Analysis of factors that influence rates of carbon monoxide uptake, distribution, and washout from blood and extravascular tissues using a multicompartment model

Margaret C. Bruce and Eugene N. Bruce

1Department of Physiology and 2Center for Biomedical Engineering, University of Kentucky, Lexington, Kentucky

Submitted 3 May 2005; accepted in final form 1 December 2005

Bruce, Margaret C, and Eugene N. Bruce. Analysis of factors that influence rates of carbon monoxide uptake, distribution, and washout from blood and extravascular tissues using a multicompartment model. J Appl Physiol 100: 1171–1180, 2006. First published December 8, 2005; doi:10.1152/japplphysiol.00512.2005.—To better understand factors that influence carbon monoxide (CO) washout rates, we utilized a multicompartment mathematical model to predict rates of CO uptake, distribution in vascular and extravascular (muscle vs. other soft tissue) compartments, and washout over a range of exposure and washout conditions with varied subject-specific parameters. We fitted this model to experimental data from 15 human subjects, for whom subject-specific parameters were known, multiple washout carboxyhemoglobin (COHb) levels were available, and CO exposure conditions were identical, to investigate the contributions of exposure conditions and individual variability to CO washout from blood. We found that CO washout from venous blood was biphasic and that postexposure times at which COHb samples were obtained significantly influenced the calculated CO half times ($P < 0.0001$). The first, more rapid, phase of CO washout from the blood reflected the loss of CO to the expired air and to a slow uptake by the muscle compartment, whereas the second, slower washout phase was attributable to CO flow from the muscle compartment back to the blood and removal from blood via the expired air. When the model was used to predict the effects of varying exposure conditions for these subjects, the CO exposure duration, concentration, peak COHb levels, and subject-specific parameters each influenced washout half times. Blood volume divided by ventilation correlated better with half-time predictions than did cardiac output, muscle mass, or ventilation, but it explained only ~50% of half-time variability. Thus exposure conditions, COHb sampling times, and individual parameters should be considered when estimating CO washout rates for poisoning victims.

Despite this concern, the factors influencing the uptake, extravascular distribution, and washout of CO have received relatively little attention in recent years. There are conflicting reports in the literature regarding the rate of removal of CO from the blood in response to specific treatment modalities, and clinical studies designed to address this issue have, of necessity, often relied on retrospective analyses of patient populations to investigate the washout kinetics of CO (8, 12, 14, 15, 23, 24). Although clinical studies have the potential advantage of including large numbers of subjects exposed to a range of CO concentrations, the lack of information regarding exposure conditions for each patient makes it difficult to assess the separate effects of subject-specific parameters and CO exposure conditions on washout rates. The degree of inter- and intrastudy variability, together with clinical observations that carboxyhemoglobin (COHb) levels are not reliable indicators of poisoning severity (8, 11, 13), indicate a need to better understand the factors that influence CO washout to optimize treatment strategies.

The majority of studies published to date have claimed that CO washout follows first-order kinetics, as assumed by the Coburn-Forster-Kane equation (5, 14, 16, 23). Monoexponential washout would be consistent with the concept that CO is being removed from either a single vascular compartment or a vascular compartment in rapid equilibrium with extravascular tissues. If, as proposed by Luomanmaki and Coburn (9), CO equilibrates rapidly with extravascular tissues, the subsequent decrease in COHb beyond this equilibration period should primarily reflect the removal of CO via the expired air. Alternatively, if, after rapid mixing within the vascular compartment, CO flows from the blood to both the extravascular muscle compartment and the expired air, the washout rate from the blood should be faster initially while CO continues to be taken up by the muscle, then slower when the CO flux is from the muscle back to the blood. Under these conditions, the resulting decrease in COHb would not be monoexponential.

Often cited as the first to propose a biphasic washout curve for CO, Wagner and colleagues (22) measured COHb levels in the arterial blood of anesthetized dogs exposed to CO, but the initial washout phase in this study occurred over a 10-min period immediately after a 3-min CO exposure. These authors proposed that the initial phase was likely to be due to vascular mixing rather than washout from the blood. Furthermore, because these investigators followed arterial COHb levels for only 85 min postexposure, stopping well before their calcu-
lated half times of 134 and 190 min, they did not have sufficient data to distinguish a linear from an exponential decrease in COHb. More recently, Shimazu (19) challenged the concept of monoexponential decay, reporting a biphasic washout curve determined from multiple COHb levels obtained over 240 min in a CO-poisoned patient. However, the severity of the poisoning episode raised questions about the extent to which the patient’s response to treatment might have influenced the shape of the CO washout curve (25). In the present study, we have used mathematical modeling to gain further insight into the washout kinetics of CO. The model simulations used in this study were based on data from 15 human subjects who inhaled the same amount of CO over a 4- to 6-min period and were then monitored for 4.5 h while breathing room air (1). Because frequent COHb samples were obtained from these subjects, we were able to fit the model to each subject individually and to compare various methods of calculating “half times” of CO washout to the actual time required for COHb to fall by 50%.

A semilog plot of the COHb levels obtained during washout demonstrated that the washout curves for these experimental subjects were not monoexponential, an observation consistent with our model predictions of a slow equilibration of CO between the blood and extravascular muscle compartment during and after the end of the exposure. In addition, we were able to use model simulations to predict for each subject the effects of CO concentration, exposure duration, and peak COHb level on washout half times and to identify the model parameters that provided the best correlation with measures of CO half times.

METHODS

Simulation Model

The basic simulation model has been described previously and validated with data from various studies of the uptake of CO during inhalation exposures (2). It is applicable to normal human subjects at rest. Briefly, it comprises three uniform tissue compartments (lungs, whole-body muscle tissue, and whole-body “other” tissue) and four vascular compartments (arterial blood, mixed venous blood, vascular subcompartment of muscle, and vascular subcompartment of the “other” tissue compartment). In the muscle compartment CO can combine with myoglobin (Mb) in competition with O2 as expressed by the Haldane equation (2). Additional details of the model, assumptions, and sensitivity analyses are available from Ref. 2.

For the present study, some minor modifications were made to the previous model. First, the whole-body metabolic rate of O2 consumption (MR) was made proportional to the body weight (BW) according to the relationship \[ MR = BW \times 224/70, \] where MR for a 70-kg subject at rest is assumed to be 224 ml/min. Second, a venous-to-arterial shunt blood flow was added to the lung compartment; however, in the present study this flow was maintained at the normal value of 5% of cardiac output. In this study, arterial PO2 is set to 100 or 500 Torr during breathing of room air or 100% O2, respectively. Partial pressure of O2 in muscle tissue is set to 21 Torr during breathing of CO in air, 17 Torr during washout of CO on air, and 40 Torr during washout of CO on O2. It is assumed that ventilation is constant during exposure to CO and during washout of CO; however, its value may be different for exposure vs. washout. It is assumed that cardiac output, muscle metabolism, and muscle blood flow do not change due to CO exposure. Unless otherwise noted, “venous” COHb refers to COHb in the venous outflow from the whole-body muscle compartment.

Parameter Estimation

To address the variability of responses in a normal population, estimates of model parameters were obtained for each subject studied by Benignus et al. (1). In this study normal male volunteers breathed a concentration of 6,683 ppm CO in air for ~5 min, or until the bag of gas was empty, followed by room air for ~4 h. The investigators measured both arterial and muscle venous COHb values every minute during the CO exposure and muscle venous COHb values at seven time points over a period of 4 h after termination of the exposure. Thus it was possible to fit the model to both exposure and washout by comparing the predicted values for arterial and muscle venous COHb with the experimental data. Values for the following parameters of individual subjects either were available in published reports (1, 20) or were supplied to us by Dr. Benignus: gender (all male), age, height, weight, hemoglobin concentration, total blood volume, resting cardiac output, lung CO-diffusing capacity, and resting ventilation measured on a separate day from the day of CO exposure. To establish which model parameters were most influential in determining the COHb responses to the specific protocol used by Benignus et al., the effects on muscle venous and arterial COHb predicted from the model were compared when this protocol was simulated and the remaining unspecified parameter values were varied individually. Those parameters having major effects were ventilation during CO exposure (VA wi); ventilation during washout (VAwo); the fraction of cardiac output that perfuses the whole-body muscle compartment (QFm); and the fraction (VFm) of the total blood volume (Vb) assumed to reside in the vascular subcompartment of the muscle compartment. Consequently, the above four parameters were estimated by fitting the model to the data from individual subjects while the remaining parameters of the model were either set to known values (as explained above) or set to average values determined from our previous study (2). The parameter estimation procedure is explained in the APPENDIX.

CALCULATION OF HALF TIMES

Ideally, calculated half-time values for washout of CO should closely estimate the actual time required for COHb to fall from its peak value to 50% of the peak value (t50); however, the accuracy of these calculations would depend on the postexposure times at which COHb levels were obtained. To address this issue, estimates of half times for washout of CO were determined by three different methods. The basic methods are described here and the specific applications of each method are described in RESULTS.

In the present study, the model fit to the seven postexposure %COHb levels from each of the Benignus subjects provides continuous values for muscle venous COHb. The first method we used determined the actual time required for venous COHb to fall from its peak value (near the end of CO exposure) to one-half of that value. This measurement is designated the “true” half time (t50) for washout of CO, although, as discussed subsequently, often it did not accurately reflect the time course of CO washout at later times when COHb was below one-half of its peak value. When more than one half time was calculated by this method, the first true half time was designated t50-.5 and the second half time was designated t50-.75.

The second, the two-point method, estimated the half time from values of venous COHb at two different time points (23). If c1 and c2 are the COHb values at times t1 and t2 during the CO washout phase, then the half time (th) based on this two-point method is given by

\[ th = \ln(2) \times \frac{t_2 - t_1}{\ln(c_1) - \ln(c_2)} \]

The selected times, t1 and t2, corresponded either to the time of...
predicted peak COHb and a later time during washout or to two times at which actual experimental data were acquired. This method assumes that COHb follows a monoexponential decay during washout.

The third, the linear regression method, involves fitting a straight line via linear regression to a plot of the natural logarithm (ln) of muscle venous COHb vs. time during washout. From the slope of the line, half time (ht) is calculated as $ht = \frac{\ln(2)}{\text{magnitude of slope}}$ (14). If COHb follows a monoexponential decay, then the plot of ln(COHb) vs. time will be a straight line and ht can be properly interpreted as a half time. Because all of our plots of this type were curvilinear, we determined linear fits to both early and late portions of the washout curves. Thus each ht value represents an “apparent” half time given only the data used to fit each line. We did not attempt to fit the COHb curves with a theoretical washout curve comprising the sum of two exponentially decaying terms (19).

Statistical Analyses

Values are means ± SD. Group means were considered to be significantly different when $P < 0.05$. Student’s paired t-test was used to compare half times calculated by the two-point method over a range of washout times and to compare the slopes of the first and second phases of washout curves. Linear regression analysis was used to determine the correlation coefficients for washout half times and subject-specific parameters.

RESULTS

Model Fitting: Parameter Ranges and Variability of Responses Within the Subject Population

We were able to fit the model to the individual data from each of the 15 subjects. The parameter estimation procedure described in the APPENDIX converged for 14 subjects. For subject 115, however, the very large difference between actual arterial and venous COHb values during CO exposure (see Ref. 1) could only be reproduced if the blood flow to the muscle compartment of the model was small. Although the final value of QFm for subject 115 was the smallest in the study group, the fit to the venous COHb data during washout was good and this subject was included in all aspects of this study, with the exception of the simulations of exposure duration and concentration effects on half time. In the case of subject 107, the parameter estimation was iterated as explained in the APPENDIX, but the criteria for stopping were never met because of a strong negative correlation between two parameters (QFm and VFm) and because the optimal values seemed to differ between the exposure and washout stages. Because the correlated parameters both varied around a value of 0.30 during the various fitting trials, this value (which provided a better fit to the washout than to the exposure data) was chosen for both of them and then the model fit to the data was determined by estimating ventilation values during exposure and washout. For all other subjects, the resulting fits of the model predictions of both arterial and venous COHb values to the data reported by Benignus et al. (1) during both CO exposure and washout were visually good (Fig. 1). However, during the time from 10 to 20 min after the end of the CO exposure, the model predictions of venous COHb were consistently slightly larger than the data values. This situation arose because the fall in COHb levels during the initial part of washout was more rapid than the model could reproduce. There may be a rapid transport or equilibration mechanism missing from the model or, possibly the assumption that ventilation was constant is incorrect. In any case, the predicted venous COHb waveform provides a visually good interpolation between the experimental data points during the rest of the washout and, by assumption, an extrapolation of the time course of venous COHb during washout beyond the last experimental data point at 264 min.

The COHb responses of these subjects to the protocol of Benignus et al. (1) varied widely, especially during the CO exposure period. Accordingly, the estimated parameter values for our model also varied widely. However, the average blood flow to whole-body muscle in the model (i.e., ~20% of cardiac
output under resting conditions) and the fraction of total blood volume resident in muscles (i.e., ~40%) are within the ranges one can infer from published data [e.g., see appendix of Smith et al. (20)]. In a few cases, the estimated ventilation values during CO exposure are higher than expected for “resting” subjects, whereas during washout ventilation rates for most subjects are notably higher than typical resting values. During the CO exposure subjects were consciously attempting to empty a 50-liter bag (from which they inspired CO in air) in 5 min; thus they may have hyperventilated. During washout it is possible that the subjects subconsciously hyperventilated because they were aware of their exposure to CO. In addition, the model does not allow cardiac output to differ between exposure and washout, but a higher cardiac output during washout would have reduced the ventilation needed to match the data.

Uptake and Washout of Carbon Monoxide in Arterial and Muscle Venous Blood

Due to the short duration of the CO exposure in the Benignus study (1), the model predicts that very little CO enters the extravascular compartments by the end of the exposure. During washout, the major flux of CO out of the blood is that which enters the lung compartment (Fig. 2A); however, because $P_{CO}$ in the blood remains high for a long time during washout on room air, CO continues to flow from blood to the muscle compartment for ~250 min after termination of the exposure (Fig. 2B). The flux of CO into the muscle compartment causes the rate of fall of arterial COHb during the early washout phase to be greater than that achieved by washout through the lungs and ventilation alone; however, later in the washout phase CO flux is from the muscle compartment back to the blood and the rate of fall of arterial COHb is less than the rate at which ventilation removes CO from the blood (Fig. 2C).

Dependence of COHb Half-Time Estimates on Sampling Times: Washout on Air

True COHb half times calculated from minute-to-minute venous COHb levels. COHb levels were measured on venous blood samples obtained from 15 subjects at 1-min intervals for the first 5 min postexposure and at 7, 11, 19, 35, 67, 131, and 259 min postexposure. Using model simulations to interpolate between the actual data points, we calculated the true COHb half time ($t_{0.50}$) for each subject by determining the time required for venous COHb to decrease to the peak value to one-half the peak level. With the use of this approach, the mean $t_{0.50}$ for the 15 subjects was 248.5 ± 39.6 min (range 206–328 min) whereas the mean value for the subsequent or second true half time ($t_{50.75}$) was 302.5 ± 50.3 min (range 251–406 min). The two values were significantly different ($P < 0.0001$). The mean difference between the two half times was 54.0 ± 12.1 min.

Factors contributing to half-time variability among subjects. As shown above, despite the fact that all subjects inhaled the same amount of CO over a 4- to 6-min period, there was considerable intersubject variability with respect to washout half times. These $t_{0.50}$ values were found to correlate better with $Vb/V_{AWO}$ ($r = 0.714$) ($P < 0.01$) than with other variables, e.g., $V_{AWO}$ (~0.537), $Vm/Qm$ (0.639), or $Vbm/Qm$ (0.621) (Fig. 3). Therefore, two parameters (similar to volume of distribution of CO and ventilation from the Coburn-Forster-Kane equation) explain roughly 50% of intersubject variability.

$COHb$ half time calculated from venous blood samples obtained during washout. To determine the extent to which COHb half-time estimates vary with postexposure sampling times, we used the two-point method described in METHODS to calculate half times for the 12 subjects for whom COHb levels were available at 5, 19, 35, 67, 131, and 259 min postexposure and compared these half times with the $t_{0.50}$ values described above. As shown in Table 1, mean half times calculated from venous samples obtained at 19 and 67 min (232 ± 39 min), at 19 and 131 min (249 ± 36 min), at 35–67 min (247 ± 78 min), and at 35–131 min (254 ± 39 min) were similar to the mean value for the $t_{0.50}$ of 246 ± 35 min for these 12 subjects. In contrast, the mean half time calculated at earlier time points, 5 and 67 min, was shorter (208 ± 78 min) whereas the mean half times were considerably longer when determined from venous COHb levels obtained at 19 and 259 min, 35 and 259 min, 67 and 131 min, and at 131 and 259 min postexposure.

![CO flux during washout for Benignus subject 108](attachment:Fig_2.png)  

A: CO flux from blood to lungs (CO FluxBL). B: CO flux from blood to muscle (CO FluxBM). C: CO uptake by and removal from hemoglobin (COHb).
Washout of CO under room air is not monoexponential. Using model extrapolations from the experimental COHb values obtained during washout under room air, we plotted COHb levels on a log scale vs. time on a linear scale, using the linear regression method to determine half time (Fig. 4). Regression lines fitted to the initial and final segments of the washout curves for each subject indicated that washout under room air was not monoexponential. The mean values of the half times for the 15 subjects were 236.2 ± 34.4 min for the first half of the washout curve and 302.3 ± 38.9 min for the second half of the washout curve. The slope of the first regression line was significantly greater (and thus washout was significantly faster) than the slope of the second (P < 0.0001).

Effects of setting the CO flux to the muscle compartment to zero. In the model, the diffusion of CO to muscle governs the rate of flow of CO between blood and muscle tissue. The blood-to-muscle diffusion coefficient, DmCO, is the analog in muscle tissue of CO-diffusing capacity in the lungs. To evaluate the extent to which uptake of CO by the muscle compartment contributes to the shape of the washout curve, we set DmCO to zero and determined, for each of the 15 subjects, the time required for the COHb level to decrease by 50% from its peak. The mean value for t0.50 increased significantly (P < 0.0001) from 248.5 ± 39.6 min when DmCO was set at 1.75 ml·min⁻¹·Torr⁻¹ (the default value) to 268.8 ± 42.6 min when DmCO was set to zero.

Washout rate decreases as exposure time to reach a constant CO level increases. The model was then used to compare washout curves under varied exposure concentrations and durations, each of which resulted in peak COHb levels of 20%. As shown in Fig. 5, because the percent carboxymyoglobin (COMb) was approximately five times higher at the end of the 600-min exposure vs. the 5-min exposure, the CO flux from blood to the muscle compartment during washout was greater after the 5-min than after the 600-min exposure. Consequently, the rate of COHb washout from venous blood was slightly faster after the 5-min exposure to 10,000 ppm than after the 600-min exposure to 160 ppm.

For relatively short exposures in which the products of CO concentration and exposure duration, and thus the CO dose, are equal, e.g., 50,000 for an exposure to 10,000 ppm for 5 min and also for an exposure to 1,250 ppm for 40 min, the predicted peak COHb levels were ~20%. However, as the CO concentration decreased and the duration of the exposure increased beyond 40 min, peak COHb levels decreased, despite the fact that the product of exposure duration and CO concentration remained constant. For example, although the product of concentration multiplied by time is also 50,000 for a 1,280-min exposure to 39 ppm, the model predicts for subject 108 a peak COHb of only 6.6% and a slower washout rate than after the 5-min exposure to 10,000 ppm. As shown in Fig. 6, A and B, during the 1,280-min exposure, some CO leaves the blood and enters the muscle, thus decreasing the total amount in the blood but keeping the body burden relatively constant. During washout after the 5-min exposure to 10,000 ppm, a substantial fraction of the CO in blood continues to enter the muscle for ~275 min, whereas after the 1,280-min exposure a much smaller fraction of the CO in blood enters the muscle compartment during washout because the %COMb is much higher at the end of the 1,280-min exposure, 3.35% vs. baseline COMb levels after the 5-min exposure.

Table 1. Half times calculated from COHb levels obtained from 5 to 259 min postexposure

<table>
<thead>
<tr>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>106</td>
<td>207</td>
<td>144</td>
<td>162</td>
<td>209</td>
<td>238</td>
<td>156</td>
<td>216</td>
<td>244</td>
<td>268</td>
<td>270</td>
</tr>
<tr>
<td>107</td>
<td>219</td>
<td>168</td>
<td>189</td>
<td>195</td>
<td>256</td>
<td>217</td>
<td>204</td>
<td>270</td>
<td>199</td>
<td>355</td>
</tr>
<tr>
<td>108</td>
<td>232</td>
<td>210</td>
<td>225</td>
<td>220</td>
<td>267</td>
<td>388</td>
<td>254</td>
<td>292</td>
<td>217</td>
<td>328</td>
</tr>
<tr>
<td>109</td>
<td>315</td>
<td>443</td>
<td>316</td>
<td>292</td>
<td>354</td>
<td>410</td>
<td>310</td>
<td>371</td>
<td>277</td>
<td>434</td>
</tr>
<tr>
<td>110</td>
<td>207</td>
<td>149</td>
<td>258</td>
<td>243</td>
<td>266</td>
<td>191</td>
<td>217</td>
<td>253</td>
<td>232</td>
<td>290</td>
</tr>
<tr>
<td>111</td>
<td>276</td>
<td>184</td>
<td>227</td>
<td>280</td>
<td>368</td>
<td>241</td>
<td>298</td>
<td>391</td>
<td>339</td>
<td>509</td>
</tr>
<tr>
<td>112</td>
<td>262</td>
<td>190</td>
<td>245</td>
<td>286</td>
<td>313</td>
<td>214</td>
<td>278</td>
<td>311</td>
<td>326</td>
<td>341</td>
</tr>
<tr>
<td>113</td>
<td>236</td>
<td>175</td>
<td>226</td>
<td>236</td>
<td>295</td>
<td>186</td>
<td>220</td>
<td>289</td>
<td>243</td>
<td>377</td>
</tr>
<tr>
<td>116</td>
<td>260</td>
<td>216</td>
<td>251</td>
<td>240</td>
<td>292</td>
<td>246</td>
<td>237</td>
<td>294</td>
<td>233</td>
<td>359</td>
</tr>
<tr>
<td>117</td>
<td>224</td>
<td>210</td>
<td>264</td>
<td>250</td>
<td>326</td>
<td>275</td>
<td>251</td>
<td>334</td>
<td>241</td>
<td>442</td>
</tr>
<tr>
<td>118</td>
<td>294</td>
<td>208</td>
<td>212</td>
<td>309</td>
<td>315</td>
<td>196</td>
<td>320</td>
<td>320</td>
<td>470</td>
<td>321</td>
</tr>
<tr>
<td>120</td>
<td>218</td>
<td>200</td>
<td>211</td>
<td>225</td>
<td>248</td>
<td>247</td>
<td>254</td>
<td>302</td>
<td>274</td>
<td>358</td>
</tr>
<tr>
<td>Mean</td>
<td>246</td>
<td>208</td>
<td>232</td>
<td>249</td>
<td>295*</td>
<td>247</td>
<td>254</td>
<td>302</td>
<td>274</td>
<td>358</td>
</tr>
<tr>
<td>SD</td>
<td>35</td>
<td>78</td>
<td>39</td>
<td>36</td>
<td>42</td>
<td>78</td>
<td>39</td>
<td>46</td>
<td>75</td>
<td>73</td>
</tr>
</tbody>
</table>

“True” half times (t₁/₂) vs. half times calculated from actual carboxyhemoglobin (COHb) data using the 2-point method. *Significantly greater than the half-time calculated at t₁ = 19 min and t₂ = 67 min (P < 0.0001). t₁, Postexposure time (in min) that the first COHb was obtained; t₂, postexposure time (in min) that the second COHb was obtained.
Influence of Washout on 100% Oxygen on COHb Half Time

COHb half time calculated from peak to half-peak venous COHb level. We then used model simulations to predict COHb values during washout under 100% O₂ for the 15 subjects after the CO exposure described above (6,683 ppm CO in air for 4–6 min). The mean half time obtained by determining the number of minutes required for the peak venous COHb level to decrease by one-half, the $t_{0.50}$, was 87.1 ± 12.8 min (range 76–102 min).

Washout of CO under 100% O₂ is not monoexponential. As observed for washout under room air, regression lines fitted to a plot of the natural logarithm of venous COHb levels vs. time for each subject indicated that washout under 100% O₂ was not monoexponential (Fig. 4B). The mean values for the 15 subjects were 86.9 ± 10.5 min for the half time of the initial phase of the washout curve and 160.7 ± 11.9 min for the half time of the second phase of the washout curve. The mean difference between the two slopes, 73.8 ± 8.6 min, was significant ($P < 0.0001$).

Effects of setting the CO flux to the muscle compartment to zero. When $D_{mCO}$ was set to zero, there was a significant increase in $t_{0.50}$ from 87.4 ± 12.4 min ($D_{mCO} = 1.75$ ml·min⁻¹·Torr⁻¹) to 96.1 ± 9.2 min ($D_{mCO} = 0$) ($P < 0.001$). The mean difference between half times was 8.7 ± 7.8 min. Although the washout rate was slower during the first phase of the washout curve, it was faster during the second phase when $D_{mCO}$ was set to zero: mean values for the $t_{0.50}$ increased 10.5 ± 2.5 min whereas mean values for the $t_{50}–75$ decreased 46.6 ± 6.3 min.

Effects of CO concentration and exposure duration on half times for washout on 100% O₂. When CO concentration and exposure duration conditions were varied for subject 108 to produce similar COHb levels, e.g., 10,000 ppm × 5 min vs. 1,250 × 40 min, resulting in peak COHb levels of 15.9 and 15.8%, respectively, the half times, calculated by the linear regression method, for both phases of the washout curves were essentially identical under the two exposure conditions. Although the CO dose was similar for an exposure to 39 ppm for 1,280 min, half times for the first and second phases of the washout curve were 20 and 15% longer, respectively, than after the shorter exposures to higher CO concentrations.

As shown in Fig. 6, C and D, the initial washout rate of venous COHb under hyperoxia is more rapid after exposure to 10,000 ppm for 5 min than after exposure to 39 ppm for 1,280 min as was the case for washout under room air for these exposure conditions. In both cases, uptake of CO by the muscle compartment increased at the onset of hyperoxic washout, but the magnitude and duration of this increased uptake were much smaller after the 1,280-min exposure to 39 ppm.

DISCUSSION

We have combined experimental data with mathematical modeling to address broadly the issue of washout of CO to better understand the factors that influence this process. To accomplish this objective, we fit a basic model of CO uptake

![Fig. 4. CO washout is not monoexponential on air or on 100% O₂. Values for ln(%COHb) vs. time for Benignus subject 108. A: washout on air; $h_{t2} > h_{t1}$ ($P < 0.0001$). B: washout on 100% O₂; $h_{t2} > h_{t1}$ ($P < 0.0001$). $h_{t1}$, First half time; $h_{t2}$, second half time; heavy dashed line, experimental data; solid line ($h_{t1}$), linear regression fit to early washout; dashed line ($h_{t2}$), linear regression fit to late washout.](http://jap.physiology.org/DownloadedFrom/10.220.33.3/1176/MULTICOMPARTMENT-MODEL-OF-FACTORS-AFFECTING-CO-WASHOUT)
and washout to data from a well-controlled study involving 15 young adult male subjects who inhaled identical amounts of CO over a 4- to 6-min period (1). The availability of subject-specific parameters relevant to our model, together with multiple COHb values obtained during the exposure and washout periods, enabled us to develop individual model fits and examine CO washout under both the reported experimental conditions and other conditions in a study population sufficiently large (15 subjects) to evaluate the range of responses among individuals.

The mathematical model we utilized is more comprehensive than the Coburn-Forster-Kane equation (5) and more adequately represents both arterial and venous uptake as well as vascular vs. extravascular distribution of CO in humans during CO exposure and washout. We determined that, for each subject, a single set of physiological parameters produces a good fit of the model to both exposure and washout on air when we allowed ventilation to differ between the exposure and washout. We found that the predicted time courses of %COHb in arterial and muscle venous blood during 5 min of CO exposure, and in muscle venous blood during >4 h of washout, closely approximate the experimental data. This result strongly suggests that these predicted time courses constitute both reasonable interpolations of muscle venous %COHb between experimental data points and a valid extrapolation beyond the last data point. Although we do not have equivalent %COHb data for these 15 subjects during washout while breathing 100% O2, the predicted half times for washout under hyperoxia are quantitatively consistent with the literature from human experimental and clinical studies.

In contrast to the half-time value of 320 min (range 128–409 min) (16) often quoted for washout under air, we found the venous muscle COHb level decreased by one-half in 248.5 ± 39 min (range 206–328 min), a considerably shorter value. Our model predictions and analyses of experimental data offer several possible explanations for the variability of reported half times both within a specific study and among similar studies. The first relates to the postexposure time at which blood samples were obtained. When we used the two-point method employed by others (16, 23) to calculate washout on room air from actual COHb values, half-time estimates were shorter (208 ± 78 min) than true half times (248 ± 39 min) for pairs of samples obtained shortly after the exposure, e.g., at 5 and 67 min, but longer than true half times when one or both samples were obtained at later postexposure times. The samples that provided the half times that best approximated true half times for washout under air were those obtained within the first 2.5 h, e.g., when \( t_1 \) was either 19 or 35 min and when \( t_2 \) was either 67 or 131 min postexposure. Thus the reported half-time variability in earlier studies could be due, in part, to differences in the postexposure times at which blood samples were obtained for analysis of COHb.

CO exposure conditions could also contribute to the observed half-time variability. Our model predictions for Benignus’ subjects demonstrated that half times for washout from blood were influenced by the CO concentration, the exposure duration, and the COHb level at the end of the exposure. In general, the initial half time decreased as the peak COHb level increased, regardless of whether the higher COHb level was due to a longer exposure or a higher inspired CO concentration.

A third explanation for the variability in reported half times is that washout rates vary widely among individuals. The range of half times observed for Benignus’ subjects (206–328 min) clearly demonstrates that washout rates can vary considerably under identical exposure conditions. Although ventilation was a significant factor, the ratio of blood volume to ventilation

---

**Fig. 6.** Model predictions for Benignus subject 108 after 2 exposure regimens, for which the products of CO concentration and exposure duration were identical, resulted in markedly different rates of CO uptake by muscle during washout. For washout on air, a large fraction of CO continued to enter the muscle compartment for ~300 min after the exposure to 10,000 ppm for 5 min (A), whereas a much smaller fraction of CO in the blood entered the muscle after the 1,280-min exposure to 39 ppm (B). For washout under 100% O2, CO uptake by muscle was much greater after the 10,000 ppm exposure for 5 min (C) than after the 39 ppm exposure for 1,280 min (D).
during washout was found to correlate better with half time than did ventilation alone.

That the ratio of blood volume to alveolar ventilation represents the dominant time constant for CO transport during washout is not surprising; neither is the fact that this correlation explains only $\sim 50\%$ of the variability in half times. At the end of an exposure, most of the CO resides in the blood. Consequently, to a first approximation, blood comprises the volume to be cleared of CO. Because CO diffuses readily from blood into lungs, the limiting factor is the rate at which CO is exhaled. Thus the ratio of blood volume to ventilation reflects the major time constant for clearing CO from the blood. At the same time, CO is flowing between the blood and extravascular compartments, and these secondary fluxes modify CO washout from that expected from the major time constant. Muscle mass, although a factor, was a less important determinant of half time because the volume of CO removed from muscle was less than the volume removed from blood. Similarly, other individual factors correlated with half times, but only weakly, because they influence only one of the two factors in the major time constant, i.e., either the volume of CO in blood, or the rate of CO expired by ventilation, or some secondary factor related to CO transport or storage.

The possibility that sampling times might influence the calculated half time has been largely discounted in the past because of the commonly cited premise that CO washout follows a monoeponential curve, i.e., that $\ln(\text{COHb})$ vs. time during washout is a straight line (14, 16, 23). However, the results presented herein provide compelling evidence that the rate of CO washout from venous blood both under air and under 100% O$_2$ changes over time and is not monoeponential. Using the extensive dataset from Benignus and colleagues (1), we were able to fit regression lines to the initial and final segments of the washout curves and to compare these slopes for each of the 15 subjects. The highly significant difference ($P < 0.0001$) between these two washout slopes under conditions in which ventilation was held constant for each subject clearly demonstrates that the initial washout phase is more rapid than the later phase. Our observation that the second half time can be considerably longer than the first, together with model predictions that washout from muscle lags behind washout from blood, emphasizes the potential need to continue therapy well beyond the time at which %COHb has returned to baseline.

A biphasic washout curve is consistent with our model predictions that CO can be taken up by the muscle compartment over a prolonged period of time, even beyond the end of the exposure. The muscle compartment accounts for $\sim 41\%$ of total body mass in young, adult men (7) and is, therefore, a potentially large storage site for CO. With the assumption of a value of 4.7 mg myoglobin (Mb)/g wet weight of muscle (10), the muscle compartment of a 70-kg man contains $\sim 135$ g of Mb, each molecule of which contains a heme group capable of binding CO, and could bind up to 178 ml CO.

Although our model is limited because its generic muscle and nonmuscle compartments do not represent specific anatomical or physiological distribution spaces for CO, nonetheless important inferences can be drawn about the movements of CO between vascular and extravascular spaces. Our model predictions of CO flux to the muscle provide new insights into the extent to which CO is taken up by muscle, the time course over which this uptake occurs both during and after the exposure, and the effects on %COMb of washout under air vs. washout under 100% O$_2$. During the exposure, inhaled CO was taken up by the blood and distributed to rapidly and slowly perfused vascular compartments as described by Smith et al. (20) and Benignus et al. (1), who proposed that the lag between the peak arterial COHb and peak venous COHb was due to CO mixing in slowly perfused vascular compartments. Our model predicts, in addition, that CO enters the muscle and nonmuscle compartments during exposure even to low CO levels and continues to be taken up by muscle well beyond the end of the exposure, in apparent contradiction to an earlier report by Luomanmaki and Coburn (9), who proposed that CO equilibrates rapidly with extravascular tissue. Although our model predictions do not support their conclusions, these predictions are, in fact, consistent with most, if not all, of their results. Their observation that the initial rate of exponential decrease in radioactivity of $^{51}$Cr red blood cells ($t_{1/2} = 14.5$ min) did not differ significantly from that for inhaled $^{14}$CO ($t_{1/2} = 12.5$ min) appears to reflect vascular mixing, which would be expected to be similar for CO and red blood cells. These results do not, however, preclude slow CO uptake by the muscle compartment beyond this initial period.

In another experiment, Luomanmaki and Coburn (9) concluded that equilibration of CO between blood and extravascular stores was rapid on the basis of their observation that the administration of CO at a constant rate over 3 h resulted in a linear increase in COHb. However, our model predicts a linear increase of COHb under these conditions at any value of $D_{mCO}$. Also open to an alternative interpretation are the results of their experiment in which CO was observed to shift "rapidly" out of the blood only when venous PO$_2$ fell below 35–40 Torr (9). However, it is likely that the concomitant fall in tissue PO$_2$ caused a rapid shift in CO from blood to extravascular compartments because severe hypoxia 1) disrupts the balance between O$_2$Mb and COMb, increasing the flow of CO from capillaries to extravascular tissue; 2) recruits muscle capillaries, effectively increasing the surface area for gas exchange and increasing the $D_{mCO}$; and 3) increases cardiac output.

Further evidence that persuaded these authors that CO equilibrates rapidly with muscle was the observed steady decline in blood $^{14}$CO concentration over a 10-h period in anesthetized dogs on a rebreathing system (9). Although the actual rate of decline was difficult to discern, given the considerable scatter of the data presented, the disappearance of CO from the blood was thought by Luomanmaki and Coburn (9) to be due to oxidation of CO to CO$_2$ as opposed to the slow uptake of CO by the muscle compartment. However, results of a similar study involving human subjects maintained on a rebreathing system after exposure to CO appear to contradict this interpretation. Burge and Skinner (3) observed a loss of 3.7 ml of CO from the blood of a male subject from 10–40 min postexposure, during which time the endogenous production of CO would have been $\sim 0.21$ ml (4). Although it is not possible to determine precisely how much of the CO body burden was oxidized to CO$_2$ in this subject, on the basis of Luomanmaki and Coburn’s conclusions that endogenous CO production, in fact, exceeds oxidation of CO to CO$_2$ at COHb levels <10%, it seems reasonable to conclude that most, if not all, of the 3.7 ml of CO lost from the blood from 10–40 min postexposure was, in fact, taken up by the muscle compartment.
In further support of our model predictions that CO in the blood does not equilibrate rapidly with the muscle compartment in human subjects, Roughton and Root (17) compared the amount of CO lost from the blood with the amount found in the expired air. For two male subjects, ~75% of CO lost from the blood from 10–40 min postexposure was accounted for in the expired air. In one of these subjects, 82% of the CO lost from the blood was accounted for in the expired air from 100–130 min postexposure, suggesting a slower but continued uptake by muscle long after the end of the exposure. In separate but similar experiments in which CO washout occurred under muscle long after the end of the exposure. In separate but similar experiments in which CO washout occurred under 100% O2, the mean percent recovery (n = 3) after 4 h was 95.6% of the CO dose administered, suggesting that the CO loss from the blood in these subjects was not due to oxidation of CO to CO2. Consistent with model predictions of CO uptake by muscle during the washout phase, CO did not wash out of the blood as rapidly when DmCO was set to zero as was the case when DmCO was set to the default value of 1.75 ml·min⁻¹·Torr⁻¹ and CO could leave the blood via two routes, the lungs and the muscle. For DmCO set at zero, washout half time increased, providing additional support for the concept that the observed decrease in COHb during washout is the net result of CO loss to the muscle compartment and to the alveolar air. Washout was also biphasic when DmCO was set to zero, although the slopes of the first and second regression lines were more nearly equal, indicating that although uptake and release of CO by the muscle compartment is a major factor in the shape of the washout curve, there are other contributing factors as well. The value for the effective blood-muscle tissue diffusion coefficient for CO (DmCO, 1.75 ml·min⁻¹·Torr⁻¹) used in these studies is similar to the corresponding effective O2 diffusion coefficient (1.51 ml·min⁻¹·Torr⁻¹) implicit in the modeling assumption that O2 flux matches O2 metabolism. The large difference between O2 flux and CO flux is due to the very small pressure gradient driving CO flux (typically a few tenths of a Torr) compared with the pressure gradient of tens of Torr driving O2 flux. Our DmCO value of 1.75 was chosen so that our model would quantitatively reproduce the slow loss of CO from the blood observed in the experiments of Burge and Skinner (3). Rapid equilibration of CO with extravascular muscle and nonmuscle tissue can only occur in our model when DmCO is set to much higher values, e.g., >50.

Our model also provided a means of predicting COHb values at times when experimental values were not available. Our calculated half times should be nearly independent of the method we used to fit the data and interpolate COHb values, so long as the fits are as good as those we achieved. Our interpretation of these data, however, is model dependent and should be considered as a hypothesis that needs to be tested experimentally. At the present time, however, experimental validation is limited by the lack of data to validate the predicted CO content of extravascular tissues. The studies of Roughton and Root (17) and Burge and Skinner (3), however, may be considered as indirect experiential validation of this hypothesis.

In summary, we have provided compelling evidence that the rate of CO washout under room air was faster immediately postexposure than later, resulting in a washout curve that was not monoexponential. Slow equilibration between vascular and extravascular compartments is a major contributor to the changing washout rates. Model simulations indicated that washout under 100% O2 was not monoexponential either. In addition, although our model simulations have demonstrated that peak COHb level, exposure duration and concentration, and the time at which COHb samples are obtained can influence calculated half-time values, these model simulations have also shown that individual variability is at least as important a factor in determining washout characteristics. A better understanding of the physiological factors involved in the uptake and washout processes will be critical to improving treatment strategies for the CO-poisoned patient.

APPENDIX

Parameter Estimation Procedure

The four parameters (VAwi, VAwo, QFm, VFm) were estimated by fitting the model to data from individual subjects from the study of Benignus et al. (1). Parameters were estimated by using an iterative method based on minimizing the mean squared error between the model predictions and the data points. Iterations were governed by a simplex algorithm that terminated when no parameter increment from the simplex algorithm reduced the error function by more than a specified threshold. Because arterial COHb values were not available beyond the first 16 min of the study, because the arterial and venous values were very similar from 10 to 16 min, and because the face mask was removed from each subject at 10 min, we performed the parameter estimation in a sequence of steps. First, we assumed that ventilation was likely to have changed when the face mask was removed 5 min after the end of the exposure; therefore, ventilation during the first 5 min of washout was assumed to equal that during the 5 min of CO exposure. In the first step QFm was set to a typical value (0.167) from our previous study (2), and VAwi and VFm were estimated by fitting the model to data from the first 10 min of the experimental protocol (VAwo is irrelevant during this time.) The error function was based on differences between model predictions and data for both arterial and venous COHb values. Second, VAwi was set to the value just estimated and the remaining three parameters were estimated by fitting the model to the entire experimental protocol (i.e., exposure and washout). The error function now included additional terms related to errors between predicted and measured venous COHb levels during washout at times (24–264 min) for which no arterial COHb data were available. If the new estimate of QFm was within 10% of 0.167, then the three estimated parameters from this step plus the value of VAwi from the first step were selected as the parameter values for the subject. Usually, however, further steps were required. If necessary, the third step comprised reestimation of VAwi, as in the first step, but with QFm and VFm set to their values from step 2. If the result of this reestimation was a lower value of the error function than found in the first step, then the parameters from step 3 were deemed to be more nearly optimal than those from step 1. In a fourth step, VAwi and QFm were set at their values from step 3, and VAwo and VFm were reestimated as in step 2 (i.e., by fitting to both exposure and washout). If VFm did not change by more than 10% from its previously estimated value, and if the error function from step 4 was lower than that from step 3, then the values from step 4 were selected as the parameters for the subject. For a few subjects a third iteration between fitting exposure data and fitting the whole protocol was required to achieve reproducible parameter estimates and minimize the error function. Although this iterative approach is theoretically suboptimal, we found that it performed better than the standard approach of simultaneously fitting all parameters using one inclusive error function.

ACKNOWLEDGMENTS

The authors thank Dr. Vernon Benignus for providing both data published in his paper (1) and unpublished measurements of parameter values from his subjects.
GRANTS

This work was supported by Grant no. NS-050289 from the National Institute of Neurological Disorders and Stroke.

REFERENCES