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Exogenous Surfactant Administration by Asymmetric High-Frequency Jet Ventilation in Experimental Respiratory Distress Syndrome

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Aim To evaluate the efficacy of surfactant administration using the instillation technique by means of asymmetric high-frequency jet ventilation (HFJV).

Methods Experiments were carried out on tracheotomized, anaesthetized, and paralyzed rabbits. Animals were lung-lavaged with saline and conventionally ventilated with 100% oxygen. After respiratory failure, they were ventilated for additional 2 hours with conventional ventilation (f = 30/min) or HFJV (f = 300/min). Subgroups were treated with porcine surfactant (Curosurf; 80 mg/mL, dose 200 mg/kg) as a bolus followed by conventional ventilation or instilled into the jet of the ventilator during asymmetric HFJV (inspiration time Ti = 30%). In controls, no material was instilled.

Results Bolus administration of Curosurf followed by conventional ventilation in comparison with conventionally ventilated animals without surfactant improved gas exchange (at 60 minutes: PaO_2/FiO_2, 9.8 ± 3.6 vs 22.3 ± 8.5 kPa, $P = 0.020$), reduced right-to-left pulmonary shunts (at 60 minutes: 47.8 ± 13.8 vs 23.9 ± 6.7 %, $P < 0.006$), and reduced inflammatory response in the lungs. Surfactant administration by HFJV in comparison with bolus instillation resulted in even better gas exchange (at 90 minutes: 20.7 ± 8.8 vs 39.1 ± 10.9 kPa, $P = 0.005$) and reduction of right-to-left pulmonary shunts (at 90 minutes: 29.1 ± 7.2 vs 8.0 ± 2.2 %, $P = 0.008$). High-frequency jet ventilation with or without surfactant treatment significantly increased pressure-volume recordings and reduced intraalveolar edema in comparison with conventional ventilation-only group. There were no significant differences in inflammatory response between the two tested methods.

Conclusion The response to surfactant therapy in experimental lung injury depends on the surfactant delivery method and may be potentiated by high-frequency jet ventilation.

Surfactant replacement has became a routine clinical practice in newborns with respiratory distress syndrome (1,2). Rapid tracheal instillation of liquid surfactant material represents the most common route of exogenous surfactant application. However, other surfactant delivery techniques have also been tested in clinical (3-6) and in experimental studies (7-10).

Surfactant therapy is usually combined with mechanical ventilation. High-frequency ventilation (HFV) is used in the management of newborns with respiratory failure, who are unresponsive to conventional ventilation. It generates tidal volumes that are smaller than dead space and relies on gas transport mechanisms related to the high frequency operation. By generating continuous distending pressure, high-frequency ventilatory modes help to keep the lungs open. The efficacy of surfactant treatment during such an open lung strategy is less dose-dependent and the con-

version of surfactant from large to small aggregates is reduced (11).

Among HFV techniques, high-frequency jet ventilation (HFJV) has also been used successfully to treat the patients with respiratory failure (12-14). Moreover, during HFJV, asymmetry in airflow can be achieved by changing the inspiration time (Ti). With Ti < 40%, airflow and moveable particles are directed toward the lungs (inpulsion effect). At Ti > 60%, airflow and materials are directed outside the lungs (expulsion effect, ref. 15). The expulsion effect of HFV was effective in the removal of bronchial secretions (16,17) or silica particles (18). It was also tested in meconium clearance (19,20), and found efficient in removal of meconium in combination with surfactant lavage (21). The opposite inpulsion effect of HFJV has not been previously tested for administration of substances into the lungs.

Based on data summarized above, using asymmetric HFJV would seem a rational approach for intratracheal instillation of exogenous surfactant. Therefore, the aim of our study was to test the efficacy of this novel surfactant delivery method in experimental respiratory distress syndrome directed at lung functions and morphology.

Material and Methods

Surfactant

Modified porcine surfactant (Curosurf®; Chiesi Pharmaceutici, Parma, Italy) was used at the concentration recommended for clinical use, 80 mg/mL, and the volume dose 2.5 mL/kg, corresponding to the dose of 200 mg/kg.

Experimental Design

The protocol followed the principles of laboratory animal care (NIH publication No. 86-23, revised 1985) and was approved by the Local Medical Ethics Committee. Twenty-eight young healthy rabbits (Chinchilla) of both sexes with mean ± standard deviation body weight (b.w.) of 1.3 ± 0.3 kg were used. Animals were anaesthetized with intramuscular 20 mg/kg ketamine (Narkamon, Spofa, Czech Republic) and 5 mg/kg xylazine (Rometar, Spofa, Czech Republic), and tracheotomized. Anesthesia was maintained by continuous infusion of ketamine (dose 20 mg/kg/h).

Catheters were inserted into the right femoral vein for infusions, into right atrium for mixed venous blood sampling and into right femoral artery for blood sampling and blood pressure monitoring. After that, the animals were paralyzed with pipecuronium bromide (Arduan, Gedeon Richter, Hungary; dose 0.3 mg/kg/30 min IV) and ventilated with a pressure-controlled ventilator (Chirana, Stará Turá, Slovakia) for 10 minutes, with a peak inspiratory pressure (PIP) of 6 cmH_2O, a frequency of 30/min, 60% inspiration time and FiO_2 of 0.21. No positive end-expiratory pressure (PEEP) was applied in this stage of the experiment. Blood gases, pH, hemoglobin, and lung function data obtained at this moment were regarded as the baseline. All animals were subjected to lung lavage according to Lachmann et al (22). At the beginning of the lavage procedure, PEEP was increased to 3 cmH_2O and FiO_2 to 1.0.

Twenty-five mL/kg b.w. of 0.9% saline warmed to 37 °C was instilled via tracheal cannula over the 15 seconds period. Lavage was repeated every 5 minutes until PaO_2 decreased below 12 kPa and lung-thorax compliance (C_{LT}) by > 30%. A 30-minute period after the last lavage was considered as a stabilization period. Animals were randomly allocated into four treatment groups. Two groups received no additional material and were ventilated by conventional ventilation or HFJV (CV and HF groups). Two other groups of animals were treated with Curosurf (80 mg/mL) at a dose of 200 mg/kg b.w. and ventilated by conventional ventilation or HFJV (CVS and HFS groups) for additional 120 minutes.

Ventilatory strategies were restricted by our protocol. During conventional ventilation, PIP was adjusted to keep the tidal volume (V_T) between 8 and 10 mL/kg b.w., PEEP was 3 cmH_2O, frequency 30/min, inspiration time 60% and FiO_2 1.0. During HFV, spontaneously generated PEEP was around 8 cmH_2O, frequency 300/min, inspiration time 50%, and FiO_2 1.0. Peak inspiratory pressure was kept at a comparable level during both types of ventilation (Table 1).

Arterial and venous blood samples were taken 30 minutes after the last lung lavage and 30, 60, 90, and 120 min during the treatment period.

At the end of the experiment, animals were killed by an overdose of anesthetics. Pulmonary pressure-volume characteristics were recorded and the lungs were fixed by vascular perfusion for histological evaluation.

Table 1. Pressures in respiratory system before and after the lung lavage, and at different time intervals in rabbits receiving conventional ventilation, conventional ventilation and surfactant, high-frequency ventilation, or high-frequency ventilation and surfactant

Parameter* and treatment group†	Finding (mean±standard deviation, cmH_2O) in relation to lavage time					
	before lavage	after lavage	30 min	60 min	90 min	120 min
PIP:						
CV	6.4±1.8	15.0±4.3	15.3±4.5	15.3±5.0	15.4±5.1	15.0±4.8
CVS	5.3±1.5	12.5±2.1	12.6±2.9	12.0±3.8	11.3±3.8	11.0±2.7
HF	5.9±1.4	12.5±0.6	13.8±1.6	13.8±1.1	13.3±1.4	12.8±1.7
HFS	4.7±1.1	13.7±2.1	14.9±2.4	14.9±2.4	14.6±2.2	14.2±2.9
PEEP:						
CV	0	2.3±0.1	3.1±0.1	3.3±0.2	3.6±0.2	3.3±0.2
CVS	0	2.3±0.2	2.8±0.2	2.7±0.2	2.8±0.3	2.2±0.2
HF	0	2.2±0.8	8.3±1.5	8.0±1.7	7.7±2.4	7.2±1.5
HFS	0	2.9±0.9	8.9±1.4	8.2±1.1	8.3±1.1	7.7±1.4
MAP:						
CV	1.9±0.5	6.1±1.9	6.8±2.0	6.9±2.1	7.1±2.3	6.7±1.7
CVS	1.6±0.5	5.4±2.1	5.8±2.6	5.5±2.6	5.3±1.9	4.7±1.2
HF	1.8±0.4	5.3±0.7	11.1±1.4	10.7±1.1	10.5±1.7	10.0±1.3
HFS	1.4±0.3	6.1±1.0	11.9±1.8	11.5±1.6	11.5±1.6	11.0±2.0

*Abbreviations: PIP – peak inspiratory pressure; PEEP – positive end-expiratory pressure; MAP – mean airway pressure; CV – conventional ventilation; CVS – conventional ventilation and surfactant; HF – high-frequency ventilation; HFS – high-frequency ventilation and surfactant.
†Within-group comparison: in all groups: PIP before lavage vs PIP after lavage, $P<0.029$. In all groups: MAP before lavage vs MAP after lavage, $P<0.036$. Between-group differences during the treatment period: PEEP. At all time points: CV and CVS vs HF and vs HFS (all $P<0.008$). MAP. At 30 and 60 min: CV and CVS vs HF and vs HFS (all $P<0.007$), at 90 and 120 minutes: CV<vs HF (both $P<0.017$), CV<vs HF, CVS vs HF and vs HFS (all $P<0.007$), ANOVA.

Surfactant Delivery

Protocol 1. Surfactant was rapidly instilled as a bolus into the airways while the animals were briefly disconnected from the ventilator and ventilated by conventional ventilation for the next 2 hours.

Protocol 2. Surfactant was instilled during asymmetric HFJV (f=300/min, Ti 30%) within 30 seconds via the jet of the ventilator without the interruption of mechanical ventilation. Asymmetric HFJV was used for additional 90 seconds. After that, Ti was adjusted to 50% and animals were ventilated for additional 2 hours by HFJV.

Lung Function Measurements

Airway pressure during conventional ventilation and HFJV was recorded via a catheter placed 0.5 cm below the distal tip of the tracheal cannula by means of a pressure transducer.

Tracheal airflow and tidal volumes were recorded with a Fleisch head connected to the pneumotachograph (ÚMMT SAV, Bratislava, Slovakia), placed temporarily between the tracheal cannula and the outlet of the ventilator circuit. Analog signals were recorded by multichannel recorder 6 NEK (RFT, Dresden, Germany). Mean airway pressure (MAP) was derived from the formula: $MAP = 0.5 \times (PIP\text{-}PEEP) \times (Ti/T) + PEEP$, where T is the duration of the respiratory cycle. The lung-thorax compliance (C_{LT}) during conventional ventilation was defined as the ratio between V_T adjusted for b.w. and the airway pressure gradient (PIP-PEEP). Airway pressures and V_T were recorded before and after the lung lavage, 30 min after allocation to treatment groups, and at 30 min intervals throughout the experiment.

Right-to-Left Pulmonary Shunt Calculation

Total right-to-left pulmonary shunts were calculated using a computer program by Fick equation: $(CcO_2 - CaO_2 / CcO_2 - CvO_2) \times 100$, where CcO_2, CaO_2, and CvO_2 are concentrations of oxygen in pulmonary capillaries, arterial, and mixed venous blood, respectively. CcO_2 was calculated using P_AO_2 (alveolar PO_2) from the equation: $P_AO_2 = (PB - PH_2O) \times FiO_2 - PaCO_2 \times [FiO_2 + (1\text{-}FiO_2) / R]$, where PB is barometric pressure and PH_2O the vapor pressure of water. Respiratory exchange ratio (R) was assumed to be 0.8 and hemoglobin necessary to calculate the oxygen concentration in the blood was measured by spectrophotometric method with Specol 11 (Carl Zeiss, Jena, Germany).

Static Lung Pressure-Volume Measurements

Pulmonary pressure-volume characteristics were recorded as described by Enhörning and Robertson (23). Airway pressure was increased from 0 to 40 cmH_2O by 5 cmH_2O increments and then reduced stepwise to 0 cmH_2O with 1 minute of equilibration at each pressure level. Lung volumes were corrected for air compression in the system and expressed per kg b.w. Volumes at deflation pressure 10 cmH_2O were used as a parameter of lung stability and expressed as percent of

maximum volume, defined as volume recorded at pressure 40 cmH_2O.

Histological Examination of the Lungs

The chest was opened, a thin catheter was inserted into the pulmonary artery, and the left ventricle was cut for drainage. The lungs were expanded for 60 seconds at a transpulmonary pressure of 35 cmH_2O. Pressure was then reduced to 10 cmH_2O, which was maintained for 30 minutes while the pulmonary arteries were perfused with 4% formaldehyde at a pressure of 65 cmH_2O. The lungs were further fixed by immersion in 4% formaldehyde and embedded in paraffin. The sections were stained with hematoxylin and eosin and with periodic acid-Schiff (PAS) stain. After randomization and coding, each section was examined blinded under a light microscope. The relative number of aerated alveoli was estimated according to a four-grade scale representing 0-25%, 26-50%, 51-75%, or 76-100% of the total alveolar spaces. The influx of inflammatory cells and the degree of edema were also graded semiquantitatively from 0 to 3 (0 – absent; 1 – slight; 2 – moderate; or 3 – severe). For all histopathological parameters, the five lung lobes from each animal were examined. A mean of the semiquantitative grading of the lung lobes was taken as a representative histopathological value for the corresponding rabbit to avoid intra-animal variation in grading (24).

Statistical Analysis

Data are expressed as mean ± SD or as median and range. The statistical software package SYSTAT 6.0.1 for Windows (SPSS Inc., Chicago, IL, USA) was used for data analysis. Between-group differences were examined by one-way analysis of variance (ANOVA), followed by Student-Neuman-Keuls *post hoc* test. Within-group differences before and after lung lavage were compared by two-way ANOVA for repeated measures. Differences in histological data (grades of alveolar expansion, intraalveolar inflammation and edema) were analysed by χ^2 test. A *P*-value < 0.05 was regarded statistically significant.

Results

General Observations

Twenty-eight rabbits were included in the study and 26 completed the experiments. Two rabbits died due to acidosis after the lung lavage before allocation to treatment groups and were excluded from the final data analysis. There were no differences in the body weight, blood gases and pH, number of lavages, and recovery of lavage fluid between the groups (Table 2).

Respiratory failure after the lung lavage was characterized by a significant decrease in both lung-thorax compliance (C_{LT}) and PaO_2/FiO_2 similar in all groups (Table 3).

PaO_2/FiO_2

Before the lung lavage, PaO_2/FiO_2 was about 45 kPa. After the lavage there was a significant drop in PaO_2/FiO_2 in all animals. In CV group, PaO_2/FiO_2 remained below 12 kPa for the rest of the experiment. After 30 minutes, values in HF and CVS groups significantly increased com-

Table 2. Survey of treatment groups, body weight of the animals, and baseline values of arterial partial pressure of oxygen (PaO_2), carbon dioxide ($PaCO_2$) and pH, number of lung lavages, and recovery of lavage fluid*

Treatment groups	Body weight (kg)	PaO_2 (kPa)	$PaCO_2$ (kPa)	pH	Lavage	
					No. of lavages	recovery (%)
Conventional ventilation (n=7)	1.20±0.23	9.2±2.2	3.1±0.6	7.45±0.10	7±2	91±4
Conventional ventilation and surfactant (n=6)	1.44±0.25	8.9±1.1	3.1±0.4	7.47±0.08	9±1	95±3
High-frequency ventilation (n=6)	1.38±0.31	9.1±0.8	3.1±0.5	7.48±0.09	8±2	93±2
High-frequency ventilation and surfactant (n=7)	1.37±0.27	9.5±2.0	3.3±0.5	7.48±0.05	8±1	93±3

*Data are expressed as mean±standard deviation. There were no statistically significant differences between the groups (ANOVA).

Table 3. Lung-thorax compliance (C_{LT}) and PaO_2/FiO_2 before and directly after the last lung lavage*

Treatment groups	Finding (mean±standard deviation) in relation to lavage time			
	C_{LT} ($mL \times kg^{-1} \times kPa^{-1}$)		PaO_2/FiO_2 (kPa)	
	before	after	before	after
Conventional ventilation	14.6±3.0	8.0±2.1	43.7±10.5	8.2±2.5
Conventional ventilation and surfactant	19.9±6.1	9.1±2.1	42.4±5.3	10.8±1.5
High-frequency ventilation	17.0±3.8	8.7±1.7	43.4±3.7	8.3±2.8
High-frequency ventilation and surfactant	20.6±4.1	9.4±2.4	45.2±9.8	8.5±2.8

*Within-group comparison: in all groups: C_{LT} after lavage vs C_{LT} before lavage, all $P<0.009$ and PaO_2/FiO_2 after lavage vs PaO_2/FiO_2 before lavage, all $P<0.002$ (ANOVA). There were no statistically significant differences between the groups (ANOVA).

pared with the CV group. Instillation of Curosurf by asymmetric HFJV led to further improvement of oxygenation in comparison with both CVS and HF-only groups. (Fig. 1).

$PaCO_2$

After the lung lavage, $PaCO_2$ significantly rose, from 3 kPa to 5 kPa. In CV group without surfactant it remained unchanged during the whole experiment. At 60 minutes, $PaCO_2$ was reduced in all other groups (CVS, HF, HFS) in comparison with the CV group. During the rest of the experiment, in the conventionally ventilated group with bolus administration of surfactant there was a gradual increase in $PaCO_2$ in comparison with both HFV groups (Fig. 2).

Right-to-Left Pulmonary Shunts

Before the lung lavage the right-to-left pulmonary shunts were about 12%. After the lung lavage right-to-left shunts significantly increased by more than 40% in all animals. In CV group the average value of this parameter remained around 50% for the rest of the experiment. At 30 min, values of right-to-left shunts were significantly reduced in all other groups (HF, CVS, HFS) in comparison with CV group. After 90 min, a further reduction in right-to-left shunts was also noticed in HF group in comparison with CVS, and in HFS group in comparison with HF and CVS groups (Fig. 3).

Ventilatory Pressures

Before and after the lavage procedure all animals were ventilated with comparable ventilatory pressures (Table 1). During the treatment period, there were no significant differences in PIP between the groups. The values of PEEP and those of MAP were significantly higher in both high-frequency ventilated groups than in CV animals due to spontaneously generated PEEP during HFJV (Table 1).

Pressure-volume recordings

Relative lung volumes at deflation pressure of 10 cmH_2O, expressed as percent of maximum volume, were larger in the HF-only group than in the CV-only group ($P=0.038$) and larger in HF animals with Curosurf in comparison with those with conventional ventilation without surfactant ($P=0.028$) (Fig. 4).

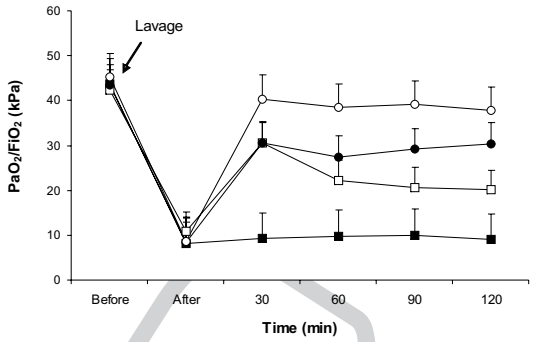

Figure 1. PaO_2/FiO_2 before and directly after lung lavage, and at different time intervals in various experimental groups: closed square – conventional ventilation (CV); open square – conventional ventilation and surfactant (CVS); closed circle – high-frequency ventilation (HF); open circle – high-frequency ventilation and surfactant (HFS). Values are given as mean±standard deviation. Within-group comparison: in all groups PaO_2/FiO_2 before lavage vs PaO_2/FiO_2 after lavage; $P<0.002$. Selected statistical analysis of between-group differences during the treatment period: At 30 min, CV vs all other groups (all $P<0.006$). At 60 min, CV vs CVS ($P=0.020$), CV vs HF ($P=0.004$), and CV vs HFS ($P=0.003$), HFS vs CVS ($P=0.005$), and HFS vs HF ($P=0.016$). At 90 min, HFS vs CV ($P=0.002$) and HFS vs CVS ($P=0.005$), HF vs CV ($P=0.003$). At 120 min, HFS vs CV ($P=0.008$) and HFS vs CVS ($P=0.021$), HF vs CV ($P=0.019$), ANOVA.

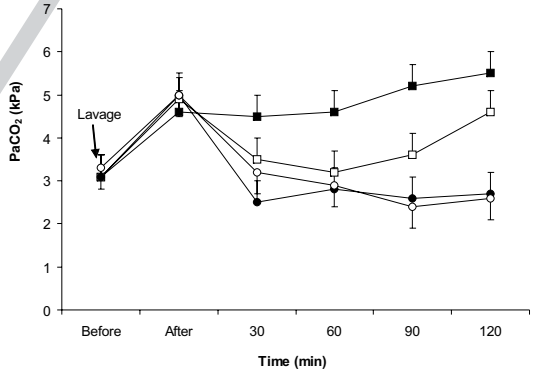

Figure 2. $PaCO_2$ before and directly after lung lavage, and at different time intervals in various experimental groups: closed square – conventional ventilation (CV); open square – conventional ventilation and surfactant (CVS); closed circle – high-frequency ventilation (HF); open circle – high-frequency ventilation and surfactant (HFS). Values are given as mean±standard deviation. Within-group comparison: in all groups $PaCO_2$ before lavage vs. $PaCO_2$ after lavage; $P<0.025$. Selected statistical analysis of between-group differences during the treatment period: At 60 min, CV vs all other groups (all $P<0.016$), at 120 min, CV vs CVS ($P<0.025$), CV vs HF ($P=0.009$) and CV vs HFS ($P=0.006$), CVS vs HF ($P=0.001$), and CVS vs HFS ($P=0.002$), ANOVA.

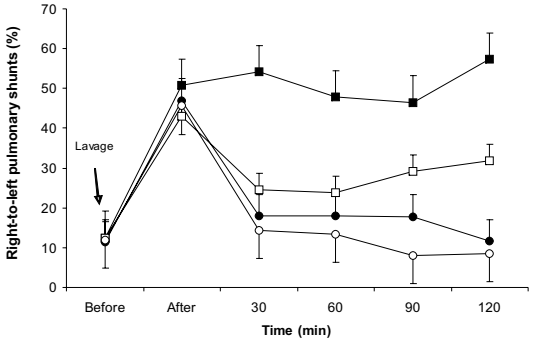

Figure 3. Right-to-left pulmonary shunts (RLS) before and directly after lung lavage, and at different time intervals in various experimental groups: closed square – conventional ventilation (CV); open square – conventional ventilation and surfactant (CVS); closed circle – high-frequency ventilation (HF); open circle – high-frequency ventilation and surfactant (HFS). Values are given as mean±SD. Within-group comparison: in all groups RLS before lavage vs RLS after lavage; $P<0.021$. Selected statistical analysis of between-group differences during the treatment period: At 30 min and 60 min CV vs all other groups (all $P<0.006$). At 90 min CV vs all other groups (all $P<0.002$), CVS vs HF ($P=0.019$) and CVS vs HFS ($P=0.008$), HF vs HFS ($P<0.021$). At 120 min CV vs all other groups (all $P<0.005$), CVS vs HF ($P=0.003$), and CVS vs HFS ($P=0.004$), ANOVA.

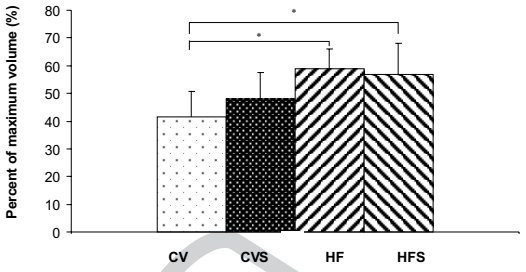

Figure 4. Static lung volume at the deflation pressure of 10 cmH_2O in various experimental groups: CV, conventional ventilation; CVS, conventional ventilation and surfactant; HF, high-frequency ventilation; HFS, high-frequency ventilation and surfactant. Values are expressed as percent of maximum volume, defined as volume recorded at pressure 40 cmH_2O. The bars represent mean values ±standard deviation. Significant between-group differences: HF>CV, *$P=0.038$ and HFS>CV, *$P=0.028$, ANOVA.

Histological Observations

Histological evidence of lung injury, defined as epithelial desquamation and necrosis, was found in all groups but was most prominent in animals subjected to conventional ventilation without surfactant. Hyaline membranes, graded as mild to moderate, were observed in a single animal in CV group. Slight to moderate intraalveolar accumulation of inflammatory cells (mostly neutrophils) was noted in most animals in CV and HF groups without surfactant (CV > CVS, $P=0.009$ and CV > HFS, $P=0.006$). The degree of intraalveolar edema was higher in the CV group without surfactant than in both groups ventilated by HFJV, with or without surfactant (CV > HF, $P=0.040$ and CV > HFS, $P=0.018$). Alveolar expansion was better in HF-ventilated animals receiving surfactant than in conventionally ventilated controls without surfactant ($P=0.027$) (Table 4).

Discussion

In this study we tested the lung functions and morphology in rabbits with experimental respiratory distress syndrome that were treated with surfactant instilled over 30 s during asymmetric high-frequency jet ventilation (HFJV) or as a bolus followed by conventional ventilation.

We found that bolus administration of Curosurf followed by conventional ventilation compared with the CV group without surfactant improved gas exchange, reduced right-to-left pulmonary shunts (RLS) and inflammatory response in the lungs. Combination of surfactant delivery by inpulsion effect of HFJV in comparison with bolus instillation followed by conventional ventilation resulted in even better gas exchange and reduction of right-to-left pulmonary shunts. HFJV with or without surfactant treatment significantly increased the static lung volumes and reduced the de-

Table 4. Semiquantitative evaluation of alveolar expansion, intraalveolar inflammation, and edema in the treatment groups.

Treatment groups	No. of lung sections per group	Grade (median, range)		
		alveolar expansion	intraalveolar inflammation	intraalveolar edema
Conventional ventilation	35	1.2 (1-2)	1.2 (0.8-1.8)	0.4 (0-1)
Conventional ventilation and surfactant	30	2 (0.4-2.4)	0 (0-0.4)*	0.3 (0-1.2)
High-frequency ventilation	30	1.4 (1.2-2.2)	0.2 (0-2)	0 (0-0.2)†
High-frequency ventilation and surfactant	35	2.1 (1.6-2.6)‡	0 (0-0.4)*	0 (0-0)†

*CV>CVS ($P=0.009$) and CV>HFS ($P=0.006$), χ^2-test.
†CV>HF ($P=0.040$) and CV>HFS ($P=0.018$), χ^2-test.
‡HFS>CV ($P=0.027$), χ^2-test.

gree of intraalveolar edema compared to the CV group without surfactant.

Bolus delivery is a recommended method for exogenous surfactant administration (1), however, several modifications of this standard mode have been used. For instance, Zola et al (3) compared different methods requiring fractional doses with or without interruption of mechanical ventilation, and found them to be equally safe and effective. Hentschel et al (6) reported an improvement in static compliance only after 90 minutes in babies with slow (30-45 minutes) instillation. In animal studies, the infusion over 5 and 45 minutes produced uneven distribution and poor physiological response (8,25) and aerosolization of surfactant, a form of slow delivery gives controversial results (10,26).

In this study, 30 seconds for surfactant administration was chosen because it is slower than bolus delivery (10 s), and shorter than periods shown to be ineffective in experimental (8,25) and clinical studies (27). In lung-lavaged rats, 60-seconds instillation of surfactant had similar effects on gas exchange and pulmonary mechanics as bolus delivery (9). One-minute period was also tested in clinical studies, when full dose of surfactant was given via catheter introduced through a side hole of a special tracheal tube adaptor (28) or via dual lumen endotracheal tube (4).

In our study, HFJV alone was superior to conventional ventilation with or without surfactant treatment as it improved gas exchange and reduced pulmonary right-to-left shunts. Surfactant delivery by asymmetric HFJV (over 30 s) was superior to all other tested groups. However, to emphasize the sole beneficial influence of the new discussed method, the group with surfactant bolus administration followed by HFJV had to be included.

Superior effect of HFV is not a standard finding. For instance, whereas Quan et al (29) found more efficient gas exchange in HFJV group of saline-lavaged rabbits, in another study HFJV was not more effective than conventional ventilation (30). Usually, when low ventilatory pressures are not sufficient to establish the critical opening pressure needed to recruit alveoli HFJV offers no advantage over conventional ventilation (30).

The value of intrapulmonary right-to-left shunts (RLS) determines the level of ventilation/perfusion mismatch in the lungs, and the beneficial effect of surfactant administration on venous admixure is known (31). In our study, lung damage induced by saline lavage increased intrapulmonary RLS up to 40-50% which is a normal value in this animal model (32). Administration of surfactant as a bolus followed by conventional ventilation and HFJV with or without surfactant immediately led to a decrease in right-to-left pulmonary shunts when compared to non-treated conventionally ventilated controls. Further reduction of RLS occurred at 90 min in HF-only group in comparison with conventional ventilation with surfactant, and by HF group with surfactant when compared to all other groups.

We also found that animals with lavaged lungs and those treated with HFJV without surfactant had, in comparison with the CV-only group, a significant relative increase in lung volume at deflation pressure of 10 cmH_2O. This observation, which is in agreement with data from other studies (33,34), indicates improved lung stability in the high-frequency ventilated group. After the instillation of surfactant there was no additional increase in static lung volume during deflation.

In other studies, even the animals ventilated with HFJV are disconnected from the ventilator and receive the surfactant directly into the endotracheal tube. In our experiments, we used a novel surfactant delivery method through the jet of the ventilator during asymmetric HFJV, without the interruption of mechanical ventilation. We speculate that when surfactant is instilled relatively slowly (over 30 s) to the jet of the ventilator, the jet pulses applied with the frequency of 300 cycles per minute cause the dispersion of the surfactant suspension and inpulsion effect of HFJV (at Ti = 30%) applied for additional 90 s promotes the movement of surfactant droplets toward the lungs. When surfactant was given as aerosol during HFV, it improved lung function without improving distribution, but with less effect on blood pressure and cerebral blood flow when compared to bolus instillation (35). We cannot say if the improved lung function in asymmetric HFJV group with surfactant may be due to better distribution. Davis et al (36) found ventilation independence of the distribution in Calf Lung Surfactant Extract labeled with ^{99m}Tc after its tracheal instillation in surfactant-deficient piglets during conventional ventilation and by HFJV with comparable PEEP.

On the other hand, Anderson et al (37) reported that the surfactant distribution depended on the time to the liquid plug rupture, which depends on the ventilation rate. At lower frequencies (20 breaths/min), liquid was localized in the gravity-dependent region of the lungs. At higher frequencies (60 breaths/min), it coated the airways, providing more uniform liquid distribution.

There is an interaction between exogenous surfactant and its alveolar environment (7) depending also on the ventilatory mode. In this term, high-frequency ventilation strategies may be of great benefit. By increasing the lung volumes they increase alveolar surface area and might indirectly facilitate the delivery of the exogenous surfactant on the base of surfactant interfacial spreading phenomena (36). Moreover, the change in surface area is small during HFV and this could reduce ventilator-induced consumption of surfactant due to repeated over-compression of the alveolar surface (38). Further, the low V_T decreases the conversion of exogenous surfactant active forms (LA) into inactive forms (SA), and the higher PEEP recruits collapsed alveoli (39). High level of PEEP also decreases the accumulation of lung water and prevents alveolar protein leakage (11,40), and thus inhibition of exogenous surfactant. In our study, spontaneously generated PEEP as a result of both given frequency and PIP in HF ventilated groups resulted in a significantly lower degree of intraalveolar edema and contributed to superior physiologic response to exogenous surfactant. As shown in our pilot experiments, such a PEEP was not tolerated by conventionally ventilated animals.

In conclusion, administration of Curosurf over 30 s during asymmetric HFJV in comparison with bolus instillation followed by conventional ventilation improved gas exchange and reduced pulmonary right-to-left shunts in rabbits with respiratory failure. HFJV with or without surfactant treatment significantly improved static lung volumes and reduced intraalveolar edema in comparison with CV animals without surfactant. This method was as effective as the bolus delivery procedure in reduction of inflammatory response. Our data indicate that the response to surfactant therapy in acute lung injury depends on the mode of surfactant delivery, and it may be potentiated by HFJV.

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