Experimental model to evaluate the effect of hydrothorax and lobar resection on lung compliance[†]

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INTRODUCTION

The model of lung resection has been extensively studied in experimental animals to determine the potential limitations of the lung's diffusive properties as well as its regeneration capability. Results have been promising to demonstrate the remarkable capacity of the lung to recover from the loss of tissue mass [1–4].

Conversely, little relevance has been given to the problem of the disruption of the mechanical coupling of the lung to the chest wall following lung resection. In fact, deciding for the best strategy for chest drainage after lobectomy still represents a critical issue for the surgeon. In practice, although pleural suction has been used for years based on the assumption that it may promote healing as well as a compensatory increase in lung volume, a recent review [5] does not support evidences for an objective advantage in face of three main postoperative complications, namely air leak, hydrothorax and 'idiopathic lung oedema'. A recent article based on respiratory and pleural space mechanics considered the problem of the mechanical coupling of the lung, whose volume has been reduced by surgery, to the chest wall. The challenging hypothesis was put forward that lung over-distension resulting from a forced chest drainage would favour a derangement of lung interstitial matrix structure, a critical cofactor for the

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complications mentioned above [6]. The objective of this investigation was to develop an experimental animal model to assess to what extent a decrease in lung volume, caused by lobar resection or hydrothorax (a common post-surgery complication), would impact on potential lung over-distension.

METHODS

Experiments were performed on adult (New Zealand White) male rabbits (weight range 2÷2.2 kg) anaesthetized with a bolus of 2.5 ml/kg of a saline solution containing 0.25 g/ml of urethane plus 10 mg/ml of pentobarbital sodium injected into a ear vein. Tracheostomy was performed, and a 50 mm-long plastic cannula (inner diameter 2.5 mm) was inserted into the distal trachea. We preferred tracheostomy rather than endotracheal intubation in order to preserve flow-resistive properties of the trachea to allow the estimate of lung compliance that is required to minimize the flow-resistive component (see also below).

Before connecting the animal to the ventilator, paralysis was accomplished by pancuronium bromide (1 mg/kg body weight initial dose, supplemented by 0.33 mg/kg every 40 min). Mechanical ventilation was provided with a tidal volume of ~20 ml and a frequency of 16 breaths/min. The inspiratory flow signal was measured by a Fleisch-type pneumotachograph connected to a differential pressure transducer (Sensym, Milpitas, CA, USA). Flow was integrated in order to obtain lung volume and volume drift was avoided by correcting flow offset. Tracheal and oesophageal (through oesophageal balloon) pressures were measured by disposable pressure transducers (Edwards Baxter, Irvine, CA, USA). All signals were digitized by an analogue-to-digital board (NI BNC-2090, National Instruments, Austin, TX, USA) and dedicated software for real-time signal acquisition and visualization was implemented in LabView (version 8.2, National Instruments) and for the signal processing in Matlab (The Mathworks, Inc., Natick, MA, USA).

Lung and chest wall compliance were estimated by volume and pressure data obtained during low flow inflation to minimize resistive pressure components leaving only elastic forces. Lung volume was increased by ~20 ml starting from end expiration.

Experiments were performed randomizing the rabbits into two groups: (i) experimental hydrothorax of various degrees and (ii) right lung lobectomy. Each animal was studied first in the control condition and then after either hydrothorax or lobectomy.

A general consensus for the experimental procedures used in the research activity was obtained from the Local Ethical Committee.

Protocol of hydrothorax

Experiments were done in five rabbits. With the animal in the supine position, an access to the right side of the chest was opened corresponding to the sixth inter-costal space by resecting the superficial tissue and chest muscles. A sealed-tipped saline-filled thin cannula (external diameter 1 mm, internal diameter 0.6 mm) with side holes was inserted tangentially into the pleural cavity for ~2 cm and left in site. The recording site of the cannula was at ~50% of the height of the pleural cavity. Lung and chest wall compliance were estimated in control conditions and after sequential injections of 2 ml of heated (37°C) saline solution through the intrapleural cannula, up to a total volume of 8 ml.

Protocol of lobectomy

Experiments were done in six rabbits. In four rabbits, the lower lobe was resected, while in two the middle and lower lobe were resected. An access to the right-hand side of the chest was cleared at ~50% of the height of the lung, along the fourth to fifth intercostal space, allowing to open the chest at the level of the hilar broncho-vascular branching of the right lung. The use of a custom curved device allowed one to pass a thread to ligate the broncho-vascular peduncle of the lower lobe or of the lower plus the middle lobes. Subsequently, the lung tissue to be removed was hooked, resected and weighted. The tidal volume of the ventilator was decreased after resection so as to match the preoperative end-inspiratory alveolar pressure.

Respiratory mechanics data analysis

Compliance is the mechanical index commonly used to describe elasticity in the respiratory system; for the lung it is defined as $C_L = \Delta V/\Delta P_L$, where $\Delta V$is the change in lung volume of its alveolar units and $\Delta P_L$is the change in distending pressure that generates ΔV [7].

The respiratory system includes the chest wall and the lung that are mechanically placed in series, so that the following relationship holds between total compliance of the respiratory system (C_{RS}) and the compliances of the chest wall (C_{CW}) and of the lung (C_L):

$$\frac{1}{C_{RS}} = \frac{1}{C_{CW}} + \frac{1}{C_L}. \tag{1}$$

Considering that the right and left lung are mechanically placed in parallel, the following relationship holds between C_L and the compliance of the right (C_{RL}) and left (C_{LL}) lung:

$$C_L = C_{RL} + C_{LL}. \tag{2}$$

Furthermore, assuming that the right and left lung have the same volume as well as the same mechanical characteristics, one can write

$$C_{RL} = C_{LL} = \tfrac{1}{2}C_L. \tag{3}$$

We measured lung compliance under positive pressure ventilation, which implies a different range of alveolar and pleural pressures relative to spontaneous breathing. However, transpulmonary pressure would be exactly the same when measured in both conditions.

Estimate of the change in compliance due to hydrothorax and lobectomy

The changes of C_{CW}, C_L and C_{RL} were estimated in control conditions and following hydrothorax and lobectomy; C_{CW} was obtained from the ratio between lung volume change and the corresponding change in transthoracic pressure, given by oesophageal pressure; C_L was derived as the ratio of lung volume to transpulmonary pressure changes, obtained as the difference between alveolar and oesophageal pressure; C_{RL} following hydrothorax and lobe resection was obtained as

$$C_{RL'} = C_{L'} - C_{LL}, \tag{4}$$

where the superscript ′ indicates the specific condition of either hydrothorax or lobectomy and the compliance of the left lung (C_{LL}) is assumed to remain unchanged.

Estimate of the decrease in lung volume caused by hydrothorax

When hydrothorax accumulates in the pleural cavity, the chest wall expands and the lung retracts and these volume changes ($\Delta V'_{CW}$ and $\Delta V'_L$, respectively) can be obtained as the product of the relative compliance of the structures by the corresponding changes in recoil pressure (ΔP). Accordingly, the relative volume changes are given by

$$\Delta V'_{CW} = C_{CW} \cdot \Delta P', \tag{5}$$

$$\Delta V'_L = C_L \cdot \Delta P'. \tag{6}$$

For the right lung exposed to hydrothorax

$$\Delta V'_{RL} = C_{RL} \cdot \Delta P'. \tag{7}$$

Estimate of the decrease in lung volume caused by lobectomy

Total lung volume at functional residual capacity (FRC) has been estimated at 20 ml [8], equally shared between the right and left lung. The percentage of lobectomy was estimated from the ratio of the weight of the resected portion (V_{res}) to the weight of the remaining lung (V_{rem}) as

$$\%\text{Volume resected} = \frac{V_{res}}{V_{res} + V_{rem}} \cdot 100. \tag{8}$$

Statistical analysis

Statistical analysis was performed by SigmaStat software v11.0 (San Jose, CA, USA).

The effects of 8 ml of hydrothorax and lobectomy on lung compliance were tested by using a paired *t*-test. All data are reported as mean values ± standard error. *P*-values lower than 0.05 were considered to indicate significant differences.

RESULTS

Figure 1 shows an example of the recording of oesophageal pressure, alveolar pressure, lung volume and pleural liquid pressure in control conditions and how these variables change, relative to end expiration, during a slow inflation manoeuvre. Note that, at FRC, oesophageal pressure was about −1 cmH_2O, while pleural liquid pressure, at ~50% of lung height, was −4 cmH_2O. Under control conditions C_L averaged 3.3 ± 0.8 (SD) ml/cmH_2O (thus, from Equation (3), $C_{RL} = C_{LL} = 1/2 \cdot C_L = 1.65$ ml/cmH_2O), and C_{CW} was 12.8 ± 4.7 ml/cmH_2O.

Hydrothorax

Hydrothorax increased pleural liquid pressure at mid lung height at a volume corresponding to FRC by about 0.4 cmH_2O. Based on a hydraulic gradient of 1 cmH_2O/cm height, we estimated that 8 ml hydrothorax led to an increase in pleural liquid pressure at the bottom of the cavity, from a control value of −1 cmH_2O up to ~2.5–3 cmH_2O. Figure 2A shows that 8 ml of hydrothorax left C_{CW} essentially unchanged while C_L decreased significantly ($P < 0.001$) due to the exposure of the right lung to positive pleural liquid pressure. With 8 ml of hydrothorax, oesophageal pressure became less sub-atmospheric by ~0.5 cmH_2O, reflecting a decrease in lung and chest wall opposite recoil pressures due to hosting of the hydrothorax volume within the pleural space. Based on Equation (5), $\Delta V'_{CW}$ was equal to 12.8·0.5 = 6.4 ml and $\Delta V'_L$ decreased by ~2 ml.

Lobectomy

In four animals, the weight of the resected portion of the right lung was 2.01 ± 0.79 g, corresponding to ~30% decrease in right lung weight. In two more animals lobectomy of middle plus lower lobes amounted to 3.24 ± 0.13 g, corresponding to ~60% decrease in right lung weight. Figure 2B shows that C_L decreased to 1.75 ± 0.3 and to 1.52 ± 0.4 ml/cmH_2O for a 30 and 60% lobectomy, respectively.

The expected values of lung compliance after hydrothorax and lobectomy

Figure 3A presents the concept that the overall change in lung volume to be considered for the estimate of compliance reflects the sum of the unitary changes in volume of the alveoli included in the lung mass. Figure 3B shows how lobectomy and hydrothorax cause a decrease in lung compliance due to the loss of alveolar units contributing to the overall change in lung volume. Lobectomy can be immediately recognized as a cause of decreased lung compliance due to the loss of alveolar units caused by volume reduction. Hydrothorax may also represent a potential cause of a decrease in alveolar units due to the increase of the pressure of the pleural fluid within the cavity relative to the control as shown in Fig. 3C and D. In particular, the increase in pleural liquid above atmospheric pressure in the lowermost parts of the cavity may actually compress the lung, thus causing alveolar atelectasis.

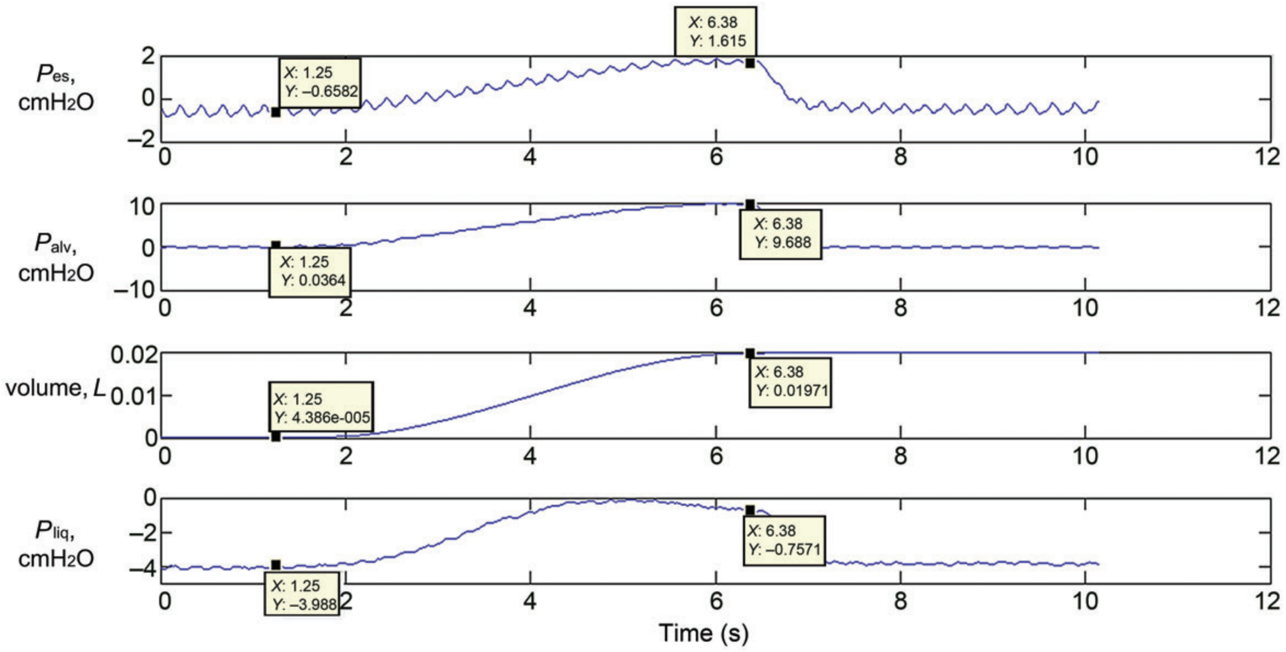

Figure 1: An example of experimental recordings of oesophageal pressure, alveolar pressure, lung volume and pleural liquid pressure (from top to bottom) in control conditions.

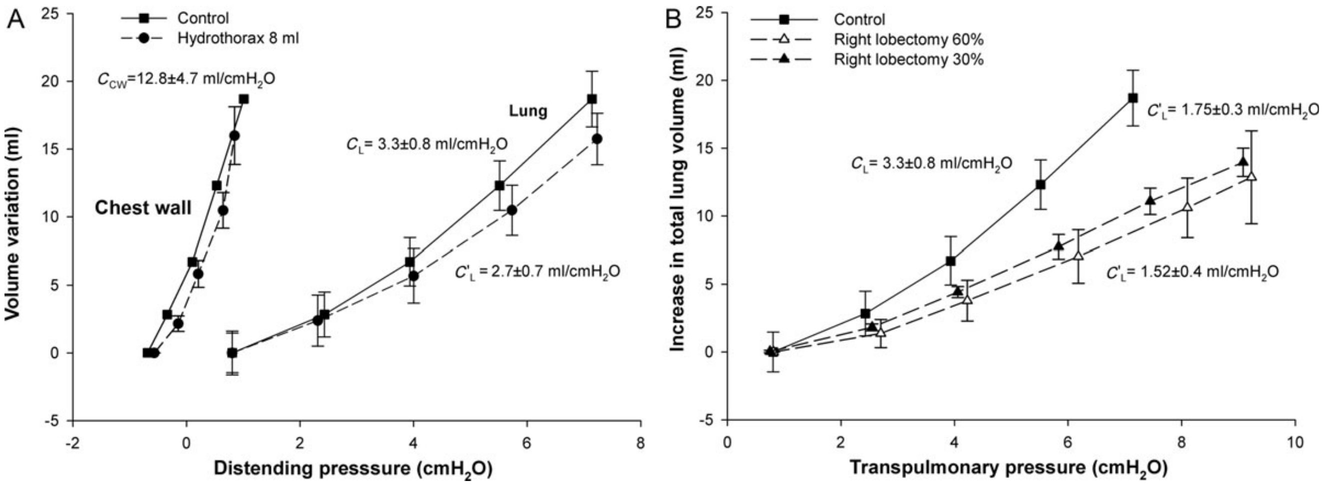

Figure 2: (**A**) Pressure–volume curves of chest wall and lung in control (square symbols) and after 8 ml of hydrothorax (circles). (**B**) Pressure–volume curves of the lung in control (closed squares) and after 30% (closed triangles) and 60% (open triangles) lobectomy. Data are shown as mean ± SD.

DISCUSSION

The data from the present study allowed us to quantitate the decrease in lung compliance due to lobectomy and hydrothorax.

The unilateral impact of hydrothorax and lobectomy

Although original data were obtained on the whole lung, it might be appropriate to refer in particular to the lung directly exposed to hydrothorax and/or subjected to lobectomy. As from equation (3), we assumed [8] for the right lung an FRC equal to 10 ml (50% of the whole lung) and a compliance equal to 50% of total lung compliance. Next, we considered that the decrease in lung volume, estimated for hydrothorax and lobectomy, was only at the expense of the right lung. Thus, for 8 ml of hydrothorax the decrease in right lung volume was ∼2 ml. It is interesting to note that with hydrothorax the increase in chest wall volume was three times greater than the decrease in lung volume, reflecting a similar difference in compliance. The decrease in right lung compliance, ∼45%, was much greater than that expected based on the estimated decrease in right lung volume (i.e. ∼20%) (Fig. 4A). As shown in Fig. 3D, this greater than expected decrease in compliance may be due to the fact that the lung, whose density is ∼0.3 g/ml, with hydrothorax becomes a buoyant, though highly compressible, organ floating

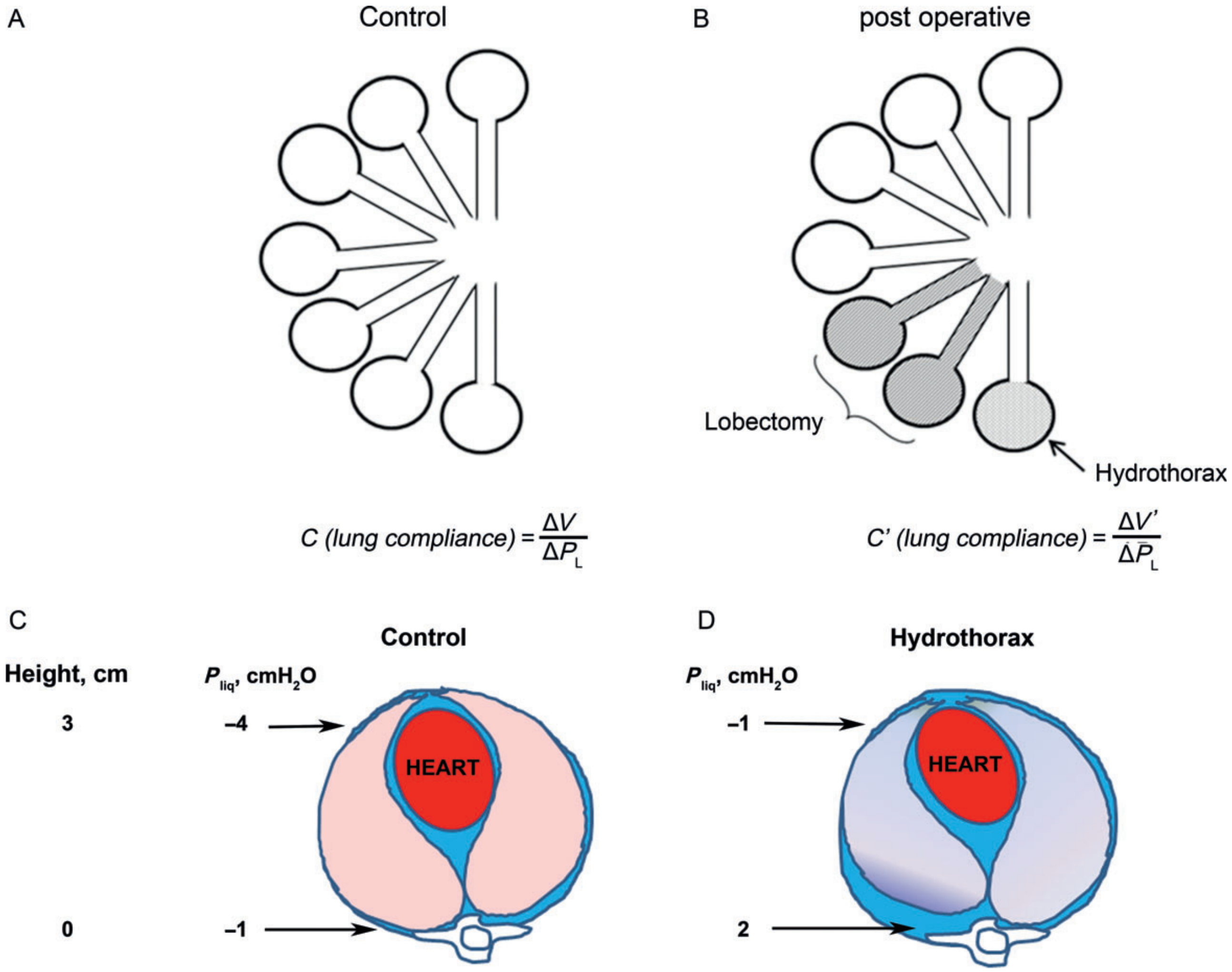

Figure 3: (**A**) Schematic drawing showing one lung in the control condition whose compliance is defined as the ratio of the change in volume of all its alveolar units divided by the lung-distending pressure. (**B**) Two potential causes of decrease in alveolar units, namely lobectomy and hydrothorax, that are expected to reduce lung compliance. (**C**) Schematic drawing to show the distribution of pleural liquid pressure as a function of height in control conditions. (**D**) Schematic drawing to show the distribution of pleural liquid pressure after hydrothorax, suggesting compression of the most dependent part of the lung due to exposure of positive pleural liquid pressure.

in fluid at positive, rather than sub-atmospheric, pressure. Figure 4A also shows that, after unilateral hydrothorax, a 4-fold increase in lobar distending pressure (from ∼1 to 4 cmH_2O) would be needed to raise the lung volume to the control value (from 8 to 10 ml). In humans, the chest wall is more rigid and its compliance is essentially equal to that of the lungs; accordingly, one can predict that lungs and chest wall would equally share a change in volume in the presence of pleural effusion. Similar to animals, however, the decrease in volume may be largely at the expense of the lung directly exposed to unilateral hydrothorax. An 8 ml unilateral hydrothorax in rabbits could be approximately comparable with a 300 ml unilateral hydrothorax in humans (10% of total lung FRC of 3L, or 20% of one-lung volume only).

Figure 4B shows the analysis for the right lobectomy. On an average, the decrease in compliance for estimated decreases in volume of 30 and 60% was greater than expected, amounting to 62 and 80%, respectively. This difference may reflect inaccuracy in the estimate of the extension of lobectomy and/or surgical damage. Figure 4B also shows that a lobar distending pressure of ∼6 cmH_2O would be needed to expand the right lung to compensate for a 30% lung resection and a pressure much >10 cmH_2O for a 60% lung resection. Even larger pressures would obviously be required to generate tidal volume. In clinical practice, hydrothorax is commonly observed as a consequence of lobectomy. Thus, based on the results of the present study, potential combined detrimental effects of hydrothorax and lobar resection on lung compliance could justify the occurrence of lung atelectasis.

The impact of over-distension on the operated lung

Evacuation of air/liquid from the cavity to allow re-expansion of the lungs is the most immediate problem after lung resection surgery. This is usually accomplished in clinical practice by a suction device. Owing to a reduced compliance of the lung on the operated side, a greater distending pressure (relative to the physiological condition) ought to be generated to bring the lung

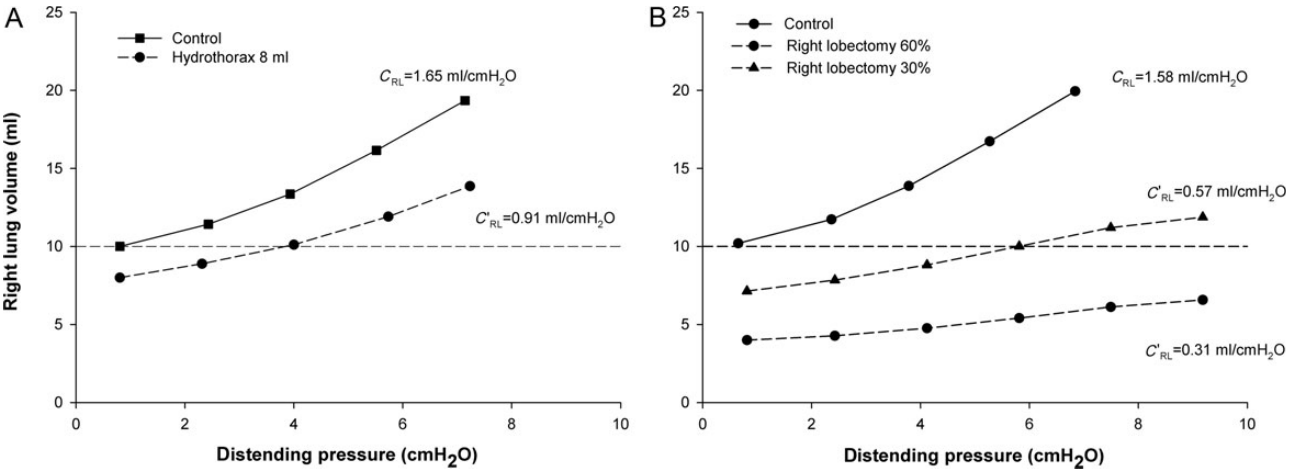

Figure 4: (**A**) Pressure–volume curve of the right lung in control condition (squares) and after 8 ml of hydrothorax (circles) causing a decrease of 2 ml of its volume at FRC. (**B**) Pressure–volume curve of the right lung in control condition (circles, solid line) and after 30% (triangles) and 60% (circles, dashed line) lobectomy.

against the chest wall; thus a rather sub-atmospheric pressure could be applied by the suction device. The mechanical consequence is over-distension of the lung parenchyma, a condition implying the risk of permanent derangement/fragmentation of the extracellular matrix of the lung [9]. Indeed, the loss of integrity of the macromolecular organization of the inter-stitial space is an important cofactor to contribute to increased leakiness of biological membranes [10, 11]. For the endothelial membrane, this may lead to oedema formation, while for the visceral pleura this favours air leak [6, 12]. Thus, the problem of over-distension ought to be considered when acutely facing the problem of air and liquid drainage from the cavity after thoracic surgery. One should also consider that inevitably over-distension of the resected lung will likely occur as time progresses until reaching the ‘final residual pleural space’. So far, it is not taken into account how a final postoperative residual pleural space is being reached, except noting that, from the standpoint of pleural mechanics, the absorption pressure of the pleural lymphatics as well as the healing process will determine the volume of the final residual pleural space. Also not acknowledged is the contribution of other structures to fill up the chest, besides the resected lung, in order to replace the surgically removed tissue. In particular, one may consider the displacement of the mediastinum and of the contralateral lung as well as the rising of the diaphragmatic dome. These compensatory mechanisms would actually prevent an excessive over-distension of the resected lung.

Limitation of the study

Although this experimental study has been done on a relatively small number of rabbits, results are clear-cut from the standpoint of respiratory mechanics, which was the main aim of our study. A further limitation is represented by the invasiveness of the operation considering the relatively small size of the animal. On the other hand, our choice for the experimental model of the rabbit was motivated by the fact that we could rely on a database we had gathered in the past on lung and pleural space mechanics [6, 9–11].

In conclusion, we found that both hydrothorax and lung resection determine a marked reduction in lung compliance, which may expose the lung to the risk of over-distension when a chest drain is applied. These findings warrant future studies investigating different strategies of drainage of air and fluid from the chest based on specific indications concerning lung-distending pressure.

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Conflict of interest: Giuseppe Miserocchi and Alessandro Brunelli have a consultant agreement with Medela Health Care, Switzerland.

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APPENDIX. CONFERENCE DISCUSSION

Dr B. Naidu *(Birmingham, UK)*: A very interesting presentation. I assume with these rodents, when you are performing a lobectomy, you had to have one-lung ventilation while you are performing the surgery. How did that affect the compliance?

Dr Miserocchi: You are saying, since we stopped ventilation on the operated side you want to know what happened after? We presented the results in terms of lung compliance that decreased more than expected based on loss of lung mass. We are interested more in describing what happened to the operated lung: we are conducting a parallel molecular biological study on the structure of the lung. I do not have that data ready now.

Dr Naidu: One of the questions is, your measures of lung compliance are global measures and there may be changes in compliance which are different on both sides.

Dr Miserocchi: I showed you the data referring to the operated side.

Dr Naidu: It is only the operated side?

Dr Miserocchi: Yes.

Dr Naidu: Is there any change on the non-operated side?

Dr Miserocchi: On the normal side?

Dr Naidu: On the normal.

Dr Miserocchi: It did not change. On average, if you consider together the operated and the normal control side, changes are not as huge as they are on the operated side.

Dr Naidu: And I may have missed this, but were these spontaneously ventilated animals or were they mechanically ventilated?

Dr Miserocchi: They were mechanically ventilated.

Dr Naidu: Would there be a difference if you were using spontaneous ventilation? Is there a difference in compliance?

Dr Miserocchi: That experiment cannot be done because the animals are paralyzed and then I have to restore the integrity on the rib cage after the operation, and we should drain out the air or the liquid, but this is impossible so far because we have been unable to have a seal of the surgical incision.

Dr D. Sugarbaker *(Boston, MA, USA)*: Your presentation is very interesting. I just want your help here a little bit. Early on following extrapleural pneumonectomy, we found that there was a significant filling of the chest on the operated side, which would cause a compression of the contralateral side. So we began leaving a catheter behind and extracting fluid on occasion.

Dr Miserocchi: On the operated side?

Dr Sugarbaker: On the operated side. And what we found was that we had a higher incidence of what I guess you are saying is overdistension of the contralateral lung, which led to your capillary leak syndrome.

Dr Miserocchi: So you had a complication on the contralateral control side?

Dr Sugarbaker: Right, which would cause hypoxia and the need for vigorous diuresis. What we have done since that time is to measure the ipsilateral pressure in the chest with a continuous monitor and to pull off small amounts of fluid rather than larger amounts intermittently. And if I understand your work, what we would be doing then would be preventing that hyperinflation, which would reduce the capillary leak within the contralateral lung. Is that what your work would suggest from the ipsilateral side?

Dr Miserocchi: Yes, but you have added one more piece of information on the contralateral side; I was analyzing only the operated side.

Dr Sugarbaker: Right.

Dr Miserocchi: Of course if you want to drain fluid from the operated side to bring the lung against the chest after lung mass reduction, by definition you cause overdistension. You are telling me that if you suck strongly from the operated side, you actually move the mediastinum towards the operated side and you also cause overdistension of the contralateral control side.

Dr Sugarbaker: That's right.

Dr Miserocchi: Both things may happen.

Dr Sugarbaker: So the question would be, would you expect this pressure transduction to occur across a very pliant mediastinum, so essentially what you were doing in the ipsilateral chest overdistending the remaining lobe.

Dr Miserocchi: Yes, one has to decide which pressure to generate in the operated chest after lobectomy, and I have no answer so far, and this pressure should avoid overdistension both on the operated side and, as you tell me, also on the contralateral side.