Surfactant Administration through Laryngeal Mask Airway: A Randomized Controlled Study in Rabbits

Cristian A. Bernardo, MD1,2  Gonzalo L. Mariani, MD1,2  María de los Ángeles Virasoro, MD1  Sebastián Burgos, RN3  María J. Labanca, MD4  Pablo Otero, DVM, PhD5

1 Division of Neonatology, Department of Pediatrics, Hospital Italiano de Buenos Aires, Ciudad de Buenos Aires, Argentina
2 School of Medicine, Instituto Universitario, Hospital Italiano de Buenos Aires, Ciudad de Buenos Aires, Argentina
3 Department of Nursing, Hospital Italiano de Buenos Aires, Ciudad de Buenos Aires, Argentina
4 Division of Pathology, Department of Diagnosis and Treatment, Hospital Italiano de Buenos Aires, Ciudad de Buenos Aires, Argentina
5 Department of Anesthesiology, Faculty of Veterinary Science, University of Buenos Aires, Buenos Aires, Argentina

Address for correspondence Cristian A. Bernardo, MD, Division of Neonatology, Department of Pediatrics, Hospital Italiano de Buenos Aires, 25 de Mayo 1551, Dept. 3, PC 7000, Tandil, Buenos Aires, Argentina (e-mail: cristian.bernardo@hospitalitaliano.org.ar).

Abstract

Background  Minimally invasive techniques for surfactant administration for infants with respiratory distress syndrome (RDS) of moderate severity have been proposed. The laryngeal mask airway (LMA) helps in securing the airway without the need of laryngoscopy, but still requires the use of positive pressure ventilation (PPV) to flush surfactant into the lungs.

Objective  This article compares the effectiveness of two techniques for LMA surfactant administration, instillation into the LMA lumen followed by PPV versus direct laryngeal instillation through a preinserted feeding tube inside the LMA during spontaneous respirations.

Study Design  This is a randomized controlled trial (RCT) of 18 rabbits with acquired respiratory distress after lung lavage. After surfactant was given, the rabbits remained on continuous positive airway pressure (CPAP). Gas exchange parameters were assessed at baseline and at 30 minutes and lung parenchyma pathology features were analyzed.

Results  Time required for surfactant administration, oxygenation improvement, and histopathologic findings did not differ between groups. The new technique decreased the need of PPV ($p < 0.05$).

Conclusion  In this animal model, surfactant administration through a preinserted feeding tube within the LMA lumen is safe and effective while providing the benefits of a minimally invasive approach. This technique reduces the need of PPV and may prevent its potential risks.
Recent studies have shown benefits of early nasal continuous positive airway pressure (nCPAP) compared with intubation for surfactant administration followed by mechanical ventilation (MV) in neonates with respiratory distress syndrome (RDS) of moderate severity.1–5 However, in neonates who have increasing fraction of inspired oxygen (FiO2) requirements, the administration of early surfactant decreases the incidence of death or bronchopulmonary dysplasia (BPD) compared with delayed administration.6 To avoid MV, the intubation–surfactant–extubation (INSURE) strategy has shown some benefits,7–10 but the potential undesirable effects of intubation and positive pressure ventilation (PPV), even for brief periods of time, are still a concern.11,12 Alternative and less invasive techniques for surfactant administration have been tested.13–16 Some studies have evaluated the use of a thin laryngeal catheter under direct laryngoscopy.17–19 This technique has shown promising results, but it does not avoid the potential trauma associated with laryngoscopy, and it does not provide a secure airway if resuscitation maneuvers were needed during or after surfactant administration.

The use of a laryngeal mask airway (LMA) for surfactant has been proven to be safe and feasible in several small studies in larger preterm infants.13,20,21 and in recent randomized controlled trials (RCTs).22,23 In all published studies about LMA use, surfactant was given in aliquots followed by PPV to flush surfactant into the lungs to prevent a transient obstruction of the airway, increasing the potential risk of volutrauma. To avoid PPV, a combined technique is proposed, which involves preinserting a K33 feeding tube inside the LMA, slightly passing the distal tip of the mask, and then proceed to insert it and instill the surfactant, while the patient keeps breathing spontaneously. This novel procedure proposes the combination of the LMA use with the Hobart technique,14,19 and can provide a secure airway while avoiding direct laryngoscopy. We aimed to evaluate this new technique in rabbits and to compare it with surfactant administration via LMA lumen.

Methods

Objective
The objective of this experiment was to compare the effectiveness of surfactant administration through instillation into the LMA lumen followed by PPV versus direct laryngeal instillation through a preinserted feeding tube inside the LMA during spontaneous breathing in an animal model of RDS.

Design and Subjects
This is a randomized controlled study in 18 adult New Zealand rabbits with acquired respiratory distress after lung lavage.

Protocol
Rabbits were sedated with intravenous (IV) ketamine (30 mg/kg) and midazolam (1 mg/kg) before intubation with a 2.5-mm endotracheal tube (ETT). Continuous propofol (1 mg/kg/min) was given during lung lavage, which was done with repeated normal saline (NS) boluses at 10 mL/kg until reaching a stable Spo2 of 80 to 90% for 3 to 5 minutes, breathing spontaneously on CPAP (5 cmH2O) with an FiO2 of 1.0. Additionally, an arterial line was set for arterial blood gas (ABG) sampling. After lung lavage, rabbits were randomized to receive surfactant into the LMA lumen followed by PPV (LMA-PPV group) or to receive surfactant directly into the larynx through a preinserted catheter inside the LMA (Cath-LMA group) (Fig. 1). Randomization was achieved through permuted blocks (blocks of two, four, and six opaque and sealed envelopes). In the LMA-PPV group, PPV was administered after surfactant instillation using a T-piece resuscitator device Neopuff (Fisher & Paykel Healthcare, Auckland, New Zealand), until surfactant was cleared out from the LMA. In the Cath-LMA group, surfactant was given directly into the larynx through a narrow feeding tube (6Fr) preinserted inside the LMA, without the use of PPV.

The LMA used was rabbit specific, V-Gel Advanced Veterinary Airway Management System (Docsinnovent, LTD, London, UK) and its placement followed a specific protocol24 Each rabbit received 3 mL of poractant alfa, Curosurf (Chiesi Farmaceutici, Parma, Italy) with 0.3 mL of methylene blue to assess leakage of surfactant around the LMA.

After the administration of surfactant, rabbits remained on CPAP (5 cmH2O) through the LMA and were monitored for 30 minutes. The FiO2 during this period was the one needed...
to maintain target SpO2 of 88 to 95%. After the 30-minute period, animals were euthanized by giving 20 mg of IV propofol and 2 mL of IV KCl.

**Measured Variables**

Total time of surfactant administration, use of PPV, values of heart rate (HR), FiO2, and SpO2 during the study was recorded. ABG samples were taken at 0 and 30 minutes after surfactant administration, and the arterial to alveolar partial pressure of oxygen (a/A PaO2) ratios were calculated.

For the histopathological analysis of the pulmonary parenchyma, the lungs of the 18 rabbits were fixed by insufflation in 10% formalin, and the pathologist performed six sections per animal for each lung lobe embedded in paraffin and stained with hematoxylin and eosin. Digital images of each histological preparation (six per animal) were obtained at 100× magnification, which were then studied by image analyzer (Image-Pro Plus, Media Cybernetics, Inc, Rockville, MD). First, the corresponding alveolar spaces surface (air surface/parenchyma surface) present in each section was calculated, and then, the average for each of the lungs was determined.

The pathologist later classified the respiratory parenchyma in four categories: 1 = alveoli within a normal range, 2 = collapsed alveoli, 3 = distended alveoli, and 4 = over-distended alveoli, by morphological benchmarks. Each image was superimposed on a grid of 120 points and the number of points that coincide with each category was counted. Finally, dividing the result of each category by the total points (excluding those that do not coincide with any alveoli), the corresponding proportion was obtained, which was also expressed as average percentage of each lung.

The pathologist (M.J.L.) who analyzed all lung samples was blinded to group assignment.

**Ethical Considerations**

The protocol was approved by the local institutional review board (IRB; Hospital Italiano de Buenos Aires). The investigators followed the regulations proposed by the Canadian Council on Animal Care (CCAC) to ensure proper management of anesthesia and analgesia to avoid any type of animal suffering.

**Statistical Analysis**

For this exploratory study, we did not calculate a sample size. The number of animals seems reasonable in comparison with other studies from different researchers in the field. Considering the sample size, the results were analyzed with the Mann–Whitney test, and a p-value of less than 0.05 was considered statistically significant.

**Results**

The 18 rabbits tolerated the procedure well and did not exhibit unfavorable changes in vital signs during LMA placement. Demographic characteristics and presurfactant status of rabbits in both groups were comparable (Table 1). Data on lung lavage, LMA placement, surfactant administration, and baseline conditions are from the 18 rabbits. Five rabbits (three from the LMA-PPV group and two from the Cath-LMA group) had to be excluded from the outcomes analyses because of technical problems with the air-oxygen blender.

Median duration for LMA placement and surfactant administration was 92 seconds [interquartile range (IQR): 75–104] in the LMA-PPV group and 87 seconds (IQR: 62–146) in the Cath-LMA group (not significant [NS]). Only one rabbit required PPV due to desaturation after surfactant administration in the Cath-LMA group (100% in the LMA-PPV group by protocol, p < 0.05).

**Table 1 Demographic and postlung lavage characteristics of rabbits**

<table>
<thead>
<tr>
<th></th>
<th>LMA-PPV group</th>
<th>Cath-LMA group</th>
</tr>
</thead>
<tbody>
<tr>
<td>N</td>
<td>10</td>
<td>8</td>
</tr>
<tr>
<td>Weight (mean ± SD)</td>
<td>3,951 ± 770 g</td>
<td>3,594 ± 760 g</td>
</tr>
<tr>
<td>Female (%)</td>
<td>50</td>
<td>38</td>
</tr>
<tr>
<td>HR (mean ± SD)</td>
<td>232 ± 37</td>
<td>227 ± 52</td>
</tr>
<tr>
<td>RR median; IQR</td>
<td>82 (52–121)</td>
<td>105 (63–134)</td>
</tr>
<tr>
<td>FiO2</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>SpO2 median; IQR</td>
<td>81% (80–83)</td>
<td>83% (82–89)</td>
</tr>
<tr>
<td>a/A PO2 median; IQR</td>
<td>0.09 (0.08–0.09)</td>
<td>0.09 (0.08–0.1)</td>
</tr>
</tbody>
</table>

Abbreviations: a/A PO2, arterial to alveolar partial pressure of oxygen ratio; Cath-LMA group, preinserted catheter through the laryngeal mask airway group; FiO2, fraction of inspired oxygen; LMA-PPV group, laryngeal mask airway-positive pressure ventilation group; HR, heart rate; IQR, interquartile range; RR, respiratory rate; SD, standard deviation; SpO2, peripheral capillary oxygen saturation.
FiO₂ was reduced after surfactant administration in both groups. Median a/A pO₂ ratio at 30 minutes was 0.18 (IQR: 0.17–1) in the LMA-PPV group versus 0.34 (IQR: 0.2–0.5) in the Cath-LMA group (NS). The proportional increase of a/A pO₂ ratio after surfactant administration was +0.09 (IQR: 0.08–0.9) in the LMA-PPV group and +0.24 (IQR: 0.12–0.42) in the Cath-LMA group (NS).

Assessment of leakage around the LMA by visual inspection of dissected upper airway revealed more intense blue staining of hard palate in the LMA-PPV group (Fig. 3). However, lung histopathology results did not differ between groups (Table 2).

Discussion

Our experimental study was designed to assess the effectiveness of surfactant administration in rabbits by two different techniques using an LMA as interface. Both the standard and the novel technique were effective and safe, achieving a significant reduction in oxygen requirement and improvements in the a/A PaO₂ ratio. The new technique, using a feeding tube preinserted within the LMA, has the additional advantage of avoiding the need to provide PPV and the risk of lung injury. We did not find any significant impact on vital signs during the procedure in this small sample of studied rabbits.

Table 2  Main results

<table>
<thead>
<tr>
<th></th>
<th>LMA-PPV group (n = 10)</th>
<th>Cath-LMA group (n = 8)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Time of LMA placement + surfactant administration (median; IQR)</td>
<td>92 s (75–104)</td>
<td>87 s (62–146)</td>
</tr>
<tr>
<td>Received PPV</td>
<td>10/10 (100%)</td>
<td>1/8 (12%)</td>
</tr>
<tr>
<td>a/A PO₂ (median; IQR)²</td>
<td>0.18 (0.17–1)</td>
<td>0.34 (0.2–0.5)</td>
</tr>
<tr>
<td>Change in a/A PO₂ (median; IQR)²</td>
<td>+0.09 (0.08–0.9)</td>
<td>+0.24 (0.12–0.42)</td>
</tr>
<tr>
<td>Collapsed alveoli (median; IQR)²</td>
<td>48% (32–70)</td>
<td>48% (40–65)</td>
</tr>
<tr>
<td>Overdistended alveoli (median; IQR)²</td>
<td>14% (1–24)</td>
<td>9% (2–18)</td>
</tr>
</tbody>
</table>

Abbreviations: a/A PO₂, arterial to alveolar partial pressure of oxygen ratio; Cath-LMA group, preinserted catheter through the LMA group; LMA, laryngeal mask airway; NS, not significant; PPV, positive pressure ventilation; IQR, interquartile range.

²These outcome analyses excluded three rabbits from the LMA-PPV group and two rabbits from the Cath-LMA group because of technical problems with the air-oxygen blender.
In preterm infants with moderate RDS, current management consensus is to start with nCPAP and to offer early rescue surfactant therapy if needed.\textsuperscript{27,28} However, in neonates who have increasing \textit{O}_2 requirements, there are controversies regarding the adequate timing of surfactant administration and the identification of predictors of early nCPAP failure.\textsuperscript{29–31} The INSURE strategy has beneficial effects when compared with intubation and MV,\textsuperscript{32} particularly when it is provided in the early stages of the RDS in neonates receiving CPAP.\textsuperscript{10} However, this technique requires adequate sedation, intubation, and PPV. Tracheal intubation may damage the airway and it is recommended to use sedatives and analgesics, which may have undesirable side effects. Furthermore, PPV can damage the preterm lung and surfactant distribution may be suboptimal.\textsuperscript{33} Currently, the dilemma has shifted toward the way of surfactant administration, considering the potential advantages of less invasive strategies\textsuperscript{34} (\textit{→ Table 3}).

A less invasive surfactant administration (LISA) technique has been described, involving surfactant administration using a feeding tube, umbilical catheter, or a small angiocatheter while the neonate is breathing spontaneously and receiving nCPAP. This technique was evaluated in 16 preterm infants with RDS using electrical impedance tomography, showing a rapid and homogeneous increase in end-expiratory lung volumes, which was associated with an improvement in oxygenation.\textsuperscript{35} Variations of this technique have been published,\textsuperscript{18,19,36} and a recent systematic review of available RCTs showed that in noninvasively ventilated preterm infants, the use of a LISA strategy is beneficial in terms of reduction in the need for MV, in the composite outcome of death or BPD at 36 weeks, and in BPD rate among survivors.\textsuperscript{37} There was also a trend toward lower pneumothorax rates. The pulmonary benefits seen with the less invasive techniques may be related to many factors including: avoiding the use of PPV, depending on spontaneous respirations for lung surfactant distribution, and allowing for continuation of nCPAP support during surfactant administration, thus preventing lung injury that could result from manual breaths, uneven surfactant incorporation, and avoidance of the temporary loss of functional lung capacity and atelectasis during the process of intubation.\textsuperscript{11,12,38} However, this LISA technique requires laryngoscopy, with its potential associated trauma, and does not provide a secure airway if resuscitation maneuvers are needed (e.g., PPV). To avoid these problems, the use of an LMA for surfactant administration seems to be a feasible option. Some reports in preterm human infants suggest that LMA use for surfactant administration is feasible and comparable to conventional therapy.\textsuperscript{19–21} A study comparing the use of an LMA versus an ETT for surfactant administration in newborn piglets showed that the improvement in oxygenation did not differ between groups and that the LMA device could be placed more easily and with less physiologic disturbances than an ETT.\textsuperscript{29} Pinheiro et al compared LMA versus INSURE for surfactant delivery in a RCT of 61 patients, showing comparable results in oxygenation improvement and subsequent need for MV.\textsuperscript{22} Similarly, Barbosa et al randomized 48 preterm infants to LMA or ETT for surfactant administration and found no difference in the need for supplementary oxygen.\textsuperscript{23} A recent unpublished RCT showed decreased need for intubation and MV with early LMA surfactant administration compared with continuing on nCPAP (K. Roberts et al. E-PAS 2016: 4470.2).

All these studies have evaluated the administration of surfactant through LMA using the intillation of surfactant midway down the airway lumen followed by manual ventilation to help drug dispersal, like our LMA-PPV group (we use a T-piece resuscitator). In comparison to this standard technique, surfactant administration using a preinserted catheter (\textit{→ Fig. 1}) as in our Cath-LMA group was done in the same time and with the advantage of using patient spontaneous breathing, potentially improving surfactant lung distribution.\textsuperscript{12} Trevisanuto and Marchetto have commented on the use of a feeding tube inserted into the LMA in three patients, stating that they did not find oxygenation improvement when the feeding tube was positioned beyond the distal part of the LMA aiming to cross the vocal cords.\textsuperscript{40} This is in contrast with our animal data, as we did find improvement in the a/A \textit{PaO}_2 ratio with this technique.

The trend toward a lesser percentage of overdistended alveoli in the Cath-LMA group might be associated with the avoidance of routine PPV administration and the hypothesis that our technique allows a more physiologic surfactant distribution during spontaneous breathing. Concerns persist about the finding of 48% of collapsed alveoli in both groups. Unfortunately, we did not have X-rays to assess lung volume during the study, so clinically we based the efficacy of the surfactant administered upon the reduction in supplemental oxygen need, evidenced by the increase in a/A \textit{PaO}_2 ratio. While the ratio of collapsed alveoli was high in both groups, there were no substantial differences among each lung lobes in terms of the average of the four alveoli categories (normal, collapse, distended, overdistended), so it might be assumed

### Table 3 Characteristics of different approaches of surfactant administration

<table>
<thead>
<tr>
<th></th>
<th>Sedation</th>
<th>Laryngoscopy</th>
<th>Intubation</th>
<th>PPV</th>
<th>On CPAP</th>
<th>Secure airway</th>
<th>ELBW infants</th>
</tr>
</thead>
<tbody>
<tr>
<td>INSURE strategy</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>–</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>LISA</td>
<td>–</td>
<td>+</td>
<td>–</td>
<td>–</td>
<td>±</td>
<td>–</td>
<td>+</td>
</tr>
<tr>
<td>Standard LMA</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>+</td>
<td>–</td>
<td>+</td>
<td>–</td>
</tr>
<tr>
<td>Cath-LMA group</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>+</td>
<td>–</td>
<td>–</td>
</tr>
</tbody>
</table>

Abbreviations: Cath-LMA group, preinserted catheter through the LMA group; CPAP, continuous positive airway pressure; ELBW, extremely low birth weight; INSURE, intubation–surfactant–extubation strategy; LISA, less invasive surfactant administration; LMA, laryngeal mask airway; PPV, positive pressure ventilation.
that surfactant’s distribution was even. We speculated that the high ratio of collapsed alveoli was probably associated with the use of a suboptimal CPAP level (5 cm H2O), which could have been too low for these adult rabbits.

We also identify certain limitations in our study. First, the sample size resulted small to reach statistical evidence of difference in the oxygenation parameters between the techniques. Second, the residual effect of sedative medication used could adversely affect the animal’s respiratory effort and might be related to the percentage of collapsed alveoli. In addition, it is noteworthy that the adult rabbits with acquired RDS may not reflect the same behavior of an immature lung with hyaline membrane disease on respiratory support. While the proposed technique looks promising, it should be studied in human infants before any definitive conclusion can be made. Unfortunately, there are no LMA support. While the proposed technique looks promising, it cannot be used in the extremely preterm infants and, for now, it is reserved for neonates above 1,200 g.13

Conclusion

In an animal model of RDS, surfactant administration through a preinserted K33 feeding tube within the LMA lumen is a safe and effective technique that allows to combine the benefits of minimally invasive approaches for moderate RDS treatment. When using an LMA for surfactant administration, our technique reduces the need of PPV and may prevent its potential risks. Further studies involving human infants are necessary to confirm these data and to explore the potential beneficial role of this approach of minimally invasive surfactant administration.

Conflict of Interest
None.

Funding
The study was funded by Fundación Carlos Giamantonio, Buenos Aires, Argentina. The Curosurf vials used were received as a donation from Chiesi Farmaceutici, Parma, Italy.

Acknowledgments
We deeply thank Prof. Waldemar A. Carlo for his critical advice and thorough review of the manuscript; Luis Sanjurjo for his technical assistance during the development of the experiment; and Chiesi Farmaceutici, Parma, Italy, for the donation of the Curosurf vials.

References
10 Isayama T, Chai-Adisaksopha C, McDonald SD. Noninvasive ventilation with vs without early surfactant to prevent chronic lung disease in preterm infants: a systematic review and meta-analysis. JAMA Pediatr 2015;169(08):731–739
17 Aguar M, Ceradina M, Brugada M, Gimeno A, Gutierrez A, Vento M. Minimally invasive surfactant therapy with a gastric tube is as effective as the intubation, surfactant, and extubation technique in preterm babies. Acta Paediatr 2014;103(06):e229–e233
20 Brimacombe J, Gandini D, Keller C. The laryngeal mask airway for administration of surfactant in two neonates with respiratory distress syndrome. Paediatr Anaesth 2004;14(02):188–190
32 Stevens TP, Harrington EW, Blennow M, Soll RF. Early surfactant administration with brief ventilation vs. selective surfactant and continued mechanical ventilation for preterm infants with or at risk for respiratory distress syndrome. Cochrane Database Syst Rev 2007;17(04):CD003063
34 Dargaville PA. CPAP, surfactant, or both for the preterm infant: resolving the dilemma. JAMA Pediatr 2015;169(08):715–717