Lung lavage and surfactant replacement during ECMO in a severe ARDS aspiration pneumonia model

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INTRODUCTION

Acute respiratory distress syndrome (ARDS) is a life-threatening form of acute respiratory failure characterized by inflammatory pulmonary edema secondary to the impairment of the alveolar-capillary barrier induced by direct (aspiration, pulmonary contusion) or indirect (sepsis, pancreatitis, trauma) mechanisms, resulting in severe hypoxemia (1, 2). ARDS is defined by the presence of clinical and radiographic variables that include acute onset, bilateral opacities on chest radiography, absence of cardiogenic causes for respiratory failure, and hypoxemia (1). The severity of ARDS is classified according to the degree of hypoxemia (PaO_2/FiO_2 ratio), with mutually exclusive categories of mild (PaO_2/FiO_2 201-300 mmHg), moderate (PaO_2/FiO_2 101-200 mmHg), and severe (PaO_2/FiO_2 ≤100 mmHg), each one associated with an increased mortality rate of 27% (95%CI, 24%-30%), 32% (95%CI, 29%-34%), and 45% (95% CI, 42%-48%), respectively (1). ARDS is common, with a crude incidence of 78.9 per 100,000 person-years, and hence has a substantial impact on public health (3, 4). Despite decades of research, there are, in fact, limited therapeutic options directed at the underlying pathological processes (5, 6), and supportive care with low tidal volumes of 6 ml/kg of predicted body weight mechanical ventilation remains the cornerstone of patient management (7, 8). However, when conventional mechanical ventilation strategies fail to provide adequate support, patients are treated with rescue therapies, including veno-venous extracorporeal membrane oxygenation (VV-ECMO), which is worldwide increasingly used with this indication (9-12).

Recently, an interesting study evaluated the therapeutic effects of saline lavage and exogenous surfactant replacement in an experimental model of aspiration-induced acute lung injury lung (13). In this model, during ex vivo lung perfusion (EVLP) of lungs injured through gastric juice instillation, saline lavage followed by surfactant replacement therapy resulted in better physiologic lung function and reduced inflammation compared to not treated controls. These results suggest that lung lavage may act to remove the cause of lung injury, including aspiration content, inflammatory mediators, and aspiration-induced dysfunctional surfactant (14, 15), which can then be replaced with exogenous active surfactant.

Exogenous surfactant instillation along with recruitment maneuvers (RM) have been shown to be beneficial in an ARDS model in rabbits (17) and demonstrated to have beneficial effects on pulmonary compliance in patients with ARDS (18). Nonetheless, these experimental results were not confirmed in randomized clinical trials, which failed to show efficacy of surfactant replacement therapy in reducing mortality in adults with ARDS. Questions remain incompletely answered surrounding the ideal timing, techniques and dosing of surfactant treatment in adults with ARDS (14-16). Moreover, studies on surfactant replacement therapy in ARDS have not included a lung lavage treatment preceding the surfactant administration.

Since VV-ECMO efficiently provides adequate gas exchange in either minimal or no contribution of injured lungs, it allows the safe administration of bronchoscopy-based treatments during mechanical ventilation even in extremely severe ARDS. Therefore, we sought to evaluate in vivo the effect of saline lavage followed by surfactant replacement therapy during ECMO in a pre-clinical severe ARDS aspiration porcine model.

EXPERIMENTAL AIMS

Establish a severe ARDS (PaO_2/FiO_2 ratio ≤100 mmHg) model in pigs that is reproducible and warrants MV and VV-ECMO support.

1. Evaluate the efficacy of saline lavage followed by surfactant replacement during VV-ECMO support in this model.

MATERIALS AND METHODS

Animal preparation: Yorkshire male domestic pigs (29-37 kg) were placed supine under general anesthesia. Animals underwent tracheostomy and received pressure control ventilation (PCV) with 15 cmH_2O of driving pressure, 5 cmH_2O of positive end expiratory pressure (PEEP), FiO_2 50%, and inspiratory:expiratory ratio 1:2. The respiratory rate set at 15 breaths per minute (bpm) was adjusted to achieve a paCO2 of 35-40 mmHg. The carotid artery was cannulated for blood pressure measurement and blood gas analysis. Subsequently, a pulmonary artery (PA) catheter was placed via pressure wave guidance. Vital signs, including pulmonary arterial pressure, and lung function including static compliance - calculated as tidal volume divided by the difference between plateau pressure and PEEP, were monitored and recorded hourly. The animals in the two groups received a similar amount of fluid during the experiment.

ECMO cannulation and management: Prior to lung injury and under systemic heparinization, open cannulation was performed introducing a 15 Fr (50 cm) drainage cannula in the femoral vein and a 14 Fr (10 cm) return cannula in the external jugular vein. Cannulas' placement was confirmed by chest radiography. ECMO blood flow was titrated to maintain mean arterial pressure (MAP) at 65 mmHg. 1 hour post lung injury ECMO sweep gas was increased from 0 to 2 L/min. 30 IU/kg of intravenous (IV) heparin were administrated every hour to prevent ECMO clotting, and a continuous IV infusion of fluid was provided at the rate of 4-10 mL/kg/hr.

ARDS induction: Severe ARDS was caused using two bronchoscopic instillations of **gastric juice** (GJ) (4 ml/kg and 2 ml/kg, pH 1.6) 30 min apart to target a PaO_2/FiO_2 ≤100 mmHg after 1 hour from the first GJ instillation. After the each instillation animals received volume controlled ventilation (VCV) with tidal volume of 10 mL/kg, unchanged respiratory rate, and 100% FiO_2, for five min, and then returned to preinjury settings maintaining 100% of FiO_2. When the ECMO sweep gas was increased to 2 L/min, the FiO_2 was lowered to 50% for the rest of the experiment. 1h post injury, a sample of broncho-alveolar lavage fluid (BAL) was collected for future analysis. Two hours post injury, a chest X-ray was performed.

Randomization: Animals were randomized after injury to receive either treatment with lavage + surfactant + recruitment maneuver (RM) (treatment group n=5), or only a RM (control group n=5).

Treatment group (LS): 2h post injury, 200 mL of saline was used to lavage the lungs and promptly recovered with active suction. Surfactant was administered immediately after the lavage. A natural surfactant extracted from bovine lung lavage (BLES® Biochemicals Inc.) was used (135 mg/kg). During surfactant instillation and for the following 5 minutes the animals received VCV with tidal volume of 10 mL/kg, 5 cmH_2O of PEEP, and 3 sustained inflations with high airway pressure (RM= 30 cmH_2O for 10s each) to facilitate alveolar recruitment and surfactant distribution.

Control group (C): 2 h post injury the animals received VCV with tidal volume of 10 mL/kg, 5 cmH_2O of PEEP, and 3 sustained inflations (RM=30 cmH_2O for 10s each).

Post-treatment time course: After the intervention (RM only or lavage+surfactant+RM), animals were supported on VV-ECMO and monitored for 4h, at the end of which a sample of BAL was collected. Then, the ECMO sweep gas was turned off, and the animals were monitored for one additional hour. During this hour, the respiratory rate was adjusted to counter the rise in pCO_2. At the end of the experiment, animals were euthanized and median sternotomy was performed for gross macroscopic exam and tissue sample collection.

RESULTS

The baseline characteristics of the animals in the two study groups were very similar without significant differences (Table 1). The bronchoscopic instillations of GJ caused a significant and consistent decrease in oxygenation and compliance, which were comparable in the two groups. In particular, the PaO_2/FiO_2 dropped to 62.8 ± 9.1 and 60.9 ± 9.6 in the control and treatment group, respectively (p=0.95). Moreover, the chest radiography performed 2 hours after injury consistently showed bilateral infiltrates in lung fields in all cases where chest radiography was available (7/10 cases, data not shown). The lung injury caused also hemodynamic instability, which was comparable in the two groups (Table 1). Gas exchange and hemodynamic parameters recovered significantly in all the animals after the ECMO sweep gas was turned on to 2 L/min to start extracorporeal respiratory support (Table 1).

The animals randomized to the treatment group received lung lavage followed by surfactant replacement. The average recovered fluid from the 200mL of saline lavage was 134 ± 16.6 mL. The mean volume of surfactant instilled was 164.8 ± 9.1 mL. During treatment, the SpO_2 remained >88% in all cases. However, the treatment caused an immediate and significant drop of expired tidal volume (delta = 66.8 ± 14.4 mL), PaO_2/FiO_2 (delta = 28.7 ± 35.1 mmHg) and Csta (delta = 4.63±1.3 mL/cmH_2O), which progressively improved over the following 4 hours. Before the ECMO sweep gas was turned off, the physiologic parameters in the two study groups were not significantly different (Figures 1 and 2).

At the end of the study period, 1 hour after the ECMO sweep gas was turned off, PaO_2/FiO_2 was higher in the treatment group compared to control (C: 203.2 ± 109.6 mmHg *vs* LS: 222.8 ± 72.8 mmHg, *p=0.69)*, however the difference did not reach statistical significance (Figure 1). Also static compliance (C: 14.2 ± 3.8 mL/cmH_2O *vs* LS: 14.9 ± 3.6 mL/cmH_2O, *p>0.999*; Figure 2), pCO2 (C: 91.1 ± 29.3 mmHg *vs* LS: 71.6 ± 38.1, *p=0.31*), respiratory rate (C: 35.6 ± 8.9 bpm *vs* LS: 32.3 ± 5.6 bpm, *p=0.59)*, and PA pressure (C: 29.8 ± 5.4 mmHg *vs* LS: 30.2 ± 12 mmHg, *p=0.6)* were not statistically different in the two study groups at the endpoint of the experiments. The wet to dry ratio, as indicator of pulmonary edema, from biopsies of the left dependent lobes were similar in both groups (C: 7.3±3.1 *vs* LS: 8.6 ± 0.85, *p=0.69*).

CONCLUSIONS

First, a complex reproducible in vivo experimental pre-clinical model of severe ARDS induced by two subsequent bronchoscopic instillations of low pH GJ was successfully established in mechanically ventilated pigs. Despite the severity of the lung injuries, the support provided by VV-ECMO allowed the maintenance of adequate gas exchange and stable hemodynamic parameters.

Secondly, in this model it was possible to investigate the efficacy of a therapeutic strategy consisting in lung lavage and surfactant replacement. In the intervention group this treatment resulted in a transient decrease in lung compliance and oxygenation immediately post-therapy, but was overall well tolerated. At the end of each experiment, the lung function parameters - PaO_2/FiO_2, pCO_2, respiratory rate and Csta - were slightly improved in the treatment group compared to control. However, the differences observed were not significant.

The analysis of the BAL fluid and tissue samples may provide more information about the effect of the treatment plan with saline lavage and surfactant replacement. Further investigations should study whether different timing and doses of the treatment strategy, including treatment with surfactant replacement only, may be more beneficial. Moreover, the effect of different mechanical ventilation approaches resulting in better alveolar recruitment, which could potentially improve the distribution of the surfactant to the injured areas of the lung, should also be investigated. Alternatively, it is possible that the treatment with saline lavage and surfactant replacement is not efficacious in this aspiration model of severe ARDS.

Table 1. Physiologic parameters of the two study groups at baseline, post-lung injury, and after initiation of VV-ECMO support before randomization. Data are expressed as mean ± SD.

	Controls (C)			Treatment (LS)		
	Baseline	Post-injury	VV-ECMO support	Baseline	Post-injury	VV-ECMO support
Weight (kg)	32.2 ± 1.7	NA	NA	33 ± 1.8	NA	NA
Hemoglobin (g/L)	102.8 ± 4	NA	NA	110.2 ± 5.8	NA	NA
GJ pH	1.68 ± 0.07	NA	NA	1.65 ± 0.05	NA	NA
MAP (mmHg)	68 ± 2	65 ± 8	67 ± 7	72 ± 17	66 ± 6	67 ± 7
HR (bpm)	111 ± 21	166 ± 17	123 ± 28	99 ± 26	144 ± 48	104 ± 32
Mean PAP (mmHg)	13 ± 3	25 ± 6	22 ± 4	13 ± 3	20 ± 7	20 ± 3
RR (bpm)	15 ± 1	28 ± 7	13 ± 2	18 ± 3	26 ± 7	17 ± 4
Vt (mL)	358 ± 18	200 ± 25	199 ± 12	349 ± 30	208 ± 40	198 ± 23
Vt (mL/kg)	11.1± 0.6	6.2 ±0.8	6.2 ± 0.6	10.6 ± 0.6	6.3 ± 1.1	6.0 ± 0.6
pH	7.42 ± 0.04	7.13 ± 0.05	7.41 ± 0.05	7.41 ± 0.01	7.11 ± 0.15	7.43 ± 0.04
PaCO2 (mmHg)	42 ± 3	85 ± 10	42 ± 6	45 ± 2	88 ± 25	39 ± 3
PaO_2/FiO_2 (mmHg)	487.9±15.9	62.8 ± 9.1	281 ± 91	490.4±24.6	60.9 ± 9.6	229 ± 22
Estimated blood loss during cannulation (mL)	12 ± 7.5	NA	NA	11 ± 4.2	NA	NA
ECMO blood flow (L/min)	1.25 ± 0.23	NA	NA	1.58 ± 0.21	NA	NA

Gj: gastric juice; MAP: mean arterial pressure; HR: heart rate; PAP: pulmonary arterial pressure; RR: respiratory rate; Vt: tidal volume; ECMO: extracorporeal membrane oxygenation.

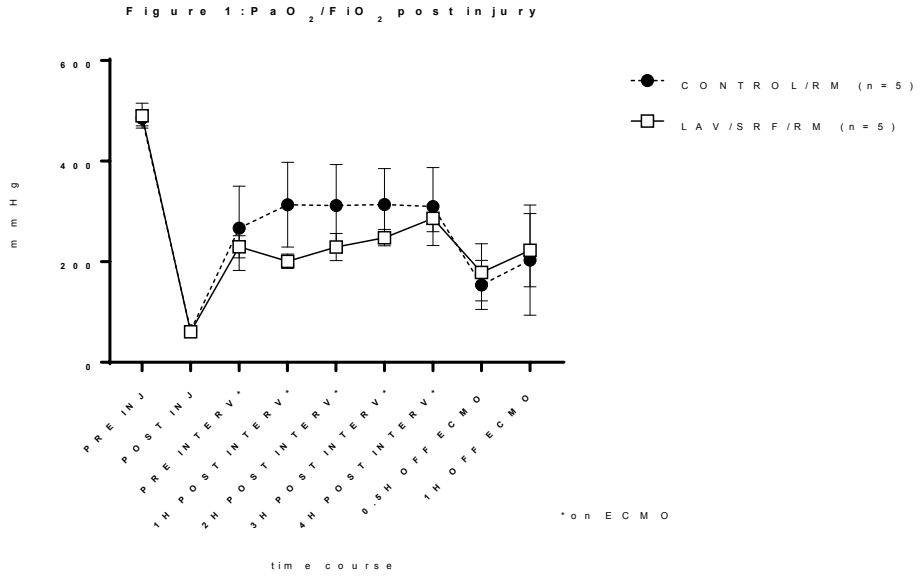

Figure 1: PaO_2/FiO_2 (mmHg) changes over time in controls (CONTROL/RM, n=5) versus treatment group (LAV/SRF/RM, n=5). The graph demonstrates the reduction in PaO_2/FiO_2 (mmHg) 1 hour post lavage/surfactant/RM and subsequent recovery by 4 hours after therapy. Data points represent mean values ± standard deviation.

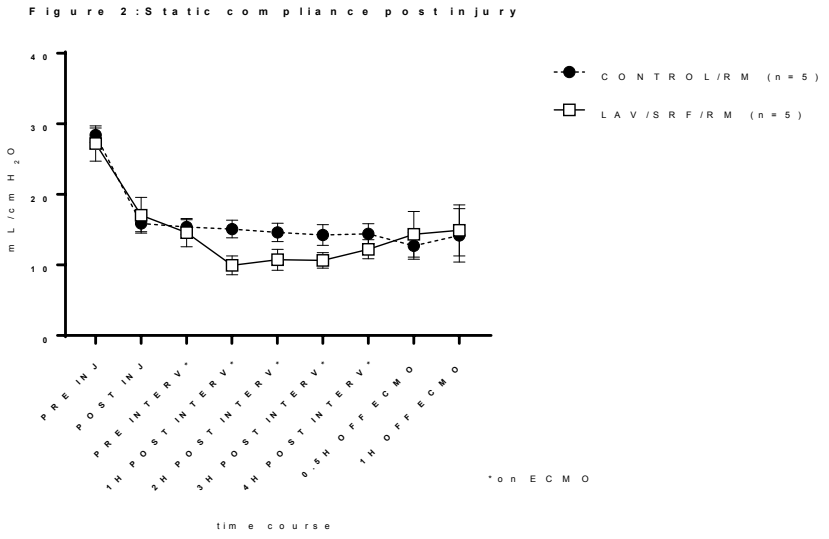

Figure 2: Static compliance, (Csta, mL/cmH_2O) changes over time in controls (CONTROL/RM, n=5) versus treatment group (LAV/SRF/RM, n=5). It is shown the reduction in Csta 1 hour post lavage/surfactant/RM and subsequent recovery by 4 hours after therapy. Data points represent mean values ± standard deviation.

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Author Contributions

RQ, YW, MG, CS, AA, MT and LDS participated in data acquisition and model development

AG provided operational assistance as the project's scientific manager

RQ, LDS, ML, SK and MC were involved in the conception and experimental design of the study

RQ, AG, LDS and MC participated in writing and review of the extended abstract

MC was the study supervisor