Oxygen Consumption and Carbon Dioxide Production During Liquid Ventilation

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Liquid ventilation with perfluorocarbon (PFCV) has advantages over conventional gas ventilation (GV) in premature and lung-injured newborn animals. Indirect calorimetric measurement of both oxygen consumption (VO₂) and carbon dioxide production (VCO₂) during PFCV has not been previously performed. In addition, comparison to indirect calorimetric measurement of VO₂ and VCO₂ during GV has not been evaluated. Ten fasted normal cats weighing 2.6 to 3.9 kg were anesthetized with pentobarbital and pancuronium. Tracheostomy was performed. Gas exchange was measured across the native lung during GV and across the membrane lung of the liquid ventilator during PFCV. VO₂ was measured using a modification of a previously described, indirect, closed-circuit, volumetric technique. VCO₂ was analyzed by capnographic assay of the mixed-expired closed-circuit air. The VO₂/VCO₂ ratio (RQ) was calculated. There was no change in VO₂, VCO₂, or RQ during PFCV when compared with GV (VO₂: GV = 5.7 ± 0.3 mL/kg/min, PFCV = 5.6 ± 0.5 mL/kg/min [P = NS]; VCO₂: GV = 4.9 ± 1.1 mL/kg/min, PFCV = 4.8 ± 0.9 mL/kg/min [P = NS]; RQ: GV = 0.85 ± 0.21, PFCV = 0.86 ± 0.21 [P = NS]). During GV the PaO₂ was higher than during PFCV (PaO₂: GV = 335 ± 70 mm Hg, PFCV = 267 ± 83 mm Hg [P = .04]), as is expected because of the relative reduction in the inspiratory PO₂ of the perfluorocarbon during liquid ventilation. There was no significant change in the PaCO₂ (PaCO₂: GV = 37.3 ± 2.2 mm Hg, PFCV = 40.4 ± 5.3 mm Hg [P = NS] or the pH (pH: GV = 7.34 ± 0.04, PFCV = 7.35 ± 0.06 [P = NS]). This study demonstrates the efficacy of measuring VO₂ and VCO₂ during gas and liquid ventilation using an indirect calorimetric technique. The data demonstrate that VO₂ and VCO₂ do not change during liquid ventilation and that excellent gas exchange can be accomplished through PFCV.

INDEX WORDS: Liquid ventilation.

PULMONARY gas exchange utilizing perfluorocarbon liquid ventilation (PFCV) has been investigated over the past 25 years. Efficacy of gas exchange in normal animals has been established. In addition, numerous studies in newborn premature animals have demonstrated efficacy in providing reduction of surface tension at the air-liquid interface in the PFC-filled lung with associated improvement in compliance, improvement in ventilation/perfusion matching, and reduction in ventilator airway pressure requirements.

Although the goal of PFCV is to provide gas exchange, few studies have actually measured VO₂ and VCO₂ during liquid ventilation. Those that have evaluated gas exchange during PFCV indicate that there is a reduction in the metabolic rate, the etiology of which remains unclear. In these studies, VO₂ and/or VCO₂ during gas and liquid ventilation were compared in different animals, under different conditions, or by methods with potential for significant measurement inaccuracy. The purpose of this study, therefore, is to relate gas exchange in comparison during PFCV and gas ventilation (GV) in the same animals under the same conditions by an indirect, calorimetric technique which allows accurate measurement of VO₂ and VCO₂.

MATERIALS AND METHODS

Ten healthy cats weighing 2.6 to 3.9 kg were anesthetized with pentobarbital, 25 mg/kg. A midline neck incision was performed and the trachea as well as the right carotid artery and internal jugular vein were isolated. A tracheostomy tube was advanced into the trachea to a point above the carina and anchored in place. A jet ventilation endotracheal tube (Mallinckrodt, Inc, Argyle, NY) was utilized in order to allow measurement of carinal airway pressure. An 18-gauge catheter was placed in the carotid artery and a 4F...
Venous Oximetrix catheter (Oximetrix, Inc, Mountain View, CA) was advanced into the right atrium via the right internal jugular vein and anchored in place. Mechanical ventilation was instituted and pancuronium bromide, 0.1 mg/kg, was administered intravenously at this point and hourly thereafter. Subsequent anesthesia was administered in the form of pentobarbital (10 mg/kg/h) intravenous infusion in 5% dextrose/Ringer’s lactate solution at a maintenance rate of 4 ml/kg/h.

Measurement of VO₂ and VCO₂

A previously described device was adapted as a flow-limited, time-cycled ventilator which allowed measurement of VO₂ and VCO₂ during gas ventilation (Fig 1). This device consists of a Cobe occlusive roller pump (Cobe Cardiovascular, Inc, Arvada, CO), which induces continuous gas flow at a prescribed rate through a closed circuit. The roller pump, the endotracheal tube, and a pinch valve are connected in series. A controller box allows alteration in inspiratory (IT) and expiratory time (ET) through variation of the duration of pinch valve closure. This, along with variation in the gas flow rate, establishes the respiratory rate and TV.

Fig 1. A flow-limited, time-cycled device that provides ventilation while simultaneously measuring VO₂ and VCO₂ during GV. The control box allows variation in the frequency and duration of pinch valve closure. This, along with variation in the gas flow rate, establishes the respiratory rate and TV.

A correction factor (CF₀) for VO₂ and VCO₂ at standard temperature and pressure dry (STPD) was applied to all data obtained during gas ventilation and was derived from the following formula:

\[
CF₀ = \frac{[273°K/(273° + T)] + [(Atm - P_H₂O)/Atm]}
\]

where \( T \) = the room temperature for VO₂ measurements and the circuit temperature at the capnometer site for VCO₂ evaluation, \( Atm = \) atmospheric pressure, and \( P_H₂O = \) water vapor pressure.

VO₂ and VCO₂ measurements during liquid ventilation were assessed utilizing a similar apparatus as seen in Fig 2. The pinch valve and endotracheal tube are deleted and replaced by the 4.5 m² membrane lung (Avecor, Minneapolis, MN) in the closed circuit. The roller pump provides a continuous sweep flow through the membrane lung as gas exchange occurs between the PFC and the closed circuit across the silicone membrane. VO₂ and VCO₂ measurements were evaluated by similar capnometric and closed circuit, volumetric techniques as described above. However, a correction factor (CF₁), incorporating a factor for the effect of liquid vapor pressure (Pₜ₀) during PFCV upon gas exchange measurements, was applied to VO₂ and VCO₂ measurements:

\[
CF₁ = \frac{[273°K/(273° + T)] + (Atm - P_H₂O - Pₜ₀)/Atm}
\]

Gas Ventilation

GV was performed utilizing the ventilator depicted in Fig 1. Use of this ventilator allowed assessment of VO₂ and VCO₂ during GV. The ventilator settings included IT = 1 second, ET = 2 seconds, TV = 15 mL/kg/breath, rate of 20 breaths/min, and circuit FiO₂ = 1.0.

Liquid Ventilation

A flow-limited, time-cycled perfluorocarbon liquid ventilator was developed and used as seen in Fig 3. This ventilator consists of an adaptation of an extracorporeal life support circuit. The inspiratory and expiratory limbs are “Y’d” at the endotracheal connection.
Res PaCO₂ at onset of GV. Anesthesia was administered by continuous infusion of pentobarbital, 10 mg/kg, throughout each study.

Arterial blood gas data were assessed utilizing an ABL 30 Blood Gas Analyzer (Radiometer A/S, Copenhagen, Denmark). Blood oxygen saturation data were evaluated using an OSM 3 co-oximeter (Radiometer A/S).

Data Analysis

VO₂ and VCO₂ data collected during each of the 10-minute periods were averaged and ±SD determined for GV and PFCV for all animals. Arterial blood gas data collected from all animals after 1 hour of either GV or PFCV were averaged and ±SD determined. The VCO₂/VO₂ ratio (RQ) was calculated, averaged, and ±SD determined for GV and PFCV. The paired Student's t test was applied for statistical comparison of data.

Approval for this study was obtained from the committee on university laboratory animal medicine at the University of Michigan Medical Center and all animal care guidelines followed. Animals were euthanized at completion of the study.

RESULTS

As seen in Fig 4, there was no change in VO₂, VCO₂, or RO during PFCV when compared to GV (VO₂: GV = 5.7 ± 0.3 mL/kg/min, PFCV = 5.6 ± 0.5 mL/kg/min [P = NS]; VCO₂: GV = 4.9 ± 1.1 mL/kg/min, PFCV = 4.8 ± 0.9 mL/kg/min [P = NS]; RQ: GV = 0.85 ± 0.21, PFCV = 0.86 ± 0.21 [P = NS]).

Arterial blood gas data (Fig 5) after 1 hour of GV or liquid ventilation demonstrated that the PaO₂ was higher during GV than during PFCV (PaO₂: GV = 335 ± 70 mm Hg, PFCV = 267 ± 83 mm Hg [P = .04]). This is expected because of the relative reduction in the inspiratory partial pressure of oxygen in PFC [PiO₂] during liquid ventilation. There was no significant change in the PaCO₂ (PaCO₂: GV = 37.3 ± 2.2 mm Hg, PFCV = 40.4 ± 5.3 mm Hg [P = NS]) or the pH (pH: GV = 7.34 ± 0.04, PFCV = 7.35 ± 0.06 [P = NS] [Fig 6]).
There was no significant change in heart rate (HR) or mean systemic blood pressure (MAP) when GV and PFCV were compared (MAP: GV = 102 ± 20 mm Hg, PFCV = 101 ± 14 mm Hg [P = NS]; HR: GV = 129 ± 21 beats/min, PFCV = 147 ± 29 beats/min [P = NS]).

DISCUSSION

This study documents that oxygen consumption and carbon dioxide production remain unchanged during liquid ventilation when related in comparison to that during GV. In addition, provision of adequate gas exchange utilizing PFCV in normal animals is demonstrated.

The concept of liquid ventilation extends back to the early 1920s when the efficacy of saline as a means of altering the physiologic effects of the pulmonary gas-liquid interface was investigated. However, because of the limited solubility of oxygen in saline (approximately 2 mL O₂/dL at PO₂ = 700 mm Hg), adequate oxygenation could not be provided during saline liquid ventilation except under hyperbaric conditions. Interest in the pulmonary applications of perfluorocarbon liquids, which incorporated properties of low surface tension and high oxygen solubility, began in 1966. At that time, the ability of the perfluorocarbon FX-80 to provide adequate gas exchange in the spontaneously breathing mouse and cat was demonstrated. Since then, a variety of perfluorocarbons have been developed and used, with the most recent generation incorporating and enhancing such desirable properties as low surface tension, high gas solubility, low viscosity to decrease airway resistance, and relatively high vapor pressure so that the perfluorocarbon might evaporate from the airways after conversion from liquid to gas ventilation.

Subsequent studies have demonstrated improved survival, enhanced gas exchange, and increased compliance in premature newborn animals, full-term lambs with meconium aspiration, and in animals with experimentally induced lung injury. In addition, the first human trials of liquid ventilation in three neonates have demonstrated feasibility of gas exchange utilizing liquid ventilation.

Although the efficacy of gas exchange during liquid ventilation has been assessed in numerous studies, measurement of VO₂ and VCO₂ has rarely been evaluated. Among the studies that have investigated gas exchange during perfluorocarbon ventilation include those by Sivieri et al in 1981 and Harris et al in 1983. In the former, VO₂ in spontaneously gas-breathing animals was compared to that during liquid ventilation as measured by a spirometric, volume-loss technique similar to the method utilized in this study. VO₂ was found to decrease 18.6% after initiation of liquid ventilation. In the second study, Harris et al evaluated VO₂ and VCO₂ in animals undergoing liquid ventilation by measuring the oxygen tension in inspired (P₈O₂) and expired (PCO₂) PFC and the carbon dioxide tension in inspired (P₁CO₂) and expired (P₂CO₂) PFC and calculating gas exchange based on the following equations:

\[ \dot{V}O₂ = V × (P₈O₂ - (P₈O₂) × àO₂) \]
\[ \dot{V}CO₂ = V × (P₂CO₂ - (P₁CO₂) × àCO₂) \]

where àO₂ and àCO₂ are the solubility coefficients for oxygen and carbon dioxide, respectively, in PFC and V is the minute volume ventilation with PFC. The derived values of VO₂ and VCO₂ were then compared to values obtained in similar fashion from another group of animals during GV. VO₂ and VCO₂ were reduced in this study after application of liquid ventilation by 42% and 35%, respectively. Therefore, in both of these studies, gas exchange appeared to be reduced during liquid ventilation, although the under-
lying physiology accounting for this finding remained unclear.

In the current study, gas exchange during liquid ventilation remained unchanged in comparison to that noted during GV. We believe that our study differs from those of others for the following reasons:

1. VO₂ and VCO₂ were evaluated by methods which allowed accurate measurement of gas exchange. In the study by Harris et al., evaluation of gas exchange was based on measurements of oxygen and carbon dioxide tensions in inspired and expired perfluorocarbon in order to calculate VO₂ and VCO₂. The authors, in discussing their results, note that the calculations for VO₂ and VCO₂ are dependent on solubility coefficients for these gases in PFC and that a variety of differing solubility coefficients have been determined, which may induce up to a 20% to 30% variation depending on the coefficient used. In contrast, the present study used an indirect, closed-circuit calorimetric technique that allowed accurate measurement of VO₂ and VCO₂ without need for application of a gas solubility coefficient.

2. In the study by Sivieri et al., animals were spontaneously respiring during gas breathing, but were pharmacologically paralyzed and ventilated during liquid breathing. In normal animals, the resting energy expenditure from breathing is approximately 1% to 5% of overall oxygen consumption and is even greater during periods of hyperventilation. In addition, pharmacological paralysis may result in an 11% reduction in VO₂. Mechanical ventilation and pharmacological paralysis, therefore, could account for the reduction in oxygen consumption noted in the study by Sivieri et al. In contrast, in the present study, animals were pharmacologically paralyzed and mechanically ventilated when gas exchange measurements were performed during both GV and PFCV.

The accuracy of the closed-circuit, indirect calorimetric technique for evaluating VO₂ and VCO₂ has been previously documented. The efficacy of a modification of this technique for the measurement of gas exchange during liquid ventilation has been demonstrated in this study. A number of factors, however, may affect the metabolic rate or the measurement of VO₂ and VCO₂. For instance:

1. Gas exchange could potentially have been altered because of variation in the PaCO₂ and pH. The metabolic rate has been shown to increase with a rising pH and diminishing PaCO₂. This change in VO₂ has been demonstrated to increase approximately 7% for each 10 mm Hg decrease in PaCO₂. PaCO₂ values during this study were, therefore, maintained at +10 mm Hg from the initial PaCO₂.

2. The metabolic rate may be altered with temperature, level of sedation, and paralysis. Therefore, all of these were carefully controlled throughout this study.

3. Stability of the VCO₂ and PaCO₂ are required for accurate assessment of VO₂. However, VCO₂, as measured by the method used in this study, would not have been affected by the absolute PaCO₂ value as long as that value was relatively stable. A stable PaCO₂ (±5 mm Hg between gas exchange measurements) was ensured during all measurements of VCO₂. Therefore, any differences in PaCO₂ seen between gas and liquid ventilation would not have affected the measured VCO₂.

Excellent gas exchange capabilities were documented during perfluorocarbon ventilation. The slight, but significant, reduction in the PaO₂ during PFCV when compared to GV is expected because of the relative reduction in inspiratory partial pressure of oxygen in PFC (PiO₂(PFC)) during liquid ventilation (PiO₂(PFC) = 380 ± 102 mm Hg) in comparison to the PiO₂ of the closed circuit during GV (FiO₂ = 1.0, PiO₂ = approximately 700 mm Hg). If adequate time is allowed, the partial pressure of oxygen in PFC will equilibrate with the partial pressure of oxygen in the gas to which the PFC is exposed. Therefore, this reduction in PiO₂(PFC) and PaO₂ is a reflection of minor limitations in the liquid ventilator device utilized, rather than being inherent to the liquid ventilation technique itself.

REFERENCES

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Discussion

F.J. Rescorla (Indianapolis, IN): Liquid ventilation with the use of perfluorocarbons has been studied for the past 25 years. Recent advances in the development of these compounds takes advantage of the low surface tension, high gas solubility, and low viscosity allowing delivery of oxygen and elimination of CO2 in a low-pressure system. Animal models using liquid ventilation to treat meconium aspiration and other experimental lung injuries have noted increased compliance, enhanced gas exchange, and improved survival. Liquid ventilation has also been shown to extend the limits of viability of immature lambs presumably by eliminating the dependency on surfactant. The recent initial human studies have demonstrated the feasibility of the model in premature babies. However, the very viable nature of the initial subjects that being a pH of 6.77 to 7.22 and short term survival and 19 hours at the maximum prevents determination of therapeutic benefits. Dr Hirschl and his colleagues have made a significant contribution to the field of liquid ventilation. They have demonstrated the efficacy of measuring oxygen consumption and carbon dioxide production during liquid ventilation using an indirect calorimetric technique. They have shown that oxygen consumption and CO2 production do not change during liquid ventilation and that excellent gas exchange can be accomplished due to fluorocarbon ventilation. My questions are as follows. In your model, the inspiratory partial pressure of oxygen in PFCV was 380 ± 102 mm Hg compared to approximately 700 mm Hg in the GV system. I understand from the manuscript that this is limited by your present system. Therefore, do you have plans to change your system to allow a higher partial pressure of oxygen in perfluorocarbon? Second, what do you foresee as the clinical application of this therapy? Will it be limited initially to preterms as in the initial clinical trial? Will it increase the viability of the fetus to a lower gestational age? Could this modality be used in the older infants and children with acute insults such as inhalation injuries and severe respiratory distress of other causes? And, finally, how can you avoid the situation experienced by other new therapies in which only the sickest and probably unsalvageable children are offered the new therapy. This appears to be the situation in the initial clinical trial. What do you foresee as the future for this modality?

W.J. Chwals (Winston-Salem, NC): Hirschl, in as much as the difference between ventilation conventionally and PFCV may be due to a difference in muscle tone in terms of energy expenditure, is the fact that your model involved paralysis not likely to reduce any differences which may be seen in conventional ventilation versus the PFCV model? In previous studies, the ventilatory mode when conventional ventilation was used is seldom complemented by paralysis.
R.B. Hirsch: Yes, the PiO₂, the inspiratory partial pressure of oxygen, was decreased with the liquid ventilator. We have already made changes in this system. These include allowing continuous flow of perfluorocarbon through the membrane lung which will alter the PiO₂ of the perfluorocarbon and which should improve the arterial oxygenation. With regard to possible clinical applications, I think that there is potential for wide-spread use not just in premature newborns, but in other neonates, children, and adults. The key benefit of PFCV is the elimination of the alveolar air-fluid interface with a reduction in alveolar surface tension. That's true especially with surfactant deficiency which is often present in the setting of respiratory failure. In addition, in situations where the alveoli are filled with fluid, perfluorocarbons may replace that fluid, allowing gas exchange to take place. These advantages of PFCV in the setting of respiratory failure will likely make this new modality applicable to all age groups. With regard to fetal application, there is a lot of speculation. It goes all the way to the point where you could imagine a premature baby submerged in the perfluorocarbon with gas exchange, in some fashion, being accomplished via the perfluorocarbon. However, there is a point where the lung is not adequately developed and ventilation with perfluorocarbons would not be applicable below that level. PFCV is at a point where evaluation in nonmoribund newborns is required. Our plan is to try to implement and explore this new modality on ECMO because it provides a safe setting for doing so. In addition, we are performing studies right now to evaluate whether PFCV will actually improve gas exchange and pulmonary function in newborns on ECMO. The pulmonary opacification that we observe after onset of bypass may be due to atelectasis and may actually be improved by PFCV. In addition, lavage with perfluorocarbons may allow removal of meconium from the lungs in the setting of the meconium aspiration syndrome. This has been shown to be true in animal models, with associated improvement in pulmonary function. With regard to the use of paralysis in this study, a previous study was done in which spontaneously breathing animals were compared to mechanically liquid ventilated animals. As would be expected, an increase in oxygen consumption in the spontaneously breathing animals was noted. We chose, therefore, to investigate whether there was a primary increase in baseline oxygen consumption and carbon dioxide production from the modality itself. I think that the best way to do that was to maintain control of all factors of oxygen consumption, which included the use of paralysis for both groups.