fig. 2** Changes in  $^{111}\text{In}$ -transferrin lung/heart ratio in the region of interest drawn around the non-instilled lung during conventional ventilation (*open circles*), high-volume ventilation at 30 cmH<sub>2</sub>O end-inspiratory pressure without PEEP (*open squares*) applied at  $t=30$  min and the same ventilation modality in animals given terbutaline in the instillate (*filled squares*). Terbutaline significantly lessened the slope of lung/heart ratio increase ( $p < 0.001$ ). The same observation was made for the flooded lung, or in both lungs in the presence of PEEP



**Fig. 3** Slopes of lung/heart ratio changes with and without terbutaline according to the ventilation modality – conventional ventilation (CV) or high-volume ventilation at 30 cmH<sub>2</sub>O end-inspiratory pressure without (HV1) or with PEEP (HV1P) – in the regions of interest drawn around the instilled (E, edema) and contralateral (CL) lung (pooled values). Terbutaline given in the instillate (intra-tracheally, IT) decreased lung/heart ratio slope in both lungs during HV1 ( $***p < 0.001$  compared with the same ventilation modality in the absence of terbutaline). Terbutaline given intra-peritoneally (IP) significantly decreased lung/heart ratio slope in the flooded lung during ventilation at 25 cmH<sub>2</sub>O end-inspiratory pressure (HV2;  $*p < 0.05$  compared with the same ventilation modality in the absence of terbutaline)

### Mean arterial pressure and heart rate

Mean arterial pressure (MAP) decreased and heart rate (HR) increased almost immediately after terbutaline was given IP. MAP was lower in rats given terbutaline IP at the beginning of the HV period (Table 1) and remained lower than in similarly ventilated controls during all this period ( $p < 0.001$  by two-way ANOVA), although it had a slight tendency to increase with time. HR was significantly higher at the beginning of the CV period in rats given terbutaline IP ( $401 \pm 19.7$  vs  $311 \pm 22.0$  bpm,  $p < 0.01$ ). HR decreased after commencement of HV in both groups (Table 1), then remained steady and higher in rats given terbutaline IP than in controls during the whole HV period ( $p < 0.001$  by two-way ANOVA).

### Lung $^{111}\text{In}$ -transferrin accumulation

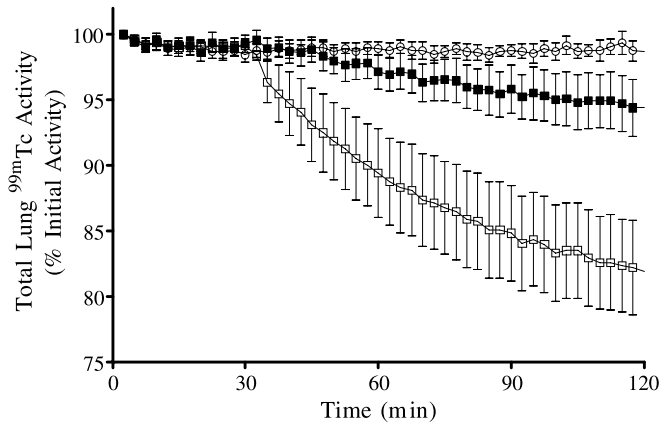
Figure 2 shows that  $^{111}\text{In}$ -transferrin accumulated slowly in the lungs of rats ventilated with CV. The increase in  $^{111}\text{In}$ -transferrin accumulation rate was almost immediate when HV was applied. Terbutaline IT significantly lessened  $^{111}\text{In}$ -transferrin accumulation rate in the instilled and the contralateral lung. The decrease observed with terbutaline IP reached significance in the instilled lung only (Fig. 3). PEEP decreased lung  $^{111}\text{In}$ -transferrin accumulation ( $p < 0.01$ ) to a value that was not significantly different from CV controls in uninstilled lungs (Fig. 3). The effect of terbutaline IT in the instilled lung did not reach significance.

### Alveolar $^{99\text{m}}\text{Tc}$ -albumin permeability

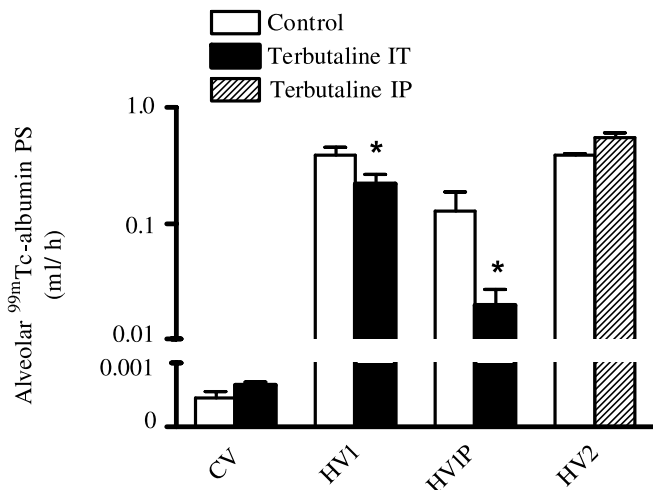
CV was not accompanied by significant  $^{99\text{m}}\text{Tc}$ -albumin leakage from airspaces. Terbutaline IT had no effect on albumin permeability during CV. HV produced an immediate decrease in lung  $^{99\text{m}}\text{Tc}$ -albumin content, as previously reported [15], which was lessened by PEEP (Fig. 4,  $p < 0.01$ ). Terbutaline IT significantly reduced the increase in albumin permeability due to HV ventilation, regardless of the presence of PEEP, as shown in Fig. 5. Albumin permeability in the presence of PEEP + terbutaline IT was higher, but not significantly so, than that in CV controls. Terbutaline IP did not affect the increase in alveolar albumin permeability due to 25 cmH<sub>2</sub>O Pei ventilation.

### Wet-to-dry lung weight ratio

Wet-to-dry lung weight ratios are shown in Fig. 6. Terbutaline IT did not affect wet-to-dry ratio in the CV group. High-volume ventilation at 30 cmH<sub>2</sub>O (HV1) or 25 cmH<sub>2</sub>O (HV2) end-inspiratory pressure (Pei) significantly increased wet-to-dry ratio ( $p < 0.001$ ).

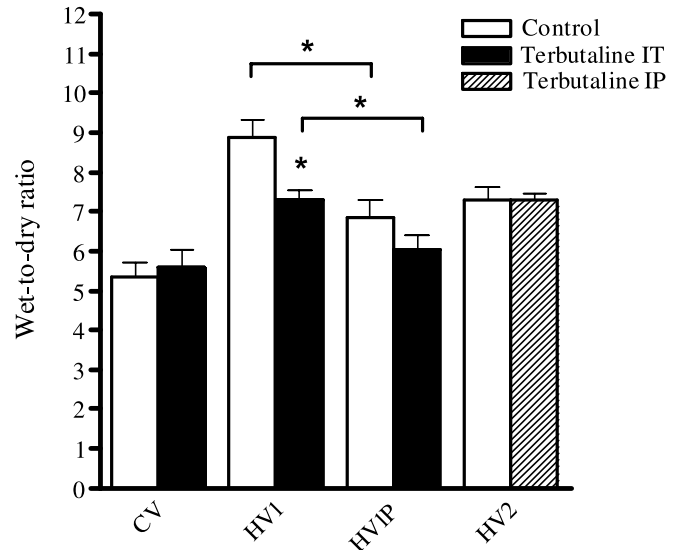


**Fig. 4** Changes in lung  $^{99m}\text{Tc}$ -albumin content (pooled values) during conventional ventilation (open circles), high-volume ventilation at 30  $\text{cmH}_2\text{O}$  without PEEP (open squares) and the same ventilation modality in animals given terbutaline in the instillate (filled squares). Terbutaline lessened the leakage of albumin from alveolar spaces produced by high-volume ventilation. The same observation was made during high-volume ventilation with PEEP. Terbutaline given intra-peritoneally did not change alveolar albumin leakage



**Fig. 5** Alveolar  $^{99m}\text{Tc}$ -albumin permeability surface area product ( $\text{PS}_A$ , log scale) calculated from the initial slopes of the curves shown in Fig. 4.  $\text{PS}_A$  was nearly 1,000 times higher ( $p < 0.001$ ) during high-volume ventilation at 30 (HV1) or 25  $\text{cmH}_2\text{O}$  (HV2) end-inspiratory pressure than during conventional (CV) ventilation, irrespective of the presence of PEEP (HV1P). Terbutaline given in the instillate (intra-tracheally, IT), but not intra-peritoneally (IP), reduced  $\text{PS}_A$  when increased by high-volume ventilation ( $*p < 0.05$  compared with the same ventilation modality)

This increase was lessened by PEEP (HV1P,  $p < 0.05$ ). Terbutaline IT significantly lessened wet-to-dry ratio in rats ventilated with 30  $\text{cmH}_2\text{O}$  Pei and ZEEP (HV1). Terbutaline IP did not affect this ratio during 25  $\text{cmH}_2\text{O}$  Pei ventilation and ZEEP.



**Fig. 6** Lung wet-to-dry ratios after conventional ventilation (CV), and high-volume ventilation at 30  $\text{cmH}_2\text{O}$  end-inspiratory pressure without (HV1) and with (HV1P) PEEP and high-volume ventilation at 25  $\text{cmH}_2\text{O}$  end-inspiratory pressure without PEEP (HV2). Wet-to-dry ratios were significantly lower when terbutaline was given in the instillate (intra-tracheally, IT), but not when it was given intra-peritoneally (IP). PEEP significantly lessened wet-to-dry ratios, independently of the presence of terbutaline IT.  $*p < 0.05$

## Discussion

This study shows that terbutaline, given either IT or IP, reduces protein fluxes across the capillary barrier during HV in vivo. Terbutaline IT, but not terbutaline IP, lessened the increase in alveolar permeability due to HV. This lessening was additive to that produced by PEEP. This is the first description, to our knowledge, of an effect of a  $\beta_2$ -adrenergic agonist on alveolar epithelial permeability alterations due to high lung stretch.

### Terbutaline and microvascular permeability alterations

Rats ventilated with CV slowly accumulated  $^{111}\text{In}$ -transferrin in their lungs, whether or not terbutaline was administered IT. HV at 25 or 30  $\text{cmH}_2\text{O}$  Pei produced a considerable increase in  $^{111}\text{In}$ -transferrin lung accumulation. We have shown that lung  $^{111}\text{In}$ -transferrin uptake increased when end-inspiratory pressure exceeded 20–25  $\text{cmH}_2\text{O}$  in rats [15, 18, 19]. This uptake was similar in aerated or flooded lungs because both experienced comparable distension at these airway pressures [15]. An increase in  $^{111}\text{In}$ -transferrin lung-to-heart ratio might also have been due to an increased lung blood volume. However, it is unlikely that HV would have produced a linear increase for at least 30 min, as shown in Fig. 2. Besides, microvascular permeability alterations during HV are well documented [2].

Ventilation with a high  $V_T$  and no PEEP dispersed variable amounts of the instilled liquid, and thus terbutaline, in the uninstilled lung, which helps to explain why  $^{111}\text{In}$ -transferrin uptake was lessened in both lungs when terbutaline was given IT. PEEP application during HV reduced microvascular permeability, which even returned to control levels in the uninstilled lung [19]. This effect of PEEP provides a further explanation for the previous observations that PEEP lessened HV-induced pulmonary edema [20, 21] and lung albumin uptake [20].

The observation that terbutaline either given IT or IP reduces in vivo the HV-induced  $^{111}\text{In}$ -transferrin accumulation in the lungs agrees with previous observations made ex vivo showing that  $\beta$ -agonists lessen microvascular permeability alterations [13, 22]. Terbutaline may also have reduced filtration pressure, because of its relaxing effect on vasculature, which resulted in a significant MAP decrease when given IP. However, terbutaline is also inotropic; the increase in cardiac output usually seen with this agent in rats (see for example [23]) may have offset the consequences of this vasodilation on mean pulmonary capillary pressure. Although a decrease in mean capillary pressure cannot be excluded as an explanation for the decrease in pulmonary  $^{111}\text{In}$ -transferrin accumulation with terbutaline, it is likely that a lessening of permeability alterations also participated in this decrease. Indeed, the lessening of microvascular permeability by  $\beta$ -adrenergic agonists has been observed in various situations [10, 13, 24–26]. Interestingly, in the above-mentioned study by Basran et al. [12], five of the six patients in whom lung  $^{113\text{m}}\text{In}$ -transferrin uptake was lower after terbutaline infusion survived, while the other four patients died.

#### Terbutaline and $^{99\text{m}}\text{Tc}$ -albumin alveolar permeability

Albumin clearance rate during CV was less than 2%/h, in keeping with previous estimates [15, 27]. This baseline clearance rate was unaffected by terbutaline. HV almost immediately increased  $\text{PS}_A$ ; PEEP application reduced this increase, as previously reported [19], despite slightly higher end-inspiratory (plateau) pressures, and thus comparable levels of overall lung distension. Terbutaline IT attenuated the increase in  $\text{PS}_A$  whatever the level of PEEP. This effect was not limited to the early leakage of albumin from airspaces to the circulation (the data used to calculate  $\text{PS}_A$ ), but lasted at least 90 min, even in the absence of PEEP. It has been shown that PEEP application may help to prevent epithelial lesions during ventilation [20] perhaps by lessening abrasion due to liquid movement in airways [28–30], but these injuries occur later because relatively short periods of HV do not damage the lungs [31, 32]. Our observations suggests that  $\beta_2$ -adrenergic agonists, when given IT, may combine with PEEP to prevent the HV-induced increase in alveolar/airway protein permeability. The route of terbutaline

administration may, however, be important, at least in rats, because terbutaline given IP did not reduce the HV-induced increase in permeability. The administered dose was rather high, was proven effective in other models [33], and may even be harmful in rats [34]. Terbutaline IP produced significant systemic effects, a sustained decrease in MAP and increase in HR, and a reduction in pulmonary  $^{111}\text{In}$ -transferrin accumulation. It is thus possible that terbutaline did not reach alveolar cell  $\beta_2$ -receptors at a sufficiently high concentration, as its effects were obvious when given IT. The issue of terbutaline dosage when administered in the systemic circulation deserves further studies.

#### Alveolar barrier permeability and lung water content

A recent randomized placebo-controlled trial showed that salbutamol infusion might decrease extravascular lung water in ALI or ARDS patients [7]. Because thermal dilution is a questionable method for measuring lung water in these patients [35], experimental studies are required to better evaluate whether  $\beta$ -adrenergic agonists really decrease lung water during permeability-type edema.

Our wet-to-dry ratio measurements suggest that  $\beta_2$ -adrenergic agonist administration may slow pulmonary edema development during HV. Microvascular permeability alterations are the main determinant of pulmonary edema during ventilator-induced lung injury [2]. Microvascular protein reflection coefficient (and thus permeability) correlates with the rate of liquid extravasation at a given filtration pressure. It is therefore not surprising that the lessening of protein permeability alteration by  $\beta_2$ -adrenergic treatment also affects wet-to-dry ratio.

It is possible that the protective effects of terbutaline observed in this study were due to hemodynamic changes. However, terbutaline is inotropic and increases cardiac output in the rat [23], so that it is difficult to infer from a decrease in MAP what occurred in the pulmonary microcirculation.

Another mechanism by which terbutaline might have decreased lung water content is stimulation of alveolar fluid clearance. Ware and Matthay have shown that alveolar liquid clearance is important for recovery in patients with ALI [8].  $\beta$ -Adrenergic agonists have long been known to stimulate alveolar liquid clearance in the rat [36]. It is possible that the increase in permeability due to HV had facilitated leakage of transport inhibitors (which are small molecules) from the airspaces and increased sodium entry, and thus that alveolar liquid clearance was not completely inhibited when permeability was high. Few studies have considered the relationship between alveolar epithelium permeability and alveolar liquid clearance. Injuries that markedly increase alveolar permeability to proteins, such as severe *Pseudomonas* pneumonia, also decrease clearance [37]. However, it is likely that epithelial lesions are

inhomogeneously distributed, so that alveolar clearance may be preserved in places. The observed decrease in wet-to-dry ratio observed with terbutaline IT may thus be related to the slowing of pulmonary edema development because of its protective effect on capillary barrier permeability and of liquid clearance stimulation. Terbutaline IP, which did not prevent alveolar permeability alterations, failed to lessen wet-to-dry ratio, which suggests that a preserved epithelium is essential to decrease the rate of edema formation. Whatever the mechanism, terbutaline IT offered a protection that was additive to that provided by PEEP.

In conclusion, this study shows that terbutaline IT reduces edema formation, possibly by lessening microvascular permeability alterations, during an injurious, high-volume, ventilation modality, and decreases the rate of edema formation in vivo. Terbutaline given IP seems

less efficient than given IT in the rat. We were surprised to find no effect on alveolar permeability or lung water. We have no clear explanation for this observation. More work is needed to clarify the effect of terbutaline and of its route of administration on pulmonary transvascular protein fluxes in vivo during high lung stretch. However, beta-adrenergic agonists have been studied for several decades for their possible benefit during permeability type pulmonary edema/acute lung injury, but this issue has not so far received a definitive answer despite many studies [38]. Our study is the first to deal with the effect of a beta-adrenergic agonist on alveolar permeability changes, an effect that cannot be explained by hemodynamic alterations. The effects of terbutaline IT are additive to those of PEEP, suggesting that combination of both treatments may be beneficial to patients with acute lung injury and ARDS.

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