Carbon dioxide added late in inspiration reduces ventilation-perfusion heterogeneity without causing respiratory acidosis

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Brogan, Thomas V., H. Thomas Robertson, Wayne J. E. Lamm, Jennifer E. Souders, and Erik R. Swenson. Carbon dioxide added late in inspiration reduces ventilation-perfusion heterogeneity without causing respiratory acidosis. J Appl Physiol 96: 1894–1898, 2004. First published December 5, 2003; 10.1152/japplphysiol.00160.2003.—We have shown previously that inspired CO2 (3–5%) improves ventilation-perfusion (V̇A/Q̇) matching but with the consequence of mild arterial hypercapnia and respiratory acidosis. We hypothesized that adding CO2 only late in inspiration to limit its effects to the conducting airways would enhance V̇A/Q̇ matching and improve oxygenation without arterial hypercapnia. CO2 was added in the latter half of inspiration in a volume aimed to reach a concentration of 5% in the conducting airways throughout the respiratory cycle. Ten mixed-breed dogs were anesthetized and, in a randomized order, ventilated with room air, 5% CO2 throughout inspiration, and CO2 added only to the latter half of inspiration. The multiple inert-gas elimination technique was used to assess V̇A/Q̇ heterogeneity. Late-inspired CO2 produced only very small changes in arterial pH (7.38 vs. 7.40) and arterial CO2 (40.6 vs. 39.4 Torr). Compared with baseline, late-inspired CO2 significantly improved arterial oxygenation (97.5 vs. 94.2 Torr), decreased the alveolar-arterial PO2 difference (10.4 vs. 15.7 Torr) and decreased the multiple inert-gas elimination technique-derived arterial-alveolar inert gas area difference, a global measurement of V̇A/Q̇ heterogeneity (0.36 vs. 0.22). These changes were equal to those with 5% CO2 throughout inspiration (arterial PO2, 102.5 Torr; alveolar-arterial PO2 difference, 10.1 Torr; and arterial-alveolar inert gas area difference, 0.21). In conclusion, we have established that the majority of the improvement in gas exchange efficiency with inspired CO2 can be achieved by limiting its application to the conducting airways and does not require systemic acidosis.

LOW CONCENTRATIONS (3–5%) of CO2 added during inspiration improve gas exchange and arterial oxygenation in normal lungs (8, 12). Our laboratory used the multiple inert-gas elimination technique (MIGET) to demonstrate that inspired CO2 improves arterial oxygenation by reducing ventilation-perfusion (V̇A/Q̇) heterogeneity (22). In further work, our group found that the influence of CO2 on V̇A/Q̇ matching was associated with decreased regional pulmonary blood flow heterogeneity, as assessed by injected fluorescent microspheres (4).

Numerous studies in whole lungs and isolated airways and blood vessels reveal that CO2 and acidosis have several effects that have the potential to improve V̇A/Q̇ matching (for review, see Ref. 23). They relax airway smooth muscle (3, 6, 7, 16, 20, 21, 27), whereas hypocapnic hyperventilation causes bronchoconstriction (12, 27). CO2 also increases collateral ventilation including dogs and humans, hypercapnic acidotic pulmonary vasoconstriction (1, 2, 13); in this way CO2 and acidosis act to match regional perfusion to changes in ventilation.

Our interest in studying CO2 limited to the airways by adding it late in inspiration was stimulated by two aims and an otherwise neglected finding of improved arterial PO2 (PaO2) occurring with pulses of CO2 added to different phases of inspiration in a control of ventilation investigation (18). Our hypothesis (and first aim) was that CO2 could be added to the inspired gas late in inspiration and could improve V̇A/Q̇ matching and do so without causing systemic respiratory acidosis. In giving CO2 in this manner, we wished to determine which of the sites of CO2 action (airways, vessels, and lung parenchyma) is most important. By adding CO2 late in inspiration, its effects could be largely restricted to the conducting airways, and effects of systemic respiratory acidosis contributing to better V̇A/Q̇ matching could be avoided. These questions arose in part in response to a finding in awake tracheotomized ponies that late pulses of CO2 in inspiration caused no change in ventilation, but yet a 4-Torr decrease in the alveolar-arterial PO2 difference [(A-a)PO2], suggesting improved V̇A/Q̇ matching (18).

METHODS

Animal instrumentation and support. The Animal Care Committee of the University of Washington School of Medicine, Seattle, Washington, approved all aspects of procurement, care, nutrition, experimental protocol, anesthetic use, and euthanasia in these experiments. Studies were performed on 10 healthy mixed breed dogs (weight range: 20–28 kg) in the supine position under barbiturate anesthesia (20–30 mg/kg iv sodium pentothal induction with supplemental doses, 1–2 mg/kg as needed every 30–60 min). The animals underwent intubation with a cuffed endotracheal tube. Femoral arterial and venous catheters were inserted, and a jugular 7–Fr thermodilution catheter was advanced into the pulmonary artery. Temperature was...
monitored by the pulmonary artery catheter. Ventilation was administered by a piston pump ventilator at a tidal volume of 12–13 ml/kg and a rate of 14–16 breaths per minute. Every 5 min, a single 30 ml/kg breath was provided to prevent atelectasis. Once an acceptable rate of ventilation was determined by arterial blood-gas analysis [arterial pH of 7.39–7.45 and arterial P\textsubscript{CO\textsubscript{2}} (P\textsubscript{aCO\textsubscript{2}}) of 35–40 Torr], no further changes in ventilator settings were made. After the experiment, the dogs were killed by an overdose of thiopental sodium.

**Hemodynamics and respiratory gas exchange measurements.** Pulmonary and systemic arterial pressures, airway pressures, and mixed expired CO\textsubscript{2} and O\textsubscript{2} fractions (F\textsubscript{ECO\textsubscript{2}} and F\textsubscript{EO\textsubscript{2}}, respectively) were continuously recorded on an eight-channel recorder (Hewlett-Packard, Palo Alto, CA). At specified intervals, pulmonary arterial occlusion pressure, thermodilution cardiac outputs, and mixed expired F\textsubscript{ECO\textsubscript{2}} and F\textsubscript{EO\textsubscript{2}} (sampled from a 3-liter mixing box) were determined. F\textsubscript{ECO\textsubscript{2}} and F\textsubscript{EO\textsubscript{2}} in expired gas were measured by a mass spectrometer (Perkin-Elmer medical gas analyzer MGA-1100, Norwalk, CT). Blood pH and partial pressures of O\textsubscript{2} and CO\textsubscript{2} in arterial and mixed venous blood were measured on a blood-gas analyzer (Radiometer BMS-3, Radiometer America, Westlake, OH) maintained at 37°C and corrected to the pulmonary artery temperature. O\textsubscript{2} consumption (V\textsubscript{O\textsubscript{2}}) and CO\textsubscript{2} production (V\textsubscript{CO\textsubscript{2}}) were calculated by analysis of expired gases using appropriate temperature, pressure, and water content corrections.

Respiratory gas exchange was assessed by analysis of arterial and mixed venous blood gases. In calculating (A–a)PO\textsubscript{2}, the complete alveolar gas equation that incorporates inspired CO\textsubscript{2} fraction (F\textsubscript{ICO\textsubscript{2}}) (25) was used.

The addition of CO\textsubscript{2} late in inspiration complicates the analysis of measured mixed expired CO\textsubscript{2} and O\textsubscript{2} for the calculations of V\textsubscript{O\textsubscript{2}} and V\textsubscript{CO\textsubscript{2}}. Because we know the tidal volume, the frequency, and the amount of 100% CO\textsubscript{2} added into the last half of inspiration, it is possible to calculate the mean weighted corrected inspired O\textsubscript{2} fraction (P\textsubscript{I O\textsubscript{2}}) and F\textsubscript{ICO\textsubscript{2}}, for purposes of calculating the V\textsubscript{O\textsubscript{2}} and V\textsubscript{CO\textsubscript{2}} under the conditions of adding a gas into only a portion of the inspirate

\[
P_{\text{CO}_2}^{(\text{corrected})} = \frac{(V_{\text{added gas}} \times F_{\text{CO}_2 \text{ added gas}}) + (V_t \times F_{\text{ICO}_2})}{(V_{\text{added gas}} + V_t)}
\]

\[
P_{\text{O}_2}^{(\text{corrected})} = \frac{(V_{\text{added gas}} \times F_{\text{O}_2 \text{ added gas}}) + (V_t \times F_{\text{I O}_2})}{(V_{\text{added gas}} + V_t)}
\]

where V\textsubscript{added gas} is the total volume of gas in the injected CO\textsubscript{2}, F\textsubscript{OCO\textsubscript{2}} (0.000) and F\textsubscript{ICO\textsubscript{2}} (0.209) are the inspired fractions of CO\textsubscript{2} and O\textsubscript{2} in the breath delivered by the ventilator, and F\textsubscript{OCO\textsubscript{2} added gas} (1.000) and F\textsubscript{ICO\textsubscript{2} added gas} (0.000) are the fractions of CO\textsubscript{2} and O\textsubscript{2} in the injected gas volume. On average these yielded a corrected F\textsubscript{ICO\textsubscript{2}} of 0.023 and a corrected F\textsubscript{I O\textsubscript{2}} of 0.209. However, in calculating alveolar PO\textsubscript{2} (P\textsubscript{AjO\textsubscript{2}}) and the (A–a)PO\textsubscript{2} from the alveolar gas equation, we assumed that none of the added CO\textsubscript{2} reached the alveolar space. Consequently we corrected the measured values for mixed expired O\textsubscript{2} and CO\textsubscript{2} with the following equations

\[
F_{\text{ICO}_2}^{(\text{corrected})} = \frac{V_{\text{added gas}} \times F_{\text{CO}_2 \text{ added gas}} + (V_t \times F_{\text{ICO}_2})}{(V_{\text{added gas}} + V_t)}
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F_{\text{I O}_2}^{(\text{corrected})} = \frac{V_{\text{added gas}} \times F_{\text{O}_2 \text{ added gas}} + (V_t \times F_{\text{I O}_2})}{(V_{\text{added gas}} + V_t)}
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**Inert gas exchange measurements.** \(V_{A/Q}\) relationships were determined by MIGET (28, 29) using a constant infusion of a dilute solution of six inert gases (sulfur hexafluoride, ethane, cyclopropane, halothane, diethyl ether, and acetone) dissolved in 5% dextrose solution as described by Wagner and coworkers (28, 29). The gas extraction method of Wagner et al. (28) was used to determine inert gas tensions and solubilities in blood samples. Inert gas indexes were calculated from the 50-compartment model of Wagner et al. (29). \(V_{A/Q}\) heterogeneity was also assessed by the arterial-alveolar difference area [(a–A)D] area, derived directly from the retention and excetration data (10). All inert gas data are the average of duplicate measurements.

**Experimental protocols.** Each dog underwent the same protocol, serving as its own control. After stabilization and a 20-min period of ventilation with room air, baseline hemodynamic (cardiac output, heart rate, systemic and pulmonary artery pressures), ventilation and respiratory gas exchange (arterial and mixed venous blood gases, minute ventilation, airway pressures, and V\textsubscript{O\textsubscript{2}} and V\textsubscript{CO\textsubscript{2}}, and inert gas exchange measurements (arterial and mixed venous blood gases, and mixed expired gas) were taken. After these measurements, the animal was ventilated with 5% CO\textsubscript{2} mixed into room air (as measured in the inspiratory gas by mass spectrometry), leading to an inspired O\textsubscript{2} of 19.9 ± 0.1%. After 20 min, all measurements were repeated, and then the animal was returned to room air ventilation. Next, the dogs received room air ventilation with a small quantity (4–15 ml) of CO\textsubscript{2} added to the last half of inspiration. This was done by smooth hand injection of the CO\textsubscript{2} over the full second half of the inspiratory phase. The order of the different CO\textsubscript{2} exposures was randomly determined by a coin toss.

**Statistical analysis.** All data are presented as means ± SD and were analyzed by repeated-measures ANOVA followed by Scheffe’s and Bonferroni/Dunn post hoc tests corrected for multiple comparisons of similar data sets and for comparisons over time. Differences between individual experimental points were analyzed by Wilcoxon’s signed-rank test.

**RESULTS**

Minute ventilation was fixed at 6.0 ± 1.5 l/min. The concentration of CO\textsubscript{2} during late CO\textsubscript{2} was 2.4 ± 0.8% of total tidal volume and 4.9 ± 0.06% during continuous inspired CO\textsubscript{2}. The F\textsubscript{ICO\textsubscript{2}} during continuous inspired CO\textsubscript{2} was 0.199 ± 0.006.

**Hemodynamic and ventilatory effects of room air, 5% CO\textsubscript{2} and late-inspired CO\textsubscript{2} are listed in Table 1.** Mean systemic blood pressure was unchanged by either CO\textsubscript{2} treatment, but mean pulmonary artery pressure increased significantly with 5% CO\textsubscript{2}. Cardiac output increased with inspired 5% CO\textsubscript{2}, approaching statistical significance (\(P = 0.06\)). Late-inspired CO\textsubscript{2} had no significant hemodynamic effects. Pulmonary artery occlusion pressure, peak airway pressure, hematocrit, and temperature did not significantly change in either experimental condition. For seven animals in which we collected expired gas for calculation of V\textsubscript{O\textsubscript{2}} and V\textsubscript{CO\textsubscript{2}}, there were no differences in V\textsubscript{O\textsubscript{2}} (room air, 0.16 l/min; late CO\textsubscript{2}, 0.16 l/min; 5% CO\textsubscript{2}, 0.15 l/min). For V\textsubscript{CO\textsubscript{2}}, there was no difference between room air and

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Values are means ± SD; \(n = 10\) dogs. SBP, mean systemic blood pressure; Ppa, mean pulmonary artery pressure; Q, cardiac output; PAOP, pulmonary artery occlusion pressure; PVR, pulmonary vascular resistance, Paw, peak airway pressure; *\(P < 0.05\) compared with baseline; †\(P < 0.01\) compared with baseline.
late CO₂ (0.15 vs. 0.16 l/min), but a small fall with 5% CO₂ to 0.12 l/min (P < 0.05).

Table 2 provides respiratory gas exchange data. Both late-inspired CO₂ and 5% CO₂ produced a significant improvement in PAO₂. A decrease in pH was significant in both experimental conditions, but late-inspired CO₂ only decreased pH from 7.40 to 7.38, a clinically insignificant change. Late-inspired CO₂ also had no significant effect on PAO₂, whereas continuous inspired 5% CO₂ significantly increased PAO₂. Late-inspired CO₂ improved mixed venous PO₂ by 1.4 Torr, whereas continuous inspired 5% CO₂ increased mixed venous PO₂ by 9.6 Torr. There were no differences in PAO₂ with either late-inspired CO₂ or 5% CO₂. However, given the fact that the FiO₂ with 5% CO₂ was 0.199, the PAO₂ of 113 Torr underestimates the improvement of PAO₂ with continuous 5% CO₂. Had the FiO₂ been held at 0.209, the comparable figure would be roughly 120 Torr. The (A-a)PO₂ decreased equally with late CO₂ and 5% CO₂, based on the seven dogs in which we had V̇O₂ and V̇CO₂ measurements to calculate the respiratory quotient.

Table 3 shows MIGET gas data for eight dogs, in which reliable inert gas data were obtained. Data from two dogs were excluded because of a high residual sum of squares (>10). (a-A)D area for the six inert gases, a global measure of VO₂/V̇Q heterogeneity, was equally and significantly reduced both with 5% CO₂ and late CO₂. The retention and excretion components of the inert gas (a-A)D area were reduced significantly with 5% CO₂ but failed to attain significance at the P < 0.05 level for late CO₂ (P = 0.11 and 0.07, respectively). 5% CO₂ reduced the log standard deviations of the perfusion distribution and ventilation distribution approaching significance with both (P = 0.08 and 0.07, respectively). Shunt fraction and inert gas dead space were not affected by either late-inspired CO₂ or 5% CO₂.

Table 4 provides the mean retention and excretion values for the six inert gases in all three experimental conditions. For each gas, except acetone, there were statistically significant increases in excretion with 5% CO₂. Otherwise there were no statistically significant differences in retention for either late CO₂ or 5% CO₂. However, inspection of Table 4 shows that for late CO₂ the mean excretion values for all the inert gases except acetone were increased, suggesting a partial dose-response effect intermediate to that of 5% CO₂.

Figure 1 plots the percentages of ventilation and perfusion across seven discrete V̇A/V̇Q ranges from shunt through dead space for the three conditions. These data show that both late CO₂ and 5% CO₂ cause a shift of ventilation out of high V̇A/V̇Q regions (V̇A/V̇Q = 10–100) into regions of midrange V̇A/V̇Q ratios (0.1–10).

**DISCUSSION**

The major finding of this study is that when CO₂ is added during the latter half of inspiration, limiting it to the conducting airways, there is improved arterial oxygenation, a reduction in the (a-a)PO₂, and less V̇A/V̇Q heterogeneity without the development of systemic respiratory acidosis. From our analysis of the inert gas data, it appears that the effect of inspired CO₂ (throughout inspiration and only during late inspiration) is largely to convert units of high V̇A/V̇Q ratio to more normal midrange values. The improvement in oxygenation with 5% CO₂ added during only the latter half of inspiration was roughly half that observed with 5% CO₂ throughout inspiration, but the improvement in V̇A/V̇Q matching was nearly equivalent. These findings demonstrate that local effects of CO₂ in the lung contribute most to improved V̇A/V̇Q matching, but that systemic respiratory acidosis with 5% CO₂ throughout inspiration further improves arterial oxygenation by changes in
for the final half of inspiration and hence similar in concentration to the continuous CO₂.

We studied dogs that were healthy by all outward appearances, but the inert gas data showed a greater degree of V/A/Q heterogeneity than normal. Some of the dogs were studied in the fall when kennel cough is prevalent, and this may have contributed to some of the greater dispersion in our baseline V/A/Q heterogeneity. Nevertheless, the effectiveness of inhaled CO₂ was evident regardless of the baseline state of gas exchange.

Effects of CO₂ on gas exchange. The nearly equal improvements in V/A/Q matching observed with late-inspired CO₂ compared with continuous CO₂ establish that the majority of the effect of inspired CO₂ can be attributed to local effects in the lung. Addition of CO₂ in the latter half of inspiration exposes the conducting airways to a near-constant CO₂ tension throughout both phases of breathing without changing alveolar PCO₂. The high diffusibility of CO₂ leads to a lower extra- and intracellular pH within the airway epithelium and surrounding smooth muscle than during breathing of CO₂-free gas. Intracellular acidosis directly relax bronchial smooth muscle (3, 6, 7, 16, 20, 21, 27). The bronchodilating effects of CO₂ may not be equal throughout the lung owing to differences in local ventilation and V/A/Q ratio. Thus areas of low ventilation and V/A/Q ratio may be intrinsically more bronchoconstricted and therefore could be more responsive to the dilating effect of a rise in local CO₂. In this manner, ventilation may be redistributed out of highly ventilated areas into poorly ventilated regions, with the net result that the ratio falls in high V/A/Q regions and rises in low V/A/Q regions. This is consistent with the changes in ventilation to V/A/Q ratio shown in Fig. 1. We have recent and direct evidence for this pattern of regional ventilation redistribution in dogs ventilated with 5% CO₂ studied with aerosolized 1-μm fluorescent microspheres (5).

Although late-inspired CO₂ would be expected to have only airway effects, the distribution of regional perfusion may also be altered. Our data in dogs with fluorescent microspheres show that inspired CO₂ produces a more homogeneous distribution of regional perfusion (4). Transairway diffusion of CO₂ to the accompanying pulmonary arterioles with hypacapnic acidotic pulmonary vasoconstriction and/or CO₂-mediated augmentation of hypoxic pulmonary vasconstriction offers an explanation for a late-inspired CO₂ effect on regional perfusion distribution despite that fact that inspired CO₂ only reaches the nonrespiratory bronchioles. Direct evidence for transairway gas exchange with accompanying pulmonary arterioles comes from human studies with H₂-sensitive platinum electrodes wedged into 2- to 3-mm pulmonary vessels (17, 20). The accompanying bronchi are sixth- to eighth-generation branches in the midportion of the conducting airway system (30). When subjects inhaled a single breath of H₂-rich gas, there was a within-breath response of the vascular electrode, in contrast to delayed signal consistent with alveolar H₂ uptake and return to the pulmonary artery via normal circulation when the electrode-tipped catheter was pulled back out of the wedged position (11, 19).

With CO₂ present in the entire breath, there is CO₂ uptake and systemic respiratory acidosis. Thus improvements in oxygenation and V/A/Q matching may not entirely result from local effects of CO₂ in the lung. Systemic hypercapnia can increase

blood-gas chemistry (Bohr effect), cardiac output, and O₂ extraction.

Critique of methods. Late CO₂ was added by manually injecting a small aliquot (4–15 ml) of 100% CO₂ gas to the latter half of inspiration by following a pneumograph tracing. Thus there likely were small variations in breath-to-breath delivery of the CO₂ with respect to its appearance during the inspiration. Nevertheless, we were successful in restricting the delivery of CO₂ to the conducting airways because we essentially had no uptake of the added CO₂. The 0.02 pH change and 1 Torr rise in PaCO₂ were trivial compared with the 0.12 pH unit and 15 Torr PaCO₂ change with 5% CO₂ throughout inspiration. The concentration of CO₂ during late CO₂ was 2.4% when calculated for the entire breath but would be 4.8%
PaO₂ in two ways, independent of any change in V̇A/Q heterogeneity. First, the Bohr effect with systemic hypercapnic acidosis will lead to a higher Po₂, in both arterial and venous blood, for the same degree of hemoglobin saturation and blood O₂ content (26). Second, an elevated mixed venous Po₂ from the Bohr effect and an increased venous O₂ content arising from a slightly greater cardiac output but unchanged VO₂ (Table 1) will lead to a higher PaO₂ for any degree of fixed V̇A/Q mismatch (9). Last, systemic hypercapnia may improve V̇A/Q matching via neural reflexes involving airways and pulmonary vessels (23) arising from the peripheral and central chemoreceptors, as is true for peripheral chemoreceptor-mediated reduction of hypoxic pulmonary vasoconstriction (32).

In conclusion, our data establish that increases in CO₂ limited to the conducting airways during inspiration improve gas exchange in the lungs of dogs because of a decrease in V̇A/Q heterogeneity. In normal lungs, an improvement in arterial oxygenation of only 4 Torr, and an equivalent drop in (A-a)Po₂, may seem a small effect, but it must be appreciated that, in health, gas exchange efficiency is already quite good and may not be much improved on. However, CO₂ addition into the latter half of inspiration in many states of lung dysfunction may yield more significant improvements when V̇A/Q mismatching is greater. Last, addition of CO₂ into the latter half of inspiration offers a potential therapeutic strategy to enhance gas exchange and suppress local inflammatory events in lung injury when patients cannot tolerate further additional respiratory acidosis with tidal volume reduction (lung-protective ventilation or permissive hypercapnia) or CO₂ added uniformly into the inspired gas (15, 17).

GRANTS

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REFERENCES