special communication

Hartmut Heller and Klaus-Dieter Schuster. Theta values for $^{16}$O$^{18}$O and $^{18}$O$_2$ related to respective pulmonary diffusing capacities

Department of Physiology, University of Bonn, 53115 Bonn, Germany

HARMUT HELLER AND KLAUS-DIETER SCHUSTER

Theta values for $^{16}$O$^{18}$O and $^{18}$O$_2$ related to respective pulmonary diffusing capacities. Am. J. Physiol. 274 (Regulatory Integrative Comp. Physiol. 43): R1496–R1499, 1998.—The single-breath diffusing capacities for singly and doubly $^{18}$O-labeled CO$_2$, DL$_{\text{C}^{16}{\text{O}}^{18}{\text{O}}}^\text{CO}_2$ and DL$_{\text{C}^{18}{\text{O}}^{2}}^\text{CO}_2$, as well as for NO, were determined in seven anesthetized rabbits to investigate whether the theoretically predicted ratio of specific blood uptake rates of both isotopic CO$_2$ species, $\theta_{\text{C}^{18}{\text{O}}^{2}}/\theta_{\text{C}^{16}{\text{O}}^{18}{\text{O}}}$ could be derived from the measured values of DL$_{\text{C}^{16}{\text{O}}^{18}{\text{O}}}^\text{CO}_2$ and DL$_{\text{C}^{18}{\text{O}}^{2}}^\text{CO}_2$. Data of DL were obtained by inflated the lungs with gas mixtures containing 0.35% $^{16}$O$^{18}$O or 0.8% C$^{18}$O$_2$ or 0.05% NO in nitrogen, with breath-holding periods of 0.05–0.5 s and 2–12 s for the CO$_2$ and NO tests, respectively. The author obtained a ratio of diffusing pressures of indicator gases within pulmonary capillary blood membranes by isotopic exchange reactions. The present work was undertaken to examine the premises of these interpretations in an animal study. We used the familiar double-reciprocal Roughton-Forster relationship (12) for data analysis

$$\frac{1}{DL} = \frac{1}{Dm} + \frac{1}{\theta \cdot Vc}$$

(1)

where DL is the overall pulmonary diffusing capacity for a test gas and the components Dm and $\theta \cdot Vc$ represent the true conductance of the alveolar-capillary membrane and the rate at which the RBC in 1 ml of pulmonary capillary blood will absorb the test gas (in $\mu$moles/min) related to the total pulmonary capillary blood volume (Vc), respectively.

If it is true that singly and doubly $^{18}$O-labeled CO$_2$ diffuse with very similar facility into pulmonary capillary blood ($Dm_{\text{C}^{16}{\text{O}}^{18}{\text{O}}} \approx Dm_{\text{C}^{18}{\text{O}}^{2}}^\text{CO}_2$) but that C$^{18}$O$_2$ is removed within the CO$_2$-bicarbonate interconversion with a twofold higher probability than C$^{16}$O$^{18}$O, then $\theta_{\text{C}^{18}{\text{O}}^{2}}/\theta_{\text{C}^{16}{\text{O}}^{18}{\text{O}}} \approx 1.9 \pm 0.2$ (mean ± SD), thus comparing reasonably with the predicted one. Therefore, our findings provide evidence that the greater value of DL$_{\text{C}^{16}{\text{O}}^{18}{\text{O}}}^\text{CO}_2$ is mainly due to the twofold higher probability (or theta value) for C$^{18}$O$_2$ than for C$^{16}$O$^{18}$O to disappear within red blood cells via isotopic exchange reactions. Artificially ventilated rabbits; oxygen-labeled carbon dioxide; single-breath method.

IN PREVIOUS STUDIES (13, 14), singly and doubly $^{18}$O-labeled CO$_2$, C$^{16}$O$^{18}$O and C$^{18}$O$_2$, were introduced to determine the pulmonary diffusing capacity for carbon dioxide in man. The major advantage of this approach is that the rapid dilution of $^{18}$O in the large water pool (55 M) by isotopic exchange between CO$_2$-bicarbonate and water limits the development of significant back pressures of indicator gases within pulmonary capillary blood. The author obtained a ratio of diffusing capacities of the human lung for C$^{16}$O$^{18}$O (DL$_{\text{C}^{16}{\text{O}}^{18}{\text{O}}}^\text{CO}_2$) and C$^{18}$O$_2$ (DL$_{\text{C}^{18}{\text{O}}^{2}}^\text{CO}_2$) of 1.28.

Because the oxygens in bicarbonate are symmetrical ($^\text{HC}^{16}{\text{O}}^{18}{\text{O}}$ from C$^{16}$O$^{18}$O and $^\text{HC}^{18}{\text{O}}^{18}{\text{O}}$ from C$^{18}$O$_2$), there is a one in three chance in the case of C$^{16}$O$^{18}$O but a two in three chance for C$^{18}$O$_2$ that the $^{18}$O label will be in the water pool because of the hydration-dehydration reactions of CO$_2$-bicarbonate interconversion that is catalyzed by carboxic anhydrase of red blood cells (RBC) and pulmonary tissue. Because of this twofold higher probability of label removal for C$^{18}$O$_2$ than for C$^{16}$O$^{18}$O, it was concluded (14) that the estimated difference between DL$_{\text{C}^{16}{\text{O}}^{18}{\text{O}}}^\text{CO}_2$ and DL$_{\text{C}^{18}{\text{O}}^{18}{\text{O}}}^\text{CO}_2$ is explainable by a higher kinetics of disappearance via isotopic exchange for C$^{18}$O$_2$ than for C$^{16}$O$^{18}$O. In addition, the author stated that, by supposing equal diffusion kinetics of both isotopic species and due to DL$_{\text{C}^{18}{\text{O}}^{18}{\text{O}}}^\text{NO} > DL_{\text{C}^{16}{\text{O}}^{18}{\text{O}}}^\text{NO}$, the value of DL$_{\text{C}^{18}{\text{O}}^{18}{\text{O}}}^\text{NO}$ can be considered to provide a lower limit of the true conductance of the alveolar-capillary membrane for CO$_2$.

We estimated the membrane diffusing capacity Dm$_{\text{C}^{16}{\text{O}}^{18}{\text{O}}}^\text{CO}_2$ by calculating Dm$_{\text{C}^{16}{\text{O}}^{18}{\text{O}}}^\text{CO}_2 = 12.3 \times$ pulmonary diffusing capacity of NO (DL$_{\text{NO}}$), falling back on the theoretical prediction that gases permeate lung tissue at rates proportional to their solubilities in the lung [estimated from the solubility constants in water: $\alpha_{\text{O}_2}/\alpha_{\text{NO}} = 15.2$ (1, 17)] and inversely proportional to the square roots of molecular weights (30/46 = 0.81). Because of the very rapid binding of NO to hemoglobin (4, 11), it is usually expected that alveolar-capillary transfer of NO is mainly limited by diffusion (2, 7, 10, 11), leading to $1/(\theta \cdot Vc) \rightarrow 0$ and DL$_{\text{NO}} = Dm_{\text{NO}}$. 

$\theta_{\text{C}^{18}{\text{O}}^{2}} = \frac{DL_{\text{C}^{18}{\text{O}}^{2}}^\text{CO}_2 \cdot (Dm_{\text{C}^{16}{\text{O}}^{18}{\text{O}}}^\text{CO}_2 - DL_{\text{C}^{16}{\text{O}}^{18}{\text{O}}}^\text{CO}_2)}{DL_{\text{C}^{18}{\text{O}}^{2}}^\text{CO}_2 \cdot (Dm_{\text{C}^{18}{\text{O}}^{2}}^\text{CO}_2 - DL_{\text{C}^{18}{\text{O}}^{2}}^\text{CO}_2)}$

(2)
We applied the single-breath method to artificially ventilated rabbits to determine values of $D_{L\text{C}^{16}\text{O}^{18}\text{O}}$, $D_{L\text{C}^{16}\text{O}_{2}}$, and $D_{L\text{NO}}$. If our assumptions are valid, the calculated ratio of specific blood uptake rates should compare reasonably with the theoretically predicted value of $\frac{\theta_{\text{C}^{18}\text{O}_{2}}}{\theta_{\text{C}^{16}\text{O}^{18}\text{O}}} = 2$.

**METHODS**

Single-breath maneuvers were performed on seven adult Chinchilla cross-breed rabbits weighing 2.8–3.7 kg under pentobarbital sodium anesthesia (20 mg·kg$^{-1}$·h$^{-1}$ iv). The animals were paralyzed by alcuronium (0.1 mg·kg$^{-1}$·h$^{-1}$ iv), endotracheally intubated (2.5–3.5 mm ID), and artificially ventilated with room air by a computerized ventilatory servo system. The animals were instrumented with carotid arterial and ear vein cannulas for blood gas analyses, determinations of the hemoglobin concentration of blood ([Hb]), and the administration of drugs.

Test gases. To avoid the formation of oxidation products of NO and to enable the comparison of DL values of the three test gases, three O$\text{2}$-free gas mixtures, containing 0.35% C$\text{16O}_{2}$, 0.8% C$\text{18O}_{2}$, or 0.05% NO in N$\text{2}$, were prepared and stored in gas-tight flexible aluminum bags. Pure NO was led through diluted KOH, subsequently collected with a KOH-containing syringe, and finally injected into the aluminum bags, which had been repeatedly washed out with N$\text{2}$. To avoid an artificial isotopic exchange with water, pure C$\text{16O}_{18}\text{O}$ or pure C$\text{18O}_{2}$ was dripped within a trap and led into the N$\text{2}$-containing aluminum bags.

Protocol of experiments. Before the series of single-breath experiments, pressure-volume curves were recorded. For this purpose, the lungs were inflated and deflated by specified volume steps and the airway pressure was measured during short breath-holds by a differential pressure transducer. The residual volume (VR) was set at the lung volume attained at $-20$ cmH$_{2}$O of airway pressure. It was calculated from the argon (Ar) washout produced by inflating the rabbit lungs with the Ar-free test gas mixtures (see Table 1). Anatomic and apparatus dead spaces were determined in separate expiration maneuvers for the three test gases and were used to calculate the effective inflation and deflation times (13).

In each animal, the series of C$\text{16O}_{18}\text{O}$, C$\text{18O}_{2}$, and NO experiments were performed in random order, starting the single-breath maneuvers from VR in each case. The respective times for inflation and deflation were set at 0.6 s for C$\text{16O}_{18}\text{O}$ and C$\text{18O}_{2}$, and at 0.8 s for NO. For the two isotopic CO$\text{2}$ species, experiments with breath-holding periods of 0.05, 0.10, 0.15, 0.20, and 0.50 s were used to calculate DL. Inflated and deflated times for inflation and deflation were set at 0.6 s for C$\text{16O}_{18}\text{O}$ and 0.26 mmHg. C$\text{16O}_{18}\text{O}$ as well as C$\text{18O}_{2}$ were removed by the Ar-free test gas mixtures (see Table 1). The NO single-breath maneuvers were performed with breath-holding periods of 2, 4, 6, 8, and 12 s. The lungs of the rabbits were inflated with 30–47 ml of the C$\text{16O}_{18}\text{O}$- or C$\text{18O}_{2}$-containing gas mixtures and with 30–55 ml of the NO-containing test gas. After breath holding, the total expired gas was sampled by deflating the lungs via a spiral stainless steel tube (3.5 mm ID, length 5 m). The gas stored within this tube was dried by freezing and was continuously sucked into the inlet system of a respiratory magnetic sector mass spectrometer (M3; Varian MAT, Bremen, Germany). As shown in Table 1, the ratio of inflation volumes for the CO$\text{2}$ and NO experiments was varied between 0.73 and 1.34 to examine the influence of changes in pulmonary capillary blood volume (and thus of Hb content) on the $\frac{\theta_{\text{C}^{18}\text{O}_{2}}}{\theta_{\text{C}^{16}\text{O}^{18}\text{O}}}$ determinations. Apart from this gas-sampling procedure, continuous recordings of alveolar partial pressures of O$\text{2}$ and C$\text{16O}_{2}$ (unlabeled CO$\text{2}$) by mass spectrometry were used to check ventilatory conditions.

Mass spectrometry. The mass spectrometer used was modified to measure also isotopic ratios (15). The relevant gases NO, O$\text{2}$, C$\text{16O}_{2}$, Ar, C$\text{16O}_{18}\text{O}$, and C$\text{18O}_{2}$ were recorded at two ion collectors and one double collector, which were set at the following mass-to-charge ratios (m/e): 30 (NO), 32 (O$\text{2}$), 44 (C$\text{16O}_{2}$), and 46 (C$\text{16O}_{18}\text{O}$). We determined C$\text{16O}_{2}$ at the first plate of the double collector, C$\text{16O}_{18}\text{O}$ at the second plate, and C$\text{18O}_{2}$ at the second plate of the same double collector (m/e = 48) by repeatedly changing the accelerating voltage (peak jump). In the same way, Ar (m/e = 40) was measured at the C$\text{16O}_{2}$-44-ion collector. To avoid drift errors and cross-talk effects (6, 11, 13), the dry sample gas was repeatedly compared with a reference gas that only differed in the content of test gases, and by subtracting the background of the mass peaks. The concentrations of C$\text{16O}_{18}\text{O}$ and C$\text{18O}_{2}$ were obtained in terms of the difference to natural abundance. The signal-to-noise ratios were 1,656:1 at 3,500 parts per million (ppm) C$\text{16O}_{18}\text{O}$, 1,905:1 at 8,000 ppm C$\text{16O}_{2}$, and 1,351:1 at 500 ppm NO.

Calculations for DL. We used the partial pressures of the test gases obtained from that end-tidal portion of the gas sample where the concentration of test gases remained unchanged. These values were processed by applying the DL calculations on the basis of a coupled system of three differential equations defining gas transfer during inflation, breath holding, and deflation, as previously described in detail (13).

Statistics. Averaged data are given as mean ± SD values. The comparison between the calculated ratios of specific blood uptake rates and the theoretically predicted value $\frac{\theta_{\text{C}^{18}\text{O}_{2}}}{\theta_{\text{C}^{16}\text{O}^{18}\text{O}}}$ = 2 was carried out using the Wilcoxon test (one-sample μ test). Multiple regression analysis was performed on $\theta_{\text{C}^{18}\text{O}_{2}}/\theta_{\text{C}^{16}\text{O}^{18}\text{O}}$ versus [Hb] and ratios of inflation volume.

**RESULTS**

Figure 1 shows the time course of alveolar-capillary transfer of test gases in a semilogarithmic plot of ratios of alveolar partial pressures at overall times t and zero (PA/PA$\text{0}$) related to the overall time period of experiments. PA$\text{0}$ values were derived by calculating the dilution of inspired test gases within the alveolar volume (1.5 mmHg < PA$\text{0}_{\text{C}^{16O}_{18}O}$ < 2.0 mmHg; 4 mmHg < PA$\text{0}_{\text{C}^{18O}_{2}}$ < 5 mmHg; 0.22 mmHg < PA$\text{0}_{\text{NO}}$ < 0.26 mmHg). C$\text{16O}_{18}\text{O}$ as well as C$\text{18O}_{2}$ were removed according to the following biexponential relationships, where t is time in seconds:

$$PA_{C^{16O}_{18}O}/PA_{0,C^{16O}_{18}O} = 0.995 \cdot e^{-3.4^t} + 0.005 \cdot e^{-0.02^t}$$

$$PA_{C^{18O}_{2}}/PA_{0,C^{18O}_{2}} = 0.995 \cdot e^{-4.6^t} + 0.0005 \cdot e^{-0.02^t}$$

### Table 1. Animal characteristics and experimental parameters

<table>
<thead>
<tr>
<th>Rabbit</th>
<th>Body Wt, kg</th>
<th>[Hb], g/l</th>
<th>VR, ml</th>
<th>Vo, ml</th>
<th>Vico$\text{2}$, ml</th>
<th>Vinc$\text{O}_{2}$, ml</th>
<th>Pa$\text{CO}_{2}$, mmHg</th>
<th>Pa$\text{NO}$, mmHg</th>
<th>$\theta_{\text{C}^{16}\text{O}<em>{18}}/\theta</em>{\text{C}^{16}\text{O}^{18}\text{O}}$</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>3.3</td>
<td>120</td>
<td>16.4</td>
<td>6.7</td>
<td>40</td>
<td>55</td>
<td>0.73</td>
<td></td>
<td></td>
</tr>
<tr>
<td>B</td>
<td>3.2</td>
<td>123</td>
<td>13.1</td>
<td>6.5</td>
<td>40</td>
<td>55</td>
<td>0.73</td>
<td></td>
<td></td>
</tr>
<tr>
<td>C</td>
<td>3.1</td>
<td>115</td>
<td>13.9</td>
<td>6.6</td>
<td>45</td>
<td>50</td>
<td>0.9</td>
<td></td>
<td></td>
</tr>
<tr>
<td>D</td>
<td>3.3</td>
<td>114</td>
<td>10.4</td>
<td>6.5</td>
<td>35</td>
<td>40</td>
<td>0.88</td>
<td></td>
<td></td>
</tr>
<tr>
<td>E</td>
<td>3.7</td>
<td>133</td>
<td>12.5</td>
<td>6.5</td>
<td>47</td>
<td>35</td>
<td>1.34</td>
<td></td>
<td></td>
</tr>
<tr>
<td>F</td>
<td>3.3</td>
<td>134</td>
<td>11.3</td>
<td>7.2</td>
<td>35</td>
<td>35</td>
<td>1.00</td>
<td></td>
<td></td>
</tr>
<tr>
<td>G</td>
<td>3.4</td>
<td>112</td>
<td>11.2</td>
<td>6.2</td>
<td>30</td>
<td>30</td>
<td>1.00</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

[Hb], hemoglobin concentration of blood; VR, residual volume; Vo, anatomic and apparatus dead space; Vico$\text{2}$, Vinc$\text{O}_{2}$, inflation volumes used for CO$\text{2}$ and for NO experiments, respectively.
but NO disappeared monoexponentially from alveolar space

\[ \frac{P_{A_{NO}}}{P_{A_{NO,0}}} = 0.93 \cdot e^{-0.56 \cdot t} \]  

(5)

During the initial phase (t < 3 s), the ratios of PA to PA0 of C16O18O and C18O2 were reduced to less than 0.01 and 0.001, respectively. The pulmonary diffusing capacities for both CO2 species were calculated from this fast component by subtracting the partial pressure of the remaining residues from the respective PAC16O18O values of the fast phase (13, 14). The smallest PAC16O18O values, measured during the slow phase (t > 3 s), came close to the level of natural abundance of C16O18O, whereas the smallest PAC18O2 values determined during the same phase of label removal were 20 times higher than the natural C18O2 abundance. By contrast to the biexponential kinetics, >99% of the inhaled NO disappeared from alveolar gas after a time period of 10 s.

The DL data of each rabbit are listed in Table 2. The overall mean ± SD values are DL-C16O18O = 9.9 ± 1.6 ml · mmHg^{-1} · min^{-1}, DL-C18O2 = 13.3 ± 2.1 ml · mmHg^{-1}.

Table 2. Single-breath diffusing capacities for C16O18O, C18O2, and NO as well as the corresponding ratio of specific blood uptake rates of both isotopic CO2 species

<table>
<thead>
<tr>
<th>Rabbit</th>
<th>Dl-C16O18o, ml·mmHg^{-1} · min^{-1}</th>
<th>Dl-C18O2, ml·mmHg^{-1} · min^{-1}</th>
<th>Dl-NO, ml·mmHg^{-1} · min^{-1}</th>
<th>( \frac{\theta_{C16O18O}}{\theta_{C18O2}} )</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>11.9 ± 0.4</td>
<td>16.6 ± 1.2</td>
<td>2.5 ± 0.1</td>
<td>1.9</td>
</tr>
<tr>
<td>B</td>
<td>10.9 ± 0.4</td>
<td>14.6 ± 0.2</td>
<td>2.0 ± 0.2</td>
<td>1.8</td>
</tr>
<tr>
<td>C</td>
<td>11.2 ± 0.9</td>
<td>14.2 ± 0.4</td>
<td>1.9 ± 0.2</td>
<td>1.7</td>
</tr>
<tr>
<td>D</td>
<td>7.7 ± 0.2</td>
<td>11.4 ± 1.0</td>
<td>1.6 ± 0.1</td>
<td>2.1</td>
</tr>
<tr>
<td>E</td>
<td>10.5 ± 0.4</td>
<td>14.2 ± 1.1</td>
<td>1.7 ± 0.2</td>
<td>2.1</td>
</tr>
<tr>
<td>F</td>
<td>8.3 ± 0.6</td>
<td>11.0 ± 1.1</td>
<td>1.4 ± 0.1</td>
<td>1.7</td>
</tr>
<tr>
<td>G</td>
<td>8.6 ± 0.3</td>
<td>11.2 ± 1.0</td>
<td>1.4 ± 0.1</td>
<td>1.9</td>
</tr>
<tr>
<td>( \overline{x} \pm SD )</td>
<td>9.9 ± 1.6</td>
<td>13.3 ± 2.1</td>
<td>1.8 ± 0.4</td>
<td>1.9 ± 0.2</td>
</tr>
</tbody>
</table>

When we applied data to DL-C16O18O and DL-NO, the calculation DL-C18O2 = 12.3 · DL-NO is also confirmed.

The objective of this study was to investigate whether the pulmonary diffusing capacities of the 18O-labeled stable isotopic species of carbon dioxide, DL-C18O2 and DL-C16O18O, reflect the corresponding ratio of specific blood uptake rates of both test gases, \( \frac{\theta_{C18O2}}{\theta_{C16O18O}} \). Our data provide evidence that the higher kinetics of disappearance from alveolar gas found for C18O2 compared with C16O18O is mainly due to the higher removal rate of C18O2 via the above-mentioned CO2-bicarbonate interconversion. This is based on the finding that the calculated ratio \( \frac{\theta_{C18O2}}{\theta_{C16O18O}} = 1.9 \pm 0.2 \) compares reasonably with its predicted value (2.0). Because Eq. 2 was applied to data of DL-C16O18O and DL-NO, calculation Dm-C16O18O = 12.3 · DL-NO is also confirmed.

The assumption that the pulmonary diffusing capacity of NO should provide a very close estimate of the true conductance of the alveolar-capillary membrane has already been used in recent studies (2, 7, 10, 11). It is mainly based on the observation that there is a very rapid binding of NO to Hb, the reaction of which is 280 times faster than that of CO (4). Therefore, it was hypothesized that NO that enters the RBC is almost instantly converted to nitrate and the fractional exchange of NO between arterial and alveolar blood is nearly 100%. This result is not significantly different from the theoretically predicted value of \( \frac{\theta_{C18O2}}{\theta_{C16O18O}} = 1.9 \pm 0.2 \) (Wilcoxon test, \( \alpha = 0.05 \)). Furthermore, there was no dependence of individual mean values of \( \theta_{C18O2}/\theta_{C16O18O} \) on the ratios of alveolar volume for the CO2 and NO experiments and the [Hb] of blood (correlation coefficient of multiple regression analysis: 0.503, \( n = 7, P < 0.6 \)).

**DISCUSSION**

The single-breath diffusing capacities for C16O18O, C18O2, and NO as well as the corresponding ratio of specific blood uptake rates of both isotopic CO2 species
a reliable value of pulmonary capillary blood volume in rabbits, 3 ml, we obtained $D_{NO} < 1.9$ ml·mmHg$^{-1}$·min$^{-1}$. Thus, by using the value of $D_{NO}$, the true NO conductance of the alveolar-capillary membrane is at worst, underestimated by 6%.

This evaluation is important to interpreting the deviation between the measured ratio $D_{C16O18O}/D_{NO} = 5.5$ and the theoretically predicted value of 12.3 that would apply if the alveolar-capillary transfer of $C^{16}O^{18}O$ were also predominantly diffusion limited $[1/(\theta_{C16O18O} \cdot Vc)]$. From the similarity of $D_{NO}$ to $D_{C18O2O}$ it can be derived that this deviation is mainly caused by a significant contribution of the specific $C^{16}O^{18}O$ blood uptake conductance $[1/(\theta_{C16O18O} \cdot Vc)]$ to the overall resistance to alveolar-capillary transfer of $C^{16}O^{18}O$ $(1/D_{C16O18O})$. By using the Roughton-Forster equation again and referring to $D_{C16O18O}$ = 12.3·$D_{NO}$, one obtains that the overall rate of disappearance of $C^{16}O^{18}O$ from alveolar space is limited by 55% because of the label removal within RBC. The corresponding value for $C^{18}O^{2}$ amounts to 40%.

The RBC act as a sink to remove $^{18}$O-labeled CO$_2$ via the CO$_2$-bicarbonate interconversion, and RBC also act as a sink to very rapidly bind NO to Hb. Therefore, determinations of the single-breath diffusing capacities of both isotopic CO$_2$ species and NO might have been biased by using different inflation volumes during the CO$_2$ and NO tests, because pulmonary capillary Hb content changes because of various inflation volumes applied to artificially ventilated animals (11). Our finding that ratios of theta were independent of variations in inflation volume provides evidence that the specific blood uptake rates of test gases used are much too high to be influenced by such variations.

Perspectives

The present animal study revealed that the ratio of pulmonary diffusing capacities of the two $^{18}$O-labeled stable isotopic CO$_2$ molecules, $C^{16}O^{18}O$ and $C^{18}O^{2}$, effectively reflects the difference between the specific blood uptake rates for both isotopic CO$_2$ species, thus confirming the assumptions previously made (13, 14) with respect to interpreting the corresponding ratio of diffusing capacities of $C^{16}O^{18}O$ and $C^{18}O^{2}$ in man. By using the classical Roughton-Forster equation and an estimated value of the true conductance of the alveolar-capillary membrane for $C^{16}O^{18}O$, we were able to show that the mean blood uptake resistances of $C^{14}O^{18}O$ and $C^{18}O^{2}$ contribute to the overall resistance to alveolar-capillary gas transfer at levels of 55 and 40%, respectively. In reviewing the pertinent literature, the latter value (of $C^{18}O^{2}$) compares very closely with the corresponding mean value for CO (42% contribution by blood uptake resistance) that we calculated from studies in humans using 169 subjects (5, 8, 12, 16). Thus we found that the blood uptake resistances to $C^{18}O^{2}$ and CO equally contribute to the corresponding overall resistances to alveolar-capillary gas transfer.

The authors are very grateful for the valuable technical assistance provided by Christa Pusch, Barbara Schreiber, and Bernd Eixmann. Address for reprint requests: H. Heller, Dept. of Physiology, Univ. of Bonn, Nussallee 11, D-53115 Bonn, Germany.

Received 11 August 1997; accepted in final form 21 J anuary 1998.

REFERENCES