Combined effects of inspired oxygen, carbon dioxide, and carbon monoxide on oxygen transport and aerobic capacity

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Crocker GH, Toth B, Jones JH. Combined effects of inspired oxygen, carbon dioxide, and carbon monoxide on oxygen transport and aerobic capacity. J Appl Physiol 115: 643–652, 2013. First published June 27, 2013; doi:10.1152/japplphysiol.01407.2012.—We hypothesized that breathing hypoxic, hypercapnic, and CO-containing gases together reduces maximal aerobic capacity (V\textsubscript{O\textsubscript{2max}}) as the sum of each gas’ individual effect on V\textsubscript{O\textsubscript{2max}}. To test this hypothesis, goats breathed combinations of inspired O\textsubscript{2} fraction (F\textsubscript{IO\textsubscript{2}}) of 0.06–0.21 and inspired CO\textsubscript{2} fraction of 0.00 or 0.05, with and without inspired CO that elevated carboxyhemoglobin fraction (F\textsubscript{HBCO}) to 0.02–0.45, while running on a treadmill at speeds eliciting V\textsubscript{O\textsubscript{2max}}. Individually, hypoxia and elevated F\textsubscript{HBCO} decreased fractional V\textsubscript{O\textsubscript{2max}} (F\textsubscript{V\textsubscript{O\textsubscript{2max}}}), fraction of a goat’s V\textsubscript{O\textsubscript{2max}} breathing air) in linear, dose-dependent manners; hypercapnia did not change V\textsubscript{O\textsubscript{2max}}. Concomitant hypoxia and elevated F\textsubscript{HBCO} decreased V\textsubscript{O\textsubscript{2max}} less than the individual gas effects summed, indicating their combined effects on V\textsubscript{O\textsubscript{2max}} are attenuated, fitting the following regression: F\textsubscript{V\textsubscript{O\textsubscript{2max}}} = 4.24 F\textsubscript{IO\textsubscript{2}} + 0.519 F\textsubscript{HBCO} – 8.22 (F\textsubscript{IO\textsubscript{2}} × F\textsubscript{HBCO}) + 0.117, (R\textsuperscript{2} = 0.965, P < 0.001). The F\textsubscript{V\textsubscript{O\textsubscript{2max}}} correlated highly with total cardiopulmonary O\textsubscript{2} delivery, not peripheral diffusing capacity, and with arterial O\textsubscript{2} concentration (Ca\textsubscript{O\textsubscript{2}}), not cardiac output. Hypoxia and elevated F\textsubscript{HBCO} decreased Ca\textsubscript{O\textsubscript{2}} by different mechanisms: hypoxia decreased arterial O\textsubscript{2} saturation (Sa\textsubscript{O\textsubscript{2}}), whereas elevated F\textsubscript{HBCO} decreased O\textsubscript{2} capacitance [concentration of hemoglobin (Hb) available to bind O\textsubscript{2} (Hb\textsubscript{avail})]. When breathing hypoxic gas (F\textsubscript{IO\textsubscript{2}}, 0.12), Ca\textsubscript{O\textsubscript{2}} did not change with increasing F\textsubscript{HBCO} up to 0.30 because higher Sa\textsubscript{O\textsubscript{2}} of Hb\textsubscript{avail} offset decrease [Hb\textsubscript{avail}] due to the following: 1) hyperventilation with hypoxia and/or elevated F\textsubscript{HBCO}; 2) increased Hb affinity for O\textsubscript{2} due to both Bohr and direct carboxyhemoglobin effects; and 3) the sigmoid relationship between O\textsubscript{2} saturation and partial pressure elevating Sa\textsubscript{O\textsubscript{2}} more with hypoxia than normoxia.

Glossary

Ca\textsubscript{O\textsubscript{2}} Arterial oxygen concentration
D\textsuperscript{\textsubscript{T}}O\textsubscript{2} Index of peripheral diffusing capacity (V\textsubscript{O\textsubscript{2max}}/mixed-venous Po\textsubscript{2})
F\textsubscript{HBCO} Carboxyhemoglobin fraction
F\textsubscript{ICO} Inspired CO fraction
F\textsubscript{ICO\textsubscript{2}} Inspired CO\textsubscript{2} fraction
F\textsubscript{IO\textsubscript{2}} Inspired O\textsubscript{2} fraction
F\textsubscript{X} Value of X expressed as a fraction of room-air value at V\textsubscript{O\textsubscript{2max}}
Hb Hemoglobin
Hb\textsubscript{avail} Hemoglobin available to bind O\textsubscript{2} (not CO-bound or methemoglobin)
HR Heart rate
LA Lactate
M\textsubscript{LA} Lactate accumulation rate
OEC O\textsubscript{2}-equilibrium curve
P\textsubscript{50} Half-saturation pressure for O\textsubscript{2} of Hb\textsubscript{avail}
P\textsubscript{ACO\textsubscript{2}} Arterial CO\textsubscript{2} partial pressure

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Table 1. Caprinometrics of goats used for this study

<table>
<thead>
<tr>
<th>Breed</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
<th>6</th>
<th>Mean</th>
<th>SD</th>
<th>CV</th>
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<tr>
<td>Age, mo</td>
<td>Alp</td>
<td>LaM</td>
<td>Tog</td>
<td>LaM</td>
<td>LaM</td>
<td>Alp</td>
<td>18</td>
<td>&lt;36</td>
<td>41</td>
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<tr>
<td>Mb, kg</td>
<td>41.8</td>
<td>46.4</td>
<td>84.3</td>
<td>70.5</td>
<td>58.7</td>
<td>50.0</td>
<td>61.7</td>
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<td>V\textsubscript{O2max}, m/s</td>
<td>5.5</td>
<td>4.3</td>
<td>3.3</td>
<td>3.6</td>
<td>3.6</td>
<td>3.5</td>
<td>5.2</td>
<td>4.6</td>
<td>1.0</td>
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<td>1.018</td>
<td>0.835</td>
<td>0.677</td>
<td>0.646</td>
<td>1.126</td>
<td>1.016</td>
<td>0.886</td>
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<td>RER</td>
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<td>1.00</td>
<td>1.02</td>
<td>1.03</td>
<td>1.03</td>
<td>0.94</td>
<td>1.03</td>
<td>0.06</td>
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<td>End-run (LA), m</td>
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<td>3.15</td>
<td>4.59</td>
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<td>3.34</td>
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<td>0.97</td>
<td>0.79</td>
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<td>6.91</td>
<td>5.27</td>
<td>6.77</td>
<td>4.97</td>
<td>9.78</td>
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<td>HR, beats/min</td>
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<td>236</td>
<td>238</td>
<td>230</td>
<td>253</td>
<td>235</td>
<td>234</td>
<td>14</td>
<td>0.060</td>
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<tr>
<td>SV, ml/kg</td>
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<td>1.34</td>
<td>1.71</td>
<td>1.30</td>
<td>1.89</td>
<td>1.89</td>
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<td>0.176</td>
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<td>[Hb], g/dl</td>
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<td>16.3</td>
<td>10.1</td>
<td>15.2</td>
<td>14.4</td>
<td>14.0</td>
<td>14.0</td>
<td>2.1</td>
<td>0.151</td>
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<td>[Hb\textsubscript{avail}], g/dl</td>
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<td>15.7</td>
<td>9.6</td>
<td>14.7</td>
<td>14.0</td>
<td>13.6</td>
<td>13.5</td>
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<td>C\textsubscript{AO2}, ml O\textsubscript{2}/dl</td>
<td>17.8</td>
<td>20.8</td>
<td>13.1</td>
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<td>17.1</td>
<td>16.9</td>
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<td>0.95</td>
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<td>0.916</td>
<td>0.875</td>
<td>0.894</td>
<td>0.929</td>
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<tr>
<td>Pa\textsubscript{O2}, Torr</td>
<td>97.3</td>
<td>92.2</td>
<td>104.3</td>
<td>85.3</td>
<td>79.3</td>
<td>79.1</td>
<td>89.6</td>
<td>10.2</td>
<td>0.114</td>
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<td>C\textsubscript{O2}, ml O\textsubscript{2}/dl</td>
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<td>4.9</td>
<td>3.1</td>
<td>5.7</td>
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<td>3.8</td>
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<td>Sv\textsubscript{O2}</td>
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<td>0.222</td>
<td>0.226</td>
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<td>0.156</td>
<td>0.171</td>
<td>0.204</td>
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<td>32.1</td>
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<td>24.6</td>
<td>26.3</td>
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<td>149</td>
<td>148</td>
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<td>149</td>
<td>149</td>
<td>149</td>
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<tr>
<td>A-s\textsubscript{AD}, Torr</td>
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<td>101.4</td>
<td>105.9</td>
<td>92.0</td>
<td>93.8</td>
<td>86.9</td>
<td>97.0</td>
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<td>0.074</td>
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<td>1.23</td>
<td>1.11</td>
<td>0.89</td>
<td>0.93</td>
<td>1.36</td>
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<td>1.13</td>
<td>0.19</td>
<td>0.166</td>
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<td>0.0293</td>
<td>0.0267</td>
<td>0.0201</td>
<td>0.0459</td>
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<td>0.309</td>
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<td>P\textsubscript{ACO2}, Torr</td>
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<td>42.5</td>
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<td>pH\textsubscript{a}</td>
<td>7.36</td>
<td>7.35</td>
<td>7.34</td>
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<td>7.28</td>
<td>7.32</td>
<td>0.04</td>
<td>0.005</td>
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<tr>
<td>VA, ml btps·s\textsuperscript{-1}·kg\textsuperscript{-1}</td>
<td>19.5</td>
<td>15.4</td>
<td>14.2</td>
<td>10.1</td>
<td>18.0</td>
<td>14.2</td>
<td>15.2</td>
<td>3.3</td>
<td>0.216</td>
</tr>
</tbody>
</table>

Goat breeds are Alpine (Alp), LaMancha (LaM), or Toggenburg (Tog). Age is at start of the study. Goat 2’s age was unknown, but <36 mo. Body mass (Mb) are mean values over the course of the experiments, but after initial training to achieve reproducible maximal aerobic capacity (V\textsubscript{O2max}). Values reported are mean, SD, and coefficient of variation (CV) are for all six goats combined. V\textsubscript{O2max} are room-air values. Mean values of all goats are referenced. Gas volumes are STPD, unless otherwise stated. Mean, SD, and coefficient of variation (CV) are for all six goats combined. V\textsubscript{O2max} is Speed eliciting maximal aerobic capacity; V\textsubscript{CO2}, rate of CO\textsubscript{2} production; RER, respiratory exchange ratio; end-run (LA), end-run lactate concentration; M\textsubscript{LAM}, lactate accumulation rate; Q, cardiac output; R, heart rate; SV, cardiac stroke volume; [Hb], hemoglobin concentration; [Hb\textsubscript{avail}], concentration of hemoglobin available to bind O\textsubscript{2}; C\textsubscript{AO2}, arterial O\textsubscript{2} concentration; P\textsubscript{ACO2}, arterial mixed-venous O\textsubscript{2} partial pressure; P\textsubscript{AO2}, inspired O\textsubscript{2} partial pressure; P\textsubscript{ACO2}, ideal arterial O\textsubscript{2} partial pressure; A-s\textsubscript{AD}, arterial arteriole-arterial O\textsubscript{2} partial pressure difference; T\textsubscript{O2max}, total cardiopulmonary O\textsubscript{2} delivery; D\textsubscript{O2}, index of peripheral diffusing capacity (V\textsubscript{O2max}/P\textsubscript{ACO2}); P\textsubscript{ACO2}, arterial CO\textsubscript{2} partial pressure; pH\textsubscript{a}, arterial pH; VA, alveolar ventilation.
0.2 mg/kg) and locally anesthetized with subcutaneous lidocaine (20 mg/ml) at the cannulation sites. Using sterile technique, we inserted one 20-gauge × 12-cm polyurethane catheter (Arrow ES-04150) percutaneously for downstream, placed both a 16-gauge × 16-cm polyurethane catheter (Arrow ES-04306) and 7-Fr × 10-cm polyurethane introducer (Arrow SI-09700) in the jugular vein. We flushed the catheters daily with heparinized saline and filled them overnight with heparin until they were removed following the fourth day of experiments. We injected 2.2 mg/kg cefiotfur Na (Naxcel) subcutaneously twice a day to prevent catheter sepsis and 1.0 mg/kg flunixin meglumine (Banamine) intravenously once a day following the experiment to reduce catheter-induced thrombophlebitis. The goats recovered for two or more weeks between experimental weeks, during which time they trained regularly to maintain their $V_{O2max}$.

Each morning before an experiment, we advanced a 6-Fr × 60-cm balloon catheter (Arrow AI-07125) through the introducer in the jugular vein and into the pulmonary artery to sample mixed-venous blood. We confirmed catheter placement by monitoring pressure at the tip of the catheter with an external pressure transducer (Statham P23Td). We passed a thermocouple encased in sterile PE (Physitemp Type T) through the 16-gauge catheter in the jugular vein to the level of the heart to measure blood temperature. We recorded blood temperature with a thermocouple thermometer (Omega DP-41) that we calibrated weekly with a NIST-traceable mercury thermometer.

**Respiratory measurements.** Each goat wore a gas-tight mask connected to a cross-current bias flow of gas into and out of which the goat breathed. The mask was affixed to a 5-cm-diameter PVC T-fitting in the bias flow line. We connected the upstream and downstream openings of the T-fitting to 2-m lengths of flexible wire-reinforced 5-cm-diameter PVC tubing that allowed the goats to move on the treadmill while running and through which a 19-kW turbine pulled a bias flow across the goat’s muzzle. Downstream of the mask and flexible PVC tubing was 7.5 m of rigid 5-cm diameter PVC pipe with two 90° elbows to promote gas mixing. Upstream of the flexible tubing affixed to the mask was 7.5 m of 7.5-cm-diameter rigid PVC pipe with three 90° bends. To create the desired $FiO2$ and $FiCO2$, we bled gases from compressed cylinders of $N2$ and/or CO2 into the upstream end of the mixing tube to dilute room air pulled into the system by the turbine. We maintained $FiO2$ of 0.21 during normoxic hypercapnic treatments ($FiCO2$ 0.05) by adding $O2$ into the upstream end of the mixing tube. We calculated flow through the system from the pressure difference created by a 2.5-cm inner diameter orifice plate in the downstream sample tube that was measured with a differential pressure transducer (Validyne DP-45).

Expired gas mixed with bias flow was continuously sampled through a 3-mm-diameter brass port downstream of the goat. A diaphragm pump (Ametek R-2) pulled gas at constant flow, set by a diaphragm pump (Ametek R-2) pulled gas at constant flow, set by a mass flow controller (Sable Systems CM-10) monitored $FICO2$. We bled CO into the mixing tube to alter $FICO2$ and thus the goat’s $FIBICO$. A hemoximeter (Radiometer Copenhagen OSM3 set to its algorithm for sheep/goat blood) measured $FIHbCO$ in 0.2-ml arterial blood samples drawn at 1- to 2-min intervals during CO exposures so $FICO2$ could be adjusted to maintain the desired $FIBICO$. Windaq hardware and software (DATAQ Windaq DI-720-USB A/D and Windaq Pro+, respectively) recorded outputs of all transducers to a PC for later analysis using Windaq Waveform Browser software (DATAQ).

The $O2$ and $CO2$ concentration differences in the inspired and expired sample lines were used to calculate $VO2$ and $CO2$ production ($VCO2$) by mass balance. We calibrated the indirect calorimeter by removing the goat from the mask, replacing its muzzle with a similarly shaped PE plug, and adding diluent $CO2$ or $N2$ through the plug. The $N2$ dilution technique for calibration of indirect calorimeters and associated calculations used for determining $VO2$ have been described in detail (14) and validated for use with hypoxic and hyperoxic gases (Birks EK, Ohmura H, Crocker GH, Jones JH, unpublished observations).

**Oxygen transport measurements.** We simultaneously sampled arterial and mixed-venous blood at each speed after $O2$ and $CO2$ concentrations in the expired gas reached steady state (~2.5 min). A hemoximeter measured and/or calculated total $Hb$ concentration ($[Hb]$), $FICO2$, methemoglobin fraction ($FIBmet$), oxyhemoglobin fraction ($FIBO2$), $O2$ saturation of the $Hb$ available to bind $O2$ (i.e., not CO-bound or methemoglobin; $SO2 = FIBO2 / (1 - FIBmet)$) and concentration of $O2$ in the blood. We calculated the [Hb] available to bind $O2$ ($[Hb]_{avdal}$) as $[Hb]_{avdal} = ([[Hb] \times (1 - FIBmet)] / (FIBO2 - FIBmet))$. A blood-gas analyzer (Radiometer ABL 705) measured $pH$, $CO2$ partial pressure, and $PO2$ in arterial and mixed-venous blood, and these measurements were temperature corrected to intracardiac blood temperature (40–42).

We calculated cardiac output ($Q$) using the Fick Principle and total cardiopulmonary $O2$ delivery ($TO2max$) as the product of $Q$ and arterial $O2$ concentration ($CaO2$). We measured heart rate (HR) from electrocardiograms recorded from amplified (Gould Brush 2200) signals from Ag-AgCl electrodes (Kendall Meditrace 535) affixed to the venral thoraxes of the goats. A surcingle held the electrodes tightly in place as the goats ran on the treadmill. However, the electrocardiogram were too noisy while the goats ran to accurately count $R$ waves, and logistics of blood sampling prevented calculating HR from blood pressure. Therefore, we used 15 consecutive $R$ waves immediately after the treadmill was abruptly stopped at the end of each running speed to measure HR. We calculated cardiac stroke volume as the quotient of $Q$ and HR.

We calculated ventilatory variables as follows: 1) alveolar ventilation ($V_{A}$) from $VC02$ and arterial $CO2$ partial pressure ($Paco2$) (33); 2) inspired $PO2$ as the product of $FiO2$ and dry gas pressure, the latter calculated from the goat’s blood temperature; and 3) ideal alveolar $PO2$ using the ideal alveolar gas equation (16) and assuming alveolar $CO2$ partial pressure equals temperature-corrected $PCO2$. We estimated an index of peripheral diffusing capacity ($D^{TO2}_{PAO2}$) as $V_{O2max}$/mixed-venous $PO2$ for comparison among $FiO2/FICO2/FIBICO$ combinations as have others when comparing normoxic and hypoxic $FiO2$ (34).

See the APPENDIX for validation of peripheral diffusion capacity calculations for the range of $FiO2/FICO2/FIBICO$ combinations used.

During the training phase, we measured $VO2$, HR, and end-run lactate concentration ($[LA]$) and plotted these variables against running speed for each goat while it breathed air. Based on these measurements, we identified each goat’s room-air $V_{O2max}$ (plateau in $VO2$ vs. running speed) and minimum $V_{O2max}$ Spd, as well as maximum $V_{O2max}$ Spd, as well as maximum HR and end-run $[LA]$ corresponding to room-air $V_{O2max}$.

However, we could not know $V_{O2max}$ or $V_{O2max}$ Spd a priori for each $FiO2/FICO2/FIBICO$ combination during the experimental phase. Therefore, for each gas combination, we measured HR at rest and submaximal speeds and then extrapolated HR to estimate the speed required to elicit maximal HR at that goat’s room-air $V_{O2max}$. We then ran the goat at that speed, measured HR, and sampled arterial blood serially at ~30-s intervals throughout the run. Samples were capped as they were collected and stored on ice until $[LA]$ was measured with a LA analyzer (YSI 2300 Stat Plus) immediately following the run. We calculated LA accumulation rate ($d[LA]/dt$) as the slope of the linear regression of $[LA]$ with respect to time (27, 29, 30, 39). If HR and $M[LA]$ were similar to room-air $V_{O2max}$ values, then we considered that the goal had run at $V_{O2max}$ for that gas combination. If not, we ran the...
goat again at a faster speed and measured $V_O$, HR, and $M_{LA}$ to determine whether they increased from the previous speed.

We used arterial and mixed-venous blood samples to quantify the effect of $F_{HbCO}$ on Hb affinity for $O_2$. The samples were collected while goats stood at rest or locomoted on the treadmill at speeds up to and at $V_{O2max}^{Sand}$ while breathing different $F_{O2}$ and with $F_{HbCO}$ of 0.02, 0.15, 0.30, and 0.45. We estimated the Bohr coefficient [Δlog(P$_{50}$/ΔpH, where P$_{50}$ is the half-saturation pressure of Hb$_{avai}$] to correct blood samples measured at 37.0°C by the blood-gas analyzer to pH 7.40 by iteratively fitting a linear regression to the 0.02 $F_{HbCO}$ samples that minimized their error variance. We then fit linear regressions to pH-corrected Hill plots (21) of blood samples with $S_{O2}$ of Hb$_{avai}$ between 0.2 and 0.8 for air at each $F_{HbCO}$ to estimate P$_{50}$ at each $F_{HbCO}$.

Statistics. Each goat did not run with every $F_{O2}/F_{CO2}/F_{HbCO}$ combination, creating an incomplete block design, but enabling us to evaluate a greater number of gas combinations. Therefore, to factor out individual goat variation from treatment effects, we compared each goat to itself by expressing O2-transport variables as fractions of each goat’s value at $V_{O2max}$ when breathing air. Data are presented for all variables as means ± SD of the goats that breathed each gas combination. For variables that were standardized as fractions of room-air $V_{O2max}$ values, there was no variance associated with room-air values: every goat’s value was 1.0. Therefore, coefficients of variation among goats breathing room air (Table 1) were used as the measure of variance within room-air values.

We determined relationships between two variables using least-squares linear regression where appropriate and Kendall’s robust line-fit method for nonparametric regression for data that were heteroscedastic, as determined with Bartlett’s homogeneity of variance test. One-way ANOVA determined significant differences in O2-transport variables among gas mixtures, and Holm-Šidák multiple comparisons tests determined differences from room-air values. One-way repeated-measures ANOVA determined if hypocapnia altered $V_{O2max}$ by comparing $V_{O2max}$ of five goats that breathed both 0.00 and 0.05 $F_{CO2}$ with 0.21 $F_{O2}$ and by comparing fractional $V_{O2max}$ ($F_{V_{O2max}}$) for six $F_{O2}/F_{HbCO}$ combinations for which goats breathed both 0.00 and 0.05 $F_{CO2}$.

We predicted how hypoxia and elevated $F_{HbCO}$ would decrease $F_{V_{O2max}}$ if they acted additively by summing the independent effects of hypoxia and elevated $F_{HbCO}$ on $F_{V_{O2max}}$ for each gas combination (additive effects). Forward stepwise regression fit to measured $F_{V_{O2max}}$ quantified effects of hypoxia and elevated $F_{HbCO}$ with an interaction term (interactive effects). One-way repeated-measures ANOVA tested for differences between measured $F_{V_{O2max}}$ described by the interactive effects model and $F_{V_{O2max}}$ predicted by the additive effects model. SigmaPlot 12 (Systat Software) performed all statistical tests with α = 0.05, except for Kendall’s robust line-fit method for nonparametric regression and Bartlett’s homogeneity of variance test, which were run using a spreadsheet algorithm.

RESULTS

Normoxic $V_{O2max}$, $V_{O2max}^{Sand}$, and values for O2-transport variables for each goat are in Table 1. These are the values to which fractional values (the ratio of the value for a goat compared with its room-air value) are referenced.

Effects of gases on $V_{O2max}$. Hypoxia decreased $F_{V_{O2max}}$ in a dose-dependent manner from 0.21 $F_{O2}$ down to 0.09 (Fig. 1A). No $V_{O2max}$ were measured for 0.06 $F_{O2}$ as, when breathing that gas mixture, the first goat studied collapsed when walking at the treadmill’s lowest speed (1.18 m/s). The other goats also showed signs of respiratory distress (e.g., dyspnea, attempting to remove the mask and vocalizing) while just standing on the treadmill when breathing 0.06 $F_{O2}$. When breathing air, elevated $F_{HbCO}$ also decreased $F_{V_{O2max}}$ in a dose-dependent manner (Fig. 1B).

We tested whether hypocapnia altered $V_{O2max}$ in five goats that breathed room air ($F_{O2}/F_{HbCO}$ 0.21/0.02, abbreviated 21/0), with both 0.00 and 0.05 $F_{CO2}$, which it did not ($F_{CO2}$ 0.00 and 0.05, 0.861 ± 0.209 and 0.816 ± 0.201 ml O$_2$ [STPD] ⋅ s$^{-1}$ ⋅ kg$^{-1}$, respectively, $P = 0.149$). We also compared $F_{V_{O2max}}$ between 0.00 and 0.05 $F_{CO2}$ for the six $F_{O2}/F_{HbCO}$ combinations for which goats breathed both 0.00 and 0.05 $F_{CO2}$: 21/0, 0.12/0.02 (12/0), 0.09/0.02 (9/0), 0.21/0.15 (21/15), 0.12/0.30 (12/30), and 0.12/0.45 (12/45). The $F_{V_{O2max}}$ did not differ for these gases when normocapnic or hypocapnic gases were breathed (for $F_{CO2}$ 0.00 and 0.05, 0.614 ± 0.251 and 0.629 ± 0.213, respectively, $P = 0.662$).

Because hypoxia did not affect $V_{O2max}$, we combined hypocapnic and normocapnic data for different $F_{O2}/F_{HbCO}$ combinations to assess how the combined effects of hypoxia and elevated $F_{HbCO}$ affect $V_{O2max}$. The additive effects model predicted $F_{V_{O2max}}$ = 4.03 $F_{O2} - 1.23 F_{HbCO}$ + 0.170 (lower surface in Fig. 2). The interactive effects model best fit the equation $F_{V_{O2max}}$ = 4.24 $F_{O2} + 0.519 F_{HbCO} - 8.22 (F_{O2} \times$
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Fig. 2. VO2max expressed as FV˙O2max, with combinations of 0.21–0.09 FIO2, 0.02–0.45 FHbCO, and FC02 of 0.00 (●) or 0.05 (○). Hypercapnic and normocapnic points with same FIO2/FHbCO are combined for regression because hypercapnia had no significant effect on FV˙O2max. Values are means for all goats that breathed that gas mixture (n = 3–6). Forward stepwise regression yielded the interactive effects model equation (upper surface, wide grid, “Interactive effects”): FV˙O2max = 4.24 FIO2 + 0.519 FHbCO + 8.22 (FIO2 × FHbCO) + 0.117 (R² = 0.965, P < 0.001). The additive effects model (lower surface, narrow grid, “Additive effects”) was generated by summing independent effects of FIO2 and FHbCO for each gas combination on FV˙O2max: FV˙O2max = (4.03 FIO2) – (1.23 FHbCO) + 0.170. The interactive effects model did not differ from measured FV˙O2max (P = 0.427); however, the additive effects model did (P = 0.007).

FHbCO) + 0.117, (R² = 0.965, P < 0.001; upper surface in Fig. 2). No differences were detected between the interactive effects model and measured FV˙O2max (P = 0.427), whereas, predictions of the additive effects model differed from measured FV˙O2max (P = 0.007), indicating the combined effects of FIO2 and FHbCO in reducing VO2max are attenuated, thus falsifying our original hypothesis that they would be additive.

Effects of gases on O2 delivery. We tested if different FIO2/FICO2/FHbCO combinations affect TO2max or D’TO2 differently. The FV˙O2max correlated with TO2max fraction (F TO2max) for all gas combinations, but not with D’TO2 fraction (FD TO2) (Fig. 3, A and B). Furthermore, FV˙O2max correlated with CaO2 fraction (FCaO2) for all gas combinations, but not with Q fraction (FDQ) (Fig. 3, C and D).

We expressed FCaO2 as a function of arterial saturation of Hbavail with O2 (SaO2) fraction (F SaO2) and Hbavail fraction (FHBavail) in Fig. 4 for all normocapnic FIO2/FHbCO combinations because CaO2 is the product of SaO2 and [Hbavail]. Both hypoxia and elevated FHbCO decrease CaO2, albeit by different mechanisms: hypoxia primarily decreases SaO2, whereas, FHbCO primarily decreases [Hbavail]. When combined with hypoxia (FIO2, 0.12), elevated FHbCO decreases FHBavail but increases FSaO2, resulting in FCaO2 remaining unchanged as FHbCO increases up to 0.30. With FHbCO > 0.30, FSaO2 cannot increase further to offset lower FHBavail because SaO2 is nearly maximized at 0.30 FHbCO. Therefore, FCaO2 decreases with FHbCO > 0.30 when breathing hypoxic gas.

Goats hypoventilated (PA CO2 52.7 ± 6.6 Torr) at VO2max when breathing air. The slope of the linear regression of PaCO2 vs. FCaO2 was greater than zero for all normocapnic FIO2/FHbCO combinations (Fig. 5A), indicating goats progressively hyperventilated with FIO2/FHbCO combinations that lowered CaO2 and, as a result, VO2max. Therefore, arterial pH increased with FIO2/FHbCO combinations that lowered FCaO2 (Fig. 5B), increasing Hb affinity for O2 due to the Bohr shift. For FIO2/FHbCO combinations that lowered CaO2, goats hyperventilated because VC02 fraction decreased proportionally with FCaO2 (and FV˙O2max) (Fig. 5C), whereas, VA fraction did not (Fig. 5D).

Target FHBCO of 0.02, 0.15, 0.30, and 0.45 resulted in measured FHBCO of 0.021 ± 0.004, 0.157 ± 0.015, 0.303 ± 0.021, and 0.460 ± 0.029, respectively. To determine how elevated FHBCO affects Hb affinity for O2, we determined that...
a Bohr coefficient of $-0.54$ minimized error variance around the Hill plot for 0.02 FHbCO. The P50 progressively decreased with increasing FHbCO such that target FHbCO of 0.02, 0.15, 0.30, and 0.45 resulted in P50 of 28.8, 23.0, 18.9, and 14.5 Torr, respectively (Fig. 6), and fit best the equation: $P_{50} = 32.0 F_{HbCO} + 28.8$ ($R^2 = 0.989$, $P = 0.006$).

**DISCUSSION**

**Summary of findings.** The present study showed that hypoxia and elevated FHbCO individually decrease VO2max in dose-dependent manners, and their effects are attenuated in combination. These findings falsify the hypothesis that combined effects of these gases lower VO2max by the sum of their individual effects. The attenuated decrease in VO2max with both gases combined results from hyperventilation with hypoxia and/or elevated FHbCO and increased Hb affinity for O2 due to direct effects of FHbCO on the O2-equilibrium curve (OEC) and the Bohr shift. Both hyperventilation and increased Hb affinity for O2 affect SO2 more with hypoxia than normoxia due to the sigmoid shape of the OEC affecting SO2 at low and high PO2 differently. To our knowledge, this is the only study demonstrating the attenuated reduction in VO2max of combined hypoxia and elevated FHbCO and the mechanisms causing it.

**Hypoxia.** Hypoxia decreased VO2max in a dose-dependent manner (Fig. 1A), as has been documented in studies in humans (5, 8, 17, 26, 31, 43, 49). Hypoxia decreases arterial PO2 (Pao2) and SaO2, although their relationship is nonlinear due to the sigmoid relationship between SO2 and PO2 (17, 37). Low SaO2 decreased CaO2, To2max, and VO2max, a finding similar to data from humans (17). We were only able to obtain VO2max measurements down to 0.09 FIO2, because goats could not walk with 0.06 FIO2. When breathing 0.06 FIO2 at rest, mean inspired PO2 was 42 Torr, slightly less than 47 Torr measured in climbers near the summit of Mt. Everest (18).

**Carboxyhemoglobin.** The FV02max decreased with increasing FHBCO (Fig. 1B), similar to studies in humans (12, 25, 48). However, the human studies elicited FHBCO < 0.20 (48), which is less than one-half the highest FHBCO in the present study. The VO2max decreased with increasing FHBCO because CO binds to Hb with $\sim 207 \times$ the affinity of O2 at 37°C (35), raising FHBCO and lowering [Hbavail] and CaO2.

**Fig. 4.** Combined effects of FIO2 and FHbCO on fraction of arterial O2 saturation (FSaO2; ordinate), fraction of hemoglobin available to bind O2 (FHbavail; abscissa), and FCaO2 (isopleths, dotted curves) for goats breathing normocapnic gas mixtures while running at speeds eliciting VO2max. Shapes denote different FIO2 (0.21 circles, 0.15 inverted triangles, 0.12 triangles, 0.09 diamonds), and shades different FHbCO (0.02 solid, 0.15 dark shaded, 0.30 light shaded, 0.45 open). Values are means ± SD of goats that breathed that gas mixture ($n = 3–6$), except room-air SD are coefficients of variation between individuals. Different from room air: *FSaO2, †FHbavail, and ‡FCaO2. Solid symbols and solid line denote effect of FIO2 alone. Circles with different shades and long-dashed line denote effect of FHbCO alone. Triangles with different shades and short-dashed curve denote effect of elevated FHbCO with hypoxia (FIO2 0.12).

**Fig. 5.** The arterial CO2 partial pressure (PaCO2; A), arterial pH (pHa; B), fraction of rate of CO2 production (FVCO2; C), and fraction of alveolar ventilation (FV; D) vs. FCaO2 with different normocapnic FIO2/FHbCO combinations in goats running at speeds eliciting VO2max. Different shapes denote different FIO2 (0.21 circles, 0.15 inverted triangles, 0.12 triangles, 0.09 diamonds), and different shades different FHbCO (0.02 solid, 0.15 dark shaded, 0.30 light shaded, 0.45 open). Linear regressions are: for A, $pH = 7.469 F_{CaO_2} + 4.48$ ($R^2 = 0.929$, $P < 0.001$), for B, $pH = -0.258 F_{CaO_2} + 7.60$ ($R^2 = 0.739$, $P = 0.001$), and for C, $F_{VCO_2} = 1.16 F_{CaO_2} - 0.180$ ($R^2 = 0.898$, $P < 0.001$). For D, linear regression of $F_{V_A}$ vs. $F_{CaO_2}$ was not significant ($P = 0.058$).
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The $V_{O_{2}max}$ decreased with $F_{IO_{2}}/F_{ICO_{2}}/F_{HBO_{2}}$ combinations that lowered $T_{O_{2}max}$ due to reduced $CaO_{2}$, not $Q$ (Fig. 3, C and D). Some studies have found that $Q$ decreases with hypoxia (5, 31, 32) or elevated $F_{HBO_{2}}$ (12), whereas others have not (43, 48). Regardless of whether $Q$ decreases with combinations of $F_{IO_{2}}/F_{ICO_{2}}/F_{HBO_{2}}$ or not, any such decrease is small compared with that of $CaO_{2}$, the primary mechanism reducing $O_2$ delivery.

The $F_{V_{O_{2}max}}$ was highly correlated with $F_{CaO_{2}}$ for all $F_{IO_{2}}/F_{ICO_{2}}/F_{HBO_{2}}$ combinations (Fig. 3D), yet $CaO_{2}$ decreased due to different mechanisms with hypoxia vs. elevated $F_{HBO_{2}}$ (Fig. 4). Hypoxia reduced $CaO_{2}$ primarily due to decreased $SaO_{2}$. In contrast, elevated $F_{HBO_{2}}$ primarily decreased $[Hbavail]$ when breathing hypoxic gas (0.12 $F_{IO_{2}}$), $CaO_{2}$ did not change as $F_{HBO_{2}}$ increased from 0.02 to 0.30, indicating that maintenance of $CaO_{2}$ attenuates the decrease in $V_{O_{2}max}$ when hypoxia and elevated $F_{HBO_{2}}$ are combined. The $CaO_{2}$ is maintained with the combined gases due to the reduced $[Hbavail]$ being offset by increased $SaO_{2}$ of $Hbavail$ resulting from hyperventilation, increased $Hb$ affinity for $O_2$, and the sigmoid shape of the OEC, as discussed below.

**Hyperventilation.** The $P_{ACO_{2}}$, decreased with $F_{IO_{2}}/F_{ICO_{2}}/F_{HBO_{2}}$ combinations that lowered $CaO_{2}$, (Fig. 5A). Hyperventilation increases alveolar $P_{O_{2}}$ and $P_{A_{O_{2}}}$, as well as $SaO_{2}$, $CaO_{2}$, and $T_{O_{2}max}$. Furthermore, arterial pH increased with $F_{IO_{2}}/F_{ICO_{2}}/F_{HBO_{2}}$ combinations that lowered $CaO_{2}$, (Fig. 5B), increasing $Hb$ affinity for $O_2$ and raising $SaO_{2}$ at similar $P_{O_{2}}$ due to the Bohr effect (1, 4). Hyperventilation results when $V_{A}$ increases relative to $V_{CO_{2}}$ (33). Hypoxia and/or elevated $F_{HBO_{2}}$ lowered $V_{O_{2}max}$ $Sp_{O_{2}}$ and, as a result, also decreased $V_{CO_{2}}$ (Fig. 5C). The $V_{A}$ did not change among $F_{IO_{2}}/F_{ICO_{2}}/F_{HBO_{2}}$ combinations (Fig. 5D), indicating hyperventilation resulted solely from decreased $V_{CO_{2}}$ with hypoxic and/or elevated $F_{HBO_{2}}$ combinations. Hyperventilation not only increases arterial pH and $Hb$ affinity for $O_2$ due to the Bohr effect (1, 4), but also raises alveolar $P_{O_{2}}$ and $P_{A_{O_{2}}}$. Both increase $P_{A_{O_{2}}}$ and the Bohr shift increases $SaO_{2}$ of $Hbavail$, helping to maintain $CaO_{2}$ when $F_{HBO_{2}}$ increases.

**Increased $Hb$ affinity for $O_2$.** Elevated $F_{HBO_{2}}$ increased $Hb$ affinity for $O_2$ independently of the Bohr effect (Fig. 6) as others have reported (19, 22, 36). The average $\Delta P_{SO_{2}}/\Delta F_{HBO_{2}}$ for goat blood was $-32$ Torr, compared with $-27$ Torr for human blood (22) and $-34$ Torr for dog blood (23). As $F_{HBO_{2}}$ increases, the reduction in blood $O_2$ capacitance due to decreased $[Hbavail]$ is partially offset by the increased $Hb$ affinity for $O_2$ causing higher $SaO_{2}$ of $Hbavail$.

**Sigmoid shape of the OEC.** Elevated $F_{HBO_{2}}$ affects $O_2$ delivery differently when breathing normoxic vs. hypoxic gas due to the sigmoid shape of the OEC affecting $SO_{2}$ of blood with high and low $P_{O_{2}}$ differently. With 21/0 $F_{IO_{2}}/F_{ICO_{2}}$, $P_{A_{O_{2}}}$ is $\sim 82$ Torr and approaching the plateau of the OEC with $SaO_{2} > 0.90$ ($P_{O_{2}} \sim 60$ Torr corresponds to 0.90 $SO_{2}$ with $pH$ 7.4 at $37^\circ C$). Therefore, when breathing air, increased $P_{A_{O_{2}}}$ (due to hyperventilation) and/or increased $Hb$ affinity for $O_2$ (due to the Bohr effect or elevated $F_{HBO_{2}}$ shifting the OEC) can raise $SaO_{2}$ only slightly. As a result, increasing $F_{HBO_{2}}$ primarily decreases $O_2$ transport by reducing $[Hbavail]$ with little compensation via increased $SaO_{2}$ because $SaO_{2}$ is already high. Conversely, with 12/0 $F_{IO_{2}}/F_{ICO_{2}}$, $P_{A_{O_{2}}}$ is $\sim 36$ Torr at $V_{O_{2}max}$ and is on the steep portion of the OEC ($SaO_{2}$ $\sim 0.65$ at $pH$ 7.4, $37^\circ C$). Therefore, increased $P_{A_{O_{2}}}$ due to hyperventilation and

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Fig. 6. Hill plots of log $[SO_{2}/(1-SO_{2})]$ vs. log $P_{O_{2}}$ for $F_{HBO_{2}}$ 0.021 $\pm$ 0.004 (solid circles), 0.157 $\pm$ 0.015 (dark shaded circles), 0.303 $\pm$ 0.021 (light shaded circles), and 0.460 $\pm$ 0.029 (open circles), with $SO_{2}$, saturation of $Hbavail$ with $O_2$: $P_{O_{2}}$, $O_2$ partial pressure. Individual points are from arterial and mixed-venous blood collected at rest and submaximal and maximal exercise with goats breathing normocapnic gases. Samples were measured at 37.0°C and corrected to pH 7.40 using an iteratively calculated Bohr coefficient (−0.54). The dotted horizontal line denotes $SO_{2}$ 0.50, and the intersection of the regressions with the dotted line denotes half-saturation pressure ($P_{50}$).

**Hypercapnia.** Hypercapnia (0.05 $F_{ICO_{2}}$) altered neither $V_{O_{2}max}$ when breathing air nor $V_{O_{2}max}$, compared with identical normocapnic $F_{IO_{2}}/F_{HBO_{2}}$ combinations. Hypercapnia decreases $Hb$ affinity for $O_2$ via the Bohr shift, which could reduce $O_2$ delivery. However, we and others (7, 13) detected no changes in $V_{O_{2}}$ at similar submaximal or maximal exercise intensities, with or without hypercapnia, suggesting increased $F_{ICO_{2}}$ (up to 0.05) does not affect $V_{O_{2}max}$ when breathing similar $F_{IO_{2}}/F_{HBO_{2}}$ combinations.

**Effects of gases in combination.** We tested the hypothesis that the reduction in $V_{O_{2}max}$ due to breathing $F_{IO_{2}}/F_{ICO_{2}}/F_{HBO_{2}}$ combinations would equal the sum of the reduction in $V_{O_{2}max}$ caused by each individual gas. The interactive effects model fit the measured $V_{O_{2}max}$ better than the additive effects model that assumes the effects of the two gases act independently and cumulatively (Fig. 2). Furthermore, the additive effects model predicted the impossibility that goats breathing 0.09/0.45 $F_{IO_{2}}/F_{HBO_{2}}$ would have $V_{O_{2}max} < 0$. These data indicate the effects of hypoxia and elevated $F_{HBO_{2}}$ in combination on $V_{O_{2}max}$ are attenuated, not additive. Horvath et al. (24) concluded that the combined effects of hypoxia and $CO$ are additive in humans, although their data showed that $V_{O_{2}max}$ was not always lower with higher $F_{ICO_{2}}$ at the same inspired $P_{O_{2}}$. However, their highest $F_{ICO_{2}}$ resulted in $F_{HBO_{2}} < 0.05$, much less than all noncontrol $F_{HBO_{2}}$ in the present study, possibly eliciting changes too small to detect interactive effects of the gases.

**Effects of changes in $O_2$ transport on $V_{O_{2}max}$.** We tested whether different $F_{IO_{2}}/F_{ICO_{2}}/F_{HBO_{2}}$ combinations affect $O_2$ delivery and peripheral diffusing capacity differently. Elevated $F_{HBO_{2}}$ might reduce peripheral diffusing capacity due to CO effects on myoglobin or cytochrome oxidase (6). However, among the 17 $F_{IO_{2}}/F_{ICO_{2}}/F_{HBO_{2}}$ combinations, $V_{O_{2}max}$ decreased in proportion to $F_{HBO_{2}}$, while $F_{D_{T_{O_{2}}}}$ did not change (Fig. 3, A and B). This suggests that altered inspired gases reduce $V_{O_{2}max}$ by impairing $O_2$ delivery without affecting peripheral diffusing capacity, similar to findings of hypoxic-only studies in humans (5, 34) and rats (20).

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changes in Hb affinity for O₂ markedly increase SaO₂, and compensate for decreased [Hbavail]. As a result, the combined effects of hypoxia and elevated F_{HbCO} acting to decrease V_{O2max} are less than the sum of their individual effects whenever hypoxia causes sufficient hypoxemia to result in P_{aO2}, on the steep part of the OEC.

Critique of methods. A fundamental limitation to this study was that not all goats received all FIO₂/FICO₂/F_{HbCO} combinations, which limited statistical power to detect small differences among treatments. To isolate treatment effects from between-subject variation, we expressed each goat’s measured values at V_{O2max} as fractions of their values when breathing air, which standardized comparisons to account for large (1.7-fold) variation in V_{O2max} among goats (Table 1). However, an advantage to having many FIO₂/FICO₂/F_{HbCO} combinations was that the large number of treatments enabled detection of linear relationships among variables, as well as fitting of a curvilinear response surface to a broad range of gas mixtures, essential for determining their interactive effects.

We used goats as models for humans in this study because of their similar body mass and V_{O2max} (45). Goats are quadrupedal, and humans bipedal. However, energy cost of locomotion is nearly identical between species that run on two or four legs for a given body size (15, 46). Goats are ruminants, and humans monogastric. Eructation of ruminal gases likely biased V_{O2}, V_{CO2}, and respiratory exchange ratio measurements. However, bias due to ruminal gases, including CO₂, on measurements of gas exchange becomes insignificant compared with the CO₂ produced by metabolically active tissues during exercise (28). Therefore, ruminal gas would bias resting measurements far more than measurements at V_{O2max}, and all measurements in this study are at V_{O2max}.

A physiologically important difference between humans and most other mammals is that, with increasing exercise intensity, humans do not hemoconcentrate (44), whereas most species do (45). The goats increased [Hb] from 9.5 ± 0.8 at rest to 14.0 ± 2.1 g/dl at V_{O2max} (P < 0.001) when breathing air. Although hemoconcentration plays a role in determining absolute aerobic capacities of nonhuman mammals, it varies little at V_{O2max} for an individual, even when breathing different FIO₂/FICO₂/F_{HbCO} combinations (+3%, data not shown).

The ventilatory response to exercise differs between humans and the goats in this study. Humans hyperventilate at V_{O2max} (10, 38), as do most mammals other than horses (11), including pygmy goats that were one-half the size of the goats in this study (45). In contrast, the goats in this study hyperventilated at V_{O2max} when breathing air (P_{aCO2} 42.8 Torr at rest, 52.7 Torr at V_{O2max} vs. 52.7 Torr at V_{O2max}). This difference in ventilatory response to exercise between humans and the goats in this study might result in humans experiencing less progressive hyperventilation with hypoxia and/or elevated F_{HbCO} than we observed in these goats (Fig. 5A); otherwise, humans would become extremely hypocapnic at V_{O2max}.

Conclusions. Breathing either hypoxic or CO-containing gas alone decreases V_{O2max} in proportion to the amount of hypoxia or elevated F_{HbCO}. Hypoxia and elevated F_{HbCO} in combination decrease V_{O2max} by less than the sum of each gas individually primarily because reductions in Cao₂, are attenuated. Mechanisms that attenuate the decrease in Cao₂ with combined hypoxia and elevated F_{HbCO} are hyperventilation (with either gas or both), increased Hb affinity for O₂ (due to both Bohr and direct F_{HbCO} effects on the OEC), and the sigmoid shape of the OEC increasing S0₂ more at low P_{aO2}, (hypoxia) than at high P_{aO2}, (normoxia). The V_{O2max} correlates highly with T_{O2max} and Cao₂, suggesting that reduced O₂ transport when breathing hypoxic and CO-containing gases plays a primary role in lowering V_{O2max}.

APPENDIX

Mean capillary P_{O2} (P̅_{O2}), not mixed-venous P_{O2} (P_{vO2}), represents the true driving pressure for diffusion in peripheral capillaries. In cycling humans breathing normoxic or hypoxic gas, P_{CO2} is proportional to both P_{vO2} and femoral-venous P_{O2} (P_{fV_{O2}}) (34), indicating that P_{CO2}, P_{vO2}, or P_{fV_{O2}} can be used for estimating changes in peripheral diffusion capacity (D_{TO2}). However, in these experiments, goats breathed combinations of hypoxic and/or hypercapnic gases, both with and without elevated F_{HbCO}, the effects of which could potentially alter the relationship between P_{CO2}, and P_{fV_{O2}}. Both CO₂ and CO change Hb affinity for O₂, although with opposite effect. Additionally, CO reduces O₂ capacitance. These changes might affect P_{CO2}, and P_{fV_{O2}} differently. Therefore, we determined whether different FIO₂/FICO₂/F_{HbCO} combinations differentially affect D_{TO2}, (V_{O2max}/ P_{CO2}) or D_{fV_{O2}} (V_{O2max}/P_{fV_{O2}}).

We calculated P_{fV_{O2}} using a Bohr integration procedure similar to the algorithm used for hypoxic humans (34). However, those studies used P_{fV_{O2}} to estimate end-capillary P_{O2}, and we did not measure P_{fV_{O2}} in the goats. However, because the goats had similar body mass, V_{O2max}, and P_{fV_{O2}} to the cycling humans in this study, we assumed the relationship between P_{fV_{O2}} and P_{fV_{O2}} in the goats was similar to that measured in humans (34). Therefore, we estimated caprine P_{fV_{O2}} based on measured P_{fV_{O2}} using the linear relationship between these variables described in the human study (Fig. 7). We then calculated the D_{fV_{O2}}/Q ratio that resulted in the estimated P_{fV_{O2}} for each gas combination. Although this assumption is an oversimplification, it

\[
\begin{align*}
\text{PO}_{2} \quad \text{Torr} & \quad \text{PO}_{2} \quad \text{Torr} \\
20 & \quad 30 \\
30 & \quad 40 \\
40 & \quad 50 \\
\end{align*}
\]

Fig. 7. Linear regression of calculated mean capillary PO₂ (P̅O₂; solid line) and estimated femoral-venous PO₂ (PfV₀₂; long-dashed line) vs. measured mixed-venous PO₂ (P_{vO₂}; abscissa and short-dashed line) with altered FICO₂, P_{fV₀₂}, and F_{HbCO} in goats running on a treadmill at V_{O2max}. Shapes denote different FIO₂ (0.21 circles, 0.15 inverted triangles, 0.12 triangles, 0.09 diamonds), shades different F_{HbCO} (0.02 solid, 0.15 dark shaded, 0.30 light shaded, 0.45 open), and a cross within the symbol denotes hypercapnia (0.05 F_{CO₂}). Legend for symbols and sample size is in Fig. 3A. Values are means ± SD for goats that breathed that gas mixture (n = 3–6). The linear regression of P_{CO₂} vs. P_{fV₀₂} is P_{fV₀₂} = 1.46 P_{vO₂} + 3.29 (R² = 0.9960, P < 0.001). Open stars denote P_{CO₂}, P_{fV₀₂}, and P_{fV₀₂} in humans breathing normoxic or hypoxic gas at V_{O2max} from Ref. 34 for comparison.
enables a first approximation of the $P_{O_2}$ vs. $P_{O_2}$ relationship to be made by eliminating the need to measure blood flow to working muscles, components of diffusing capacity (e.g., HbO2 reaction rate and capillary volume), and capillary transit time and is similar to the approach used by Roca et al. (34).

Results from the Bohr integrations show that $P_{CO_2}$ and $P_{O_2}$ decrease proportionally (Fig. 7), similar to their relationships in humans (34), dogs (2), and rats (20). The proportionality between $P_{CO_2}$ and $P_{O_2}$ for all $FIO_2/FICO_2/FHbCO$ combinations tested suggests that changes in peripheral diffusing capacity can be demonstrated whether diffusing capacity is calculated using measured $P_{O_2}$ or calculated $P_{CO_2}$. However, absolute values of $D^{\prime}_{O_2}$ are biased high compared with $D_{O_2}$, because $P_{O_2}$ is less than $P_{CO_2}$.

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DISCLOSURES

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AUTHOR CONTRIBUTIONS


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