Surfactant replacement partially restores the activity of pulmonary stretch receptors in surfactant-depleted cats

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1Department of Women’s and Children’s Health and Department of Physiology and Medical Biophysics, Uppsala University, Uppsala, Sweden; and 2Division of Neonatology, Department of Obstetrics and Gynecology, Klinikum Grosshadern, Ludwig Maximilian University, Munich, Germany

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Sindelar, Richard, Anders Jonzon, Andreas Schulze, and Gunnar Sedin. Surfactant replacement partially restores the activity of pulmonary stretch receptors in surfactant-depleted cats. J Appl Physiol 100: 594–601, 2006. First published October 6, 2005; doi:10.1152/japplphysiol.00389.2005.—Single units of slowly adapting pulmonary stretch receptors (PSRs) were investigated in anesthetized cats during spontaneous breathing on continuous positive airway pressure (2–5 cmH2O), before and after lung lavage and then after instillation of surfactant to determine the PSR response to surfactant replacement. PSRs were classified as high threshold (HT) and low threshold (LT), and their instantaneous impulse frequency (fimp) after lung lavage (55 and 45%, respectively) in the presence of increased Ptp and decreased VT. While Ptp decreased markedly and VT remained unchanged after surfactant instillation, all except one PSR responded with increased total number of impulses and maximal fimp (42 and 26%, respectively). Some HT PSRs ceased to discharge after lung lavage but recovered after surfactant instillation. The end-expiratory activity of LT PSRs increased or was regained after surfactant instillation. After instillation of surfactant, respiratory rate increased further with a shorter inspiratory time, resulting in a lower inspiratory-to-expiratory time ratio. Arterial pH decreased (7.31 ± 0.04 vs. 7.22 ± 0.06) and Pco2 increased (5.5 ± 0.7 vs. 7.2 ± 1.3 kPa) after lung lavage, but they were the same after as before instillation of surfactant (pH = 7.21 ± 0.08 and Pco2 = 7.6 ± 1.4 kPa) during spontaneous breathing. In conclusion, surfactant instillation increased lung compliance, which, in turn, increased the activity of both HT and LT PSRs. A further increase in respiratory rate due to a shorter inspiratory time after surfactant instillation suggests that the partially restored PSR activity after surfactant instillation affected the breathing pattern.

slowly adapting pulmonary stretch receptor; lung mechanics; breathing pattern; surfactant

THE IMPORTANCE OF MAINTAINED vagal input from pulmonary stretch receptors to establish regular breathing and normal pulmonary gas exchange after birth has been investigated in several studies in which both antenatal and early postnatal vagotomy caused deterioration in lung mechanics, impairment of surfactant function, and a lower rate of breathing (6, 18, 26, 45).

The pulmonary stretch receptors consist mainly of slowly adapting pulmonary stretch receptors (PSRs), which are further divided into low-threshold (LT) and high-threshold (HT) PSRs (35). PSRs have been shown to modulate the depth and duration of each breath in response to not only changes in lung volume or transpulmonary pressure (1, 24, 38, 43) but also the rate of change in lung inflation (10, 33). In this context, PSRs have been considered to play an important role in eliciting the Hering-Breuer inspiratory inhibitory reflex (19), the strength of which has been found to be increased by immaturity and respiratory distress syndrome (RDS) in newborn infants, resulting in a prolonged expiration (12, 23). The PSRs might, therefore, respond differently to changes in lung mechanics, as in RDS (3, 8) and following instillation of surfactant (13, 16, 20, 28), and, consequently, influence the breathing pattern. Hitherto, no known study has addressed the specific effect of surfactant instillation on PSR activity in spontaneously breathing animals.

The aim of this study was to investigate whether LT and HT PSRs respond with an increased activity to surfactant instillation in surfactant-depleted lungs of spontaneously breathing cats and to determine whether this response is related to changes in lung mechanics.

MATERIALS AND METHODS

General. Fourteen young cats with a mean body weight of 3.07 ± 0.47 kg were initially anesthetized with chloroform while endotracheal intubation and dissection of the femoral artery and vein were performed. An endotracheal tube (ETT) with an inner diameter of 4 mm was inserted orally into the trachea and connected to an infant ventilator (Stephanie Infant Ventilator, F. Stephan Biomedical, Gackenback, Germany), which was set on controlled mechanical ventilation during the surgical procedures. The right femoral vein and artery were dissected, and catheters were inserted so that their tips were located in the thorax. The venous catheter was used for maintenance of anesthesia with 0.72% enflurane (C-0128, Sigma-Aldrich), with an initial dose of 10 ml/kg and additional doses of 2.5–5.0 ml·kg−1·h−1 at regular intervals. A mixture containing two-thirds of 10% glucose and one-third of 5% bicarbonate was given through the same catheter at a rate of 6.4 ml·kg−1·h−1 throughout the experiment. Blood samples were obtained from the arterial line for intermittent blood-gas measurements with an automatic acid-base analyzer (Acid-Base Laboratory ABL 300, Radiometer, Copenhagen, Denmark). Additional doses of 5% bicarbonate were given, if necessary. Care was taken to maintain a normal body temperature with the use of heating pads.

A midline incision was made in the pretracheal region, and a ligature was tied around the trachea to prevent leakage around the tube. An 8-French catheter with an esophageal balloon (40 × 7.5 mm; flat frequency response up to 5 Hz) was advanced into the lower part of the esophagus until the largest pressure swing occurred, and a ligature was tied softly around the esophagus to prevent air entrance (4).

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The cervical part of the vagal nerve was exposed, and the connective sheath was removed under a microscope and further dissected into filaments, leaving the rest of the nerve intact. Nerve strands from the filaments were placed on a single platinum electrode, and their activity was analyzed until the signal from a single PSR was found. A reference electrode was placed in the nearby connective tissue. The receptor was identified by its characteristic pattern of discharge during the ventilatory cycle and its slowly adapting activity during maintained inflation, fulfilling the adaptation index as described by Widdicombe (43) and Davis et al. (10). The nerves were immersed in mineral oil to prevent drying and for electrical insulation.

**Measurements and recordings.** Arterial blood pressure and heart rate were measured continuously with a transducer (Druck Transducer, Leicestershire, UK) placed at the level of the tip of the arterial catheter.

Airway pressure (Paw) and airflow were measured by a pneumotachograph with the same dynamic properties as an original Fleisch 00 pneumotachograph but with less dead space and resistance (39). Its sensor was interposed between the ETT and the ventilator’s connector piece.

Esophageal pressure (Pes) was recorded with a pressure transducer (Druck Transducer), which was referenced to atmospheric pressure before the recording. The validity of the esophageal balloon pressure was checked by an airway occlusion technique before each recording (4).

The nerve signals from PSRs were amplified, filtered, and rectified with a Neurolog system (Digitimer Research Instrumentation, Welwyn Garden City, Hertfordshire, UK; preamplifier NL 103, AC-amplifier NL 105, filters NL 115) and passed through an analog window discriminator to eliminate background noise and to ensure that only spike amplitudes of a certain size were recorded (21, 32). These spikes were fed to a spike generator to produce spikes of a uniform duration (0.5 ms) and amplitude (Digitimer D130 and Spike Trigger NL 200, Digitimer Research Instrumentation) to ensure that the analog-to-digital converter picked up every single spike.

Signals from Paw, Pes, airflow, and the nerve activities were digitized and recorded online by a data-acquisition system (Windaq Data Acquisition, Datalog Instruments, Austin, TX) at a sampling rate of 5 kHz. The Windaq Analysis Software (Datalog Instruments) was used to review, analyze, and process the acquired signals.

**Protocol.** The cats were kept normoventilated with mechanical ventilation before each period of spontaneous breathing on continuous positive airway pressure (CPAP), as verified by intermittent arterial blood-gas measurements.

After 2–3 min of spontaneous breathing on CPAP (2–5 cmH2O), the first recording of 10–15 breaths was made. In some healthy cats, CPAP of 5 cmH2O induced hypocapnea and a tendency to apnea. To maintain spontaneous breathing in these hypocapneic cats, the adjustments in FIO2 were made, allowing for adjustments in FIO2 (see Table 2). Each recording with CPAP was followed by arterial blood-gas measurements (see Table 2).

To estimate the degree of lung injury inflicted by lung lavage, the respiratory system compliance was measured during mechanical ventilation before lavage and 30 min after its completion. The change in respiratory system compliance caused by surfactant replacement was estimated by measuring the compliance immediately before and 10 min after surfactant instillation. The experiments were performed at the Biomedical Center of Uppsala University, and the protocol was approved by the Uppsala University Animal Research Ethics Board (D.no. C130/98; C224/0).

**Analysis of results.** Ten breaths were evaluated in each cat before and after lung lavage and after surfactant replacement.

Tidal volume (Vt) was integrated from the airflow recording in the laboratory computer (Windaq Analysis Software, Datalog Instruments), with correction for airflow leakage in the respiratory system. Transpulmonary pressure (Ptp) was calculated as the difference between Paw and Pes. The lung compliance (Cl) was calculated from Vt and Ptp.

Inspiratory time and breathing rate were calculated from the airflow recordings. Zero airflow turning to positive airflow was considered as the start of inspiration. When airflow returned to zero, this indicated the end of inspiration and coincided with maximal Vt. Inspiratory time was defined as the interval between these two points. The interval between two consecutive inspiratory starts defined one breath and permitted calculation of the breathing rate.

To detect an irregular breathing pattern, the coefficient of variation (CV) was derived from the breath-to-breath variability, i.e., the root mean square value of differences between successive intervals of breaths [RMSSD = \( \sqrt{\frac{\sum(x_i - x_{i+1})^2}{N - 1}} \)], where \( N \) is the total number of intervals with \( x_i \) being the \( i \)th interval in the sequence] (30), divided by their mean value (\( \bar{x} \)) (41):

\[
CV = \frac{RMSSD}{\bar{x}} \times 100
\]

Normal variations in the breathing pattern could thus be excluded when calculating the CV for respiratory rate, inspiratory and expiratory time, and Vt.

### Table 1. Lung mechanics and breathing pattern during spontaneous breathing on CPAP, before and after lung lavage, and after instillation of surfactant

<table>
<thead>
<tr>
<th></th>
<th>Before Lung Lavage</th>
<th>After Lung Lavage</th>
<th>After Surfactant</th>
</tr>
</thead>
<tbody>
<tr>
<td>Airway pressure, cmH2O</td>
<td>2.21±0.80</td>
<td>5.00±0.49</td>
<td>5.00±0</td>
</tr>
<tr>
<td>Transpulmonary pressure, cmH2O</td>
<td>6.86±3.23</td>
<td>12.35±3.50*</td>
<td>10.57±3.98†</td>
</tr>
<tr>
<td>Tidal volume, ml/kg</td>
<td>13.33±4.13</td>
<td>8.12±2.23*</td>
<td>7.32±1.33</td>
</tr>
<tr>
<td>Respiratory rate, breaths/min</td>
<td>10.44±2.24</td>
<td>15.27±3.62*</td>
<td>18.25±6.44†</td>
</tr>
<tr>
<td>Inspiratory time, s</td>
<td>1.67±0.24</td>
<td>1.32±0.16*</td>
<td>1.04±0.16†</td>
</tr>
<tr>
<td>Expiratory time, s</td>
<td>4.58±1.68</td>
<td>2.91±1.00*</td>
<td>2.94±1.50</td>
</tr>
<tr>
<td>Lung compliance, ml/cmH2O</td>
<td>6.68±2.30</td>
<td>2.00±0.58*</td>
<td>2.41±0.92†</td>
</tr>
</tbody>
</table>

Values are means ± SD. CPAP, continuous positive airway pressure. Significant difference between *pre- and postlavage values and †postlavage value and value after surfactant instillation: \( P < 0.05 \) (ANOVA, two-tailed paired Student’s t-test).
The instantaneous impulse frequency of PSR activity (fimp) was calculated from the time interval between two consecutive spikes, giving the moving fimp of the entire PSR discharge activity (impulses/s) during the course of every breath. The fimp was plotted against Ptp and Vt, and the maximal fimp was noted for every breath during spontaneous breathing on CPAP. To define the adaptation properties of each receptor (adaptation index <15%) (10, 43), fimp of all PSRs was calculated during sustained inflation on mechanical ventilation. The PSRs were divided into two groups: 1) HT PSRs, if they continued to discharge at end expiration; and 2) LT PSRs, if they continued to discharge at end expiration.

Receptors studied. Initially, there were 14 cats in the study, but, because of technical problems in maintaining the same nerve fiber from a single unit of PSR throughout the two interventions (lung lavage and surfactant instillation), only nine cats completed the entire protocol. In seven cats, one and the same single-unit PSR was studied before and after lung lavage and after surfactant instillation; and in two cats the same single-unit PSR was studied after lung lavage and after instillation of surfactant. Four of the nine PSRs were defined as LT PSRs, and the other five were defined as HT PSRs.

Statistics. Student’s t-test for two-tailed paired observations was used for statistical analysis, and differences were considered significant at P < 0.05.

RESULTS

Lung mechanics, arterial blood gases, and oxygenation. Lung lavage resulted in a 40% decrease in Vt (P < 0.001), an 80% increase in Ptp, and a 70% decrease in Ct (P < 0.001; Table 1). Although Vt remained the same after instillation of surfactant, there was a 25% increase in Ct (P = 0.003) and a marked decrease in Ptp by 15% (P = 0.007; Table 1). All cats developed respiratory acidosis of the same degree during spontaneous breathing on CPAP after lung lavage and after surfactant instillation, as observed from blood-gas measurements after each recording (Table 2). The oxygenation was kept adequate and at the same level during each setting on CPAP, with a decrease in the Fio2 after instillation of surfactant (P = 0.023; Table 2).

PSR activity during individual breaths. The HT PSRs (Figs. 1B and 2B) appeared to be more vulnerable to lung lavage than the LT PSRs (Fig. 3B). Two of the five HT PSRs lost their activity altogether after lung lavage and recovered only part of it after instillation of surfactant, compared with the prelavage value (Fig. 2). The LT PSRs never ceased to discharge completely after lung lavage, but their activity was markedly reduced after lung lavage, especially in relation to Ptp (Fig. 3B). The LT PSR activity increased and recovered with a close relation to both Vt and Ptp after instillation of surfactant (Fig. 3C).

PSR activity related to volume, Ptp, and end-expiratory pressure. Without taking into account changes in Ptp or Vt, the total number of PSR impulses per breath and the maximal fimp of PSRs increased in eight of nine cats after instillation of surfactant (P = 0.041 and P = 0.006, respectively). One of the HT receptors, which lost its activity after instillation of surfac-

Table 2. Arterial blood gases and Fio2 measured before and after lung lavage and after instillation of surfactant during MV before CPAP and during spontaneous breathing on CPAP

<table>
<thead>
<tr>
<th></th>
<th>Before Lung Lavage</th>
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<th>After Lung Lavage</th>
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<th>After Surfactant</th>
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<tbody>
<tr>
<td></td>
<td>MV</td>
<td>CPAP</td>
<td>MV</td>
<td>CPAP</td>
<td>MV</td>
<td>CPAP</td>
</tr>
<tr>
<td>pH</td>
<td>7.38±0.07</td>
<td>7.31±0.04</td>
<td>7.34±0.08</td>
<td>7.22±0.06*</td>
<td>7.35±0.04</td>
<td>7.21±0.08</td>
</tr>
<tr>
<td>Pco2, kPa</td>
<td>4.53±0.72</td>
<td>5.55±0.75</td>
<td>4.74±1.27</td>
<td>7.20±1.32*</td>
<td>5.23±1.31</td>
<td>7.65±1.39</td>
</tr>
<tr>
<td>Po2, kPa</td>
<td>13.24±2.35</td>
<td>10.71±2.72</td>
<td>11.18±6.41</td>
<td>8.97±2.73</td>
<td>10.01±3.83</td>
<td>9.57±1.91</td>
</tr>
<tr>
<td>Base deficit</td>
<td>−4.96±1.9</td>
<td>−5.19±1.57</td>
<td>−5.98±2.56</td>
<td>−6.00±2.63</td>
<td>−3.64±3.50</td>
<td>−4.40±2.57</td>
</tr>
<tr>
<td>FiO2</td>
<td>0.21±0.00</td>
<td>0.21±0.00</td>
<td>1.00±0*</td>
<td>0.84±0.21*</td>
<td>0.80±0.20†</td>
<td>0.68±0.26†</td>
</tr>
</tbody>
</table>

Values are means ± SD. MV, mechanical ventilation; Fio2, inspiratory fraction of oxygen. Significant difference between *pre- and postlavage values and †postlavage value and value after surfactant instillation: P < 0.05 (ANOVA, two-tailed paired Student’s t-test).
tant during spontaneous breathing on CPAP, did show activity during intermittent positive pressure ventilation (peak inspiratory pressure = 20 and positive end-expiratory pressure = 5 cmH₂O), proving that this receptor was not lost.

The maximal f_{imp} of LT PSRs per Ptp was higher than the maximal f_{imp} of HT PSRs per Ptp before lung lavage (range 8–26 vs. 6–11 impulses·s⁻¹·cmH₂O⁻¹; P = 0.0023). All PSRs decreased their maximal f_{imp} per Ptp after lung lavage (Fig. 4; range 0–12 impulses·s⁻¹·cmH₂O⁻¹; P < 0.001), and, in all but one of these nine receptors, the maximal f_{imp} per Ptp increased after instillation of surfactant (Fig. 4; range 0–16 impulses·s⁻¹·cmH₂O⁻¹; P < 0.001).

Three of four LT PSRs increased their maximal f_{imp} per Vt after lung lavage, and two of three HT PSRs decreased their maximal f_{imp} per Vt (Fig. 5). Although Vt remained the same after instillation of surfactant, the increase in f_{imp} of all receptors but one resulted in an increased maximal f_{imp} per Vt (Table 3 and Fig. 5; P < 0.001).

The end-expiratory f_{imp} of all LT PSRs decreased after lung lavage (34 ± 2.0 vs. 19 ± 2.7 impulses/s; means ± SE; P < 0.001) and increased after instillation of surfactant (38 ± 3.4 impulses/s; P < 0.001), also when related to the end-expiratory pressure (Fig. 6 and Table 3).

**Breathing pattern and variability.** The respiratory rate increased after each intervention, with a concomitant decrease in inspiratory time (Table 1). The expiratory time decreased markedly after lung lavage (P < 0.001), but it remained the same after instillation of surfactant (Table 1). The inspiratory-to-expiratory time ratio was 0.36 before lung lavage, 0.45 after lung lavage, and 0.35 after surfactant instillation. The general CV for all parameters measured was low, both after lung lavage and after instillation of surfactant (<6%; Table 4). Both after lung lavage and after instillation of surfactant, the CV for Vt was significantly higher than before lung lavage (Table 4; P = 0.016 and P = 0.017, respectively).

**DISCUSSION**

The most important finding in this study is that PSRs increase their activity after instillation of surfactant in spontaneously breathing surfactant-depleted young cats, but this increase not in parallel with changes in measured lung mechanical parameters such as Ptp and Vt. This increased PSR activity is accompanied by a shorter inspiratory time and a lower inspiratory-to-expiratory time ratio while Vt remains low. The improved receptor activity and Cl in the presence of a continued low ventilatory response...
after surfactant instillation might indicate a stronger inspiratory inhibition by vagally mediated PSRs, as recorded in all animals.

As PSRs have been shown to be mainly sensitive to changes in VT and Ptp in healthy animals (1, 10, 24, 33, 43), it may be anticipated that a decreased compliance, as observed in neonates with RDS (3, 8), might influence PSR discharge and thereby the control of breathing. If the major mechanical stimulus for PSRs is Ptp (9, 10, 43), then the increase in Ptp required to attain a normal inspired volume in a less compliant lung would elicit a higher PSR discharge. The effect of increased PSR activity would hence be an earlier inhibition of inspiration and a prolonged expiration, according to the Hering-Breuer inhibitory inspiratory reflex (19), not taking into account the influence of central and peripheral chemoreceptors or other pulmonary receptors. An increase in compliance after instillation of surfactant, as observed in mechanically ventilated preterm lambs and in spontaneously breathing infants on mechanical ventilation before CPAP (11, 20), would have the reverse effect, i.e., a decrease in PSR activity and less influence by the Hering-Breuer reflex. In the other hand, VT alone would constitute a weaker stimulus on PSR activity in a less compliant lung if the present Ptp were unable to attain the same VT as in the compliant lung.

In artificially ventilated dogs, Sant’Ambrogio et al. (38) found that increased compliance after overinflation (and thereby lowered Ptp) decreased the \( f_{\text{imp}} \) of a majority of intrapulmonary-situated PSRs. Yu et al. (49) noted that a decrease in compliance induced by removing the positive end-expiratory pressure (increased Ptp to maintain the same VT) in artificially ventilated cats led to increased discharge of all PSRs at the peak of inflation, but it led to decreased deflation discharge in HT PSRs and loss of end-expiratory activity in LT PSRs. It is possible that both of these studies represent the response of PSRs to overextended alveoli, in the first example due to hyperinflation of already recruited alveoli (38), and in the second example due to a compensation of collapsed alveoli (49). However, these studies (38, 49) were performed in healthy lungs with intact surfactant content.

In this study, lung lavage was used to reduce the surfactant content of the lungs and thereby decrease compliance (5, 25). Lavage of the lung also causes a decrease in functional residual capacity (FRC), atelectasis, and hyaline membranes (25), but this lung condition still responds to recruitment maneuvers (31, 42) and surfactant treatment (40). In the present study, the compliance decreased by 70% after lung lavage, and the PSRs responded with a markedly lower total number of impulses per breath, and the maximal PSR \( f_{\text{imp}} \) decreased by 46% in the presence of increased Ptp and decreased VT (Tables 1 and 2). The reduced sensitivity of the PSRs to changes in Ptp after lung lavage (Table 3 and Fig. 6) might be explained by changes in the immediate surroundings of the PSRs, especially of the distally located HT PSRs. However, this idea is somewhat contradicted by the finding by Widdicombe (44) that PSRs submitted to major mechanical deformation by pulmonary edema and atelectasis still showed increased sensitivity to Ptp, but in that study lung lavage was not performed. Furthermore, the displacement or possible destruction of surfactant by instillation of a nonionic detergent (Tween 20) has been shown to reduce the number of impulses per ventilator stroke in mechanically ventilated rats (7), an effect also observed in our study after lung lavage.

The specific response of PSRs to instillation of surfactant in the present study might imply an effect on the PSRs that is not directly related to Ptp and VT. It has been postulated from studies of bronchoconstriction, bronchodilation, and microscopy that the likely location of PSRs is within the smooth muscle layer (2). Recent studies of the receptive fields of PSRs have shown that terminal knobs of these receptors are situated in the most peripheral parts of the lung with no relation to smooth muscle (46). In addition, single units of PSRs seem to possess multiple receptive fields with different encoders and discharge characteristics (47), thereby providing more complex information about the lung than is yielded by measurements such as Ptp and VT.

Increased CO2 in inhaled air in cats and increased Pco2 in the pulmonary circulation in dogs have been shown to depress both HT and LT PSR activity (17, 36). In the present study, \( f_{\text{imp}} \) of HT and LT PSRs increased after instillation of surfactant, even though there was a slight but nonsignificant increase in arterial Pco2 after such instillation. In addition, the animals in the present study invariably showed normal arterial Pco2 during mechanical ventilation before CPAP (Table 2), and, after switching from mechanical ventilation to CPAP, the observed PSR discharge did not change during that setting. It is there-
fore, unlikely that arterial Pco2 influenced the PSR activity in any substantial way during CPAP. On the other hand, the influence of central chemoreceptors on the breathing pattern in the surfactant-depleted cats cannot be excluded, although no difference in arterial Pco2 could be observed before and after surfactant instillation during these short recording intervals.

FRC of the lung in RDS and in models of RDS increases after instillation of surfactant, both in infants and in animals, and is accompanied by a reduction in arterial Pco2 (13, 16, 37). In the present study, the increase in Cl (Table 1) after instillation of surfactant may be accompanied by an increase in FRC, although arterial Pco2 was the same as before surfactant instillation. An increase in FRC could also explain the regained end-expiratory activity of LT PSRs (Figs. 3 and 6), as suggested by other authors (9, 29). When corrected for the end-expiratory pressure, the end-expiratory activity of LT PSRs still remained higher after than before instillation of surfactant (Table 3; Fig. 6), implying a regained tonic activity of these receptors.

One hypothesis tested in this study was that instillation of surfactant would improve lung mechanics and PSR activity and thereby lead to a more optimal breathing pattern in terms of frequency and depth. Without excluding the influence of central chemoreceptors on the setting of the respiratory rate in the surfactant-depleted cats before and after surfactant instillation, and regional changes in peripheral airways, the breathing remained rapid and shallow after instillation of surfactant in the present study, although Cl and fimp increased (Figs. 1–3, Tables 1 and 3). Increased PSR activity would result in an earlier inhibition of inspiration and prolonged expiration, as suggested by Zuperku et al. (50) and Gautier et al. (15). This was verified in the present study by a decrease in inspiratory time after surfactant instillation and resumption of a lower inspiratory-to-expiratory time ratio comparable to that observed before lung lavage (Table 1; Figs. 1–3).

The importance of establishing continuous and regular breathing at birth to achieve effective gas exchange has been investigated in several studies by performing pre- and postnatal vagotomy (6, 26, 45). Both prenatal and postnatal vagal denervation in lambs have been shown to cause major early postnatal reductions in gas exchange, breathing frequency, minute ventilation, and Cl, as well as surfactant dysfunction with increased surface tension (23, 45). A gradual increase in frequency of augmented breaths was noted over time in the postnatally vagally denervated lambs (26), which meant that other compensatory mechanisms to improve gas exchange could not be excluded. In the present study, no reductions in breathing frequency or irregularities in the breathing pattern as a consequence of diminished or absent vagal input from PSRs were observed, either before or after instillation of surfactant. Not excluding the influence of chemoreceptors on the setting of the respiratory rate, we cannot rule out the possibility that the rapid shallow breathing, especially when the lung is surfactant depleted, could be caused by increased activity of other vagal afferents, such as C fibers, and/or of rapidly adapting pulmonary stretch receptors (RARs). As the activity of RARs has been shown to be inversely related to Cl both in dogs and in cats (22, 48), then the increase in compliance after surfactant instillation should have reduced their activity and thereby their influence on the breathing pattern in the present study.

The observed changes in lung mechanics and breathing pattern, in the present study of surfactant-depleted spontaneously breathing cats, show similarities to findings in clinical studies of spontaneously breathing infants with RDS (11, 13, 16, 28). Davis et al. (11) assessed lung mechanics in infants

Table 3. PSR fimp related to Ptp, Vt, and EEP before and after lung lavage and after instillation of surfactant

<table>
<thead>
<tr>
<th></th>
<th>Before Lung Lavage</th>
<th>After Lung Lavage</th>
<th>After Surfactant</th>
</tr>
</thead>
<tbody>
<tr>
<td>Maximal PSR fimp per breath, impulses/s</td>
<td>70.5±5.6</td>
<td>37.2±10.1*</td>
<td>50.6±9.6†‡</td>
</tr>
<tr>
<td>Total number of PSR impulses per breath, impulses/s</td>
<td>40.3±4.3</td>
<td>18.3±3.7*</td>
<td>31.2±6.4‡</td>
</tr>
<tr>
<td>Maximal PSR fimp related to Ptp, impulses s⁻¹·cmH₂O⁻¹</td>
<td>14.09±2.29</td>
<td>4.69±1.31*</td>
<td>6.16±1.54‡‡</td>
</tr>
<tr>
<td>Maximal PSR fimp related to EEP, impulses s⁻¹·cmH₂O⁻¹</td>
<td>1.95±0.25</td>
<td>1.97±0.63</td>
<td>2.65±0.78‡‡</td>
</tr>
<tr>
<td>End-expiratory LT PSR fimp related to EEP, impulses s⁻¹·cmH₂O⁻¹</td>
<td>23.35±2.159</td>
<td>5.32±0.79*</td>
<td>11.86±1.12‡‡</td>
</tr>
</tbody>
</table>

Values are means ± SE. PSR, slowly adapting pulmonary stretch receptor; fimp, impulse frequency; Ptp, transpulmonary pressure; Vt, tidal volume; LT, low threshold; EEP, end-expiratory pressure. Significant difference between *pre- and postlavage values, †prelavage value and value after instillation of surfactant, and ‡prelavage value and value after instillation of surfactant: P < 0.01 (ANOVA, two-tailed paired Student’s t-test).

Table 4. CV for defining irregularity of breathing pattern, before and after lung lavage and after surfactant instillation during spontaneous breathing on CPAP

<table>
<thead>
<tr>
<th></th>
<th>Before Lung Lavage</th>
<th>After Lung Lavage</th>
<th>After Instillation of Surfactant</th>
</tr>
</thead>
<tbody>
<tr>
<td>CV for respiratory rate, %</td>
<td>2.40±0.95</td>
<td>3.01±1.33</td>
<td>3.38±1.70</td>
</tr>
<tr>
<td>CV for duration of inspiration, %</td>
<td>5.84±0.89</td>
<td>5.58±3.38</td>
<td>5.23±1.14</td>
</tr>
<tr>
<td>CV for duration of expiration, %</td>
<td>4.45±2.84</td>
<td>3.64±1.59</td>
<td>4.06±1.83</td>
</tr>
<tr>
<td>CV for Vt, %</td>
<td>2.56±1.30</td>
<td>5.70±3.59*</td>
<td>5.46±3.34†</td>
</tr>
</tbody>
</table>

Values are means ± SD. CV, coefficient of variation. Significant difference between *pre- and postlavage values and †prelavage value and value after surfactant instillation: P < 0.05 (Wilcoxon’s signed rank test).
with RDS, spontaneously breathing on CPAP, before and after instillation of surfactant, and found an increase in Ct. (29%) after this instillation, together with maintained rapid and shallow breathing, suggesting similarities in the control of breathing between infants with RDS and young cats submitted to lung lavage.

In summary, the most important finding in this study is that all PSRs respond with increased activity after instillation of surfactant in surfactant-depleted, spontaneously breathing cats. The decrease in the inspiratory-to-expiratory time ratio to the same level as before lung lavage indicates increased influence of PSRs on the breathing pattern during recovery from RDS.

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