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| 13. ABSTRACT (Maximum 200 words) The amount of O ₂ available to tissues is essentially the product of cardiac output, [Hb], and O ₂ saturation. Saturation depends on PO ₂ and the O ₂ Hb dissociation curve. With altitude, increased [2,3-DPG] shifts the dissociation curve rightward, but hypocapnia and alkalosis move it leftward. We determined both standard and in vivo P50 in 5 fit subjects decompressed over 42 days in an altitude chamber to the equivalent of the Mt. Everest summit (Operation Everest II). Arterial and venous blood was sampled at five "altitudes" (PB = 760, 429, 347, 282, 253 mmHg), and PO ₂ , PCO ₂ , pH, O ₂ saturation, [Hb] and [2,3-DPG] were measured. As reported previously, 2,3-DPG levels increased from 1.7 (PB = 760) to 3.8 mmol/L (PB = 282). Standard P50 also increased (from 28.2 mmHg at sea level to 33.1 on the summit, p < 0.001). Alone, this would have lowered saturation by 12 percentage points at a summit arterial PO ₂ of 30 mmHg. However, in vivo P50 remained between 26 and 27 mmHg throughout due to progressive hypocapnia and alkalosis. Calculations suggest that the increase in standard P50 did not affect summit VO ₂ MAX, alveolar, arterial and venous PO ₂ 's, but reduced arterial and venous O ₂ saturations by 8.4 and 17.4 points, respectively, and increased O ₂ extraction by 7.9 percentage points. Reduced saturation was balanced by increased extraction, resulting in no significant overall O ₂ transport benefit, thus leaving unanswered the question of the purpose of increased [2,3-DPG] concentrations at altitude. | | | |
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Hemoglobin P₅₀ During a Simulated Ascent of Mt. Everest, Operation Everest II

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ABSTRACT

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Key Words: oxygen transport; altitude; exercise; hemoglobin dissociation curve; 2,3-diphosphoglycerate

INTRODUCTION

IN 1985, OPERATION EVEREST II (OE II) was conducted within the USARIEM hypobaric chambers at Natick, Massachusetts (Houston et

al., 1987). In this study, 8 healthy young fit men were gradually decompressed over 42 days from sea level to a pressure equivalent in P_{I_{O₂}} to that on the Everest summit (West, 1983). Blood gas measurements were made at rest and

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during exercise at sea level and at barometric pressures of 429, 347, 282, and 253 mmHg, corresponding to altitudes of approximately 15,000, 20,000, 25,000, and 29,000 ft. Samples were taken from a radial or brachial artery and, except at 15,000 ft, the pulmonary artery as well. From these samples, P_{O₂}, P_{CO₂}, pH, O₂ saturation, Hb concentration, and hematocrit were all measured, along with blood temperature. These data provided the basis for several reports documenting changes in pulmonary gas exchange with simulated ascent, including the effects of exercise (Wagner et al., 1987; Sutton et al., 1988; Cymerman et al., 1989).

What has never been examined from this unique data set is the P₅₀ of hemoglobin (HbP₅₀). As original OE II investigators, the authors have reexamined the raw data and determined both standard and in vivo HbP₅₀ of the subjects as a function of simulated altitude. The present paper reports on the methodology for determining HbP₅₀ from such data sets, the effects of the simulated Everest ascent on that variable, and the calculated consequences for maximal O₂ transport and $\dot{V}_{O_{2MAX}}$. Based on previous work (Lenfant et al., 1968; Mulhausen et al., 1968; Lenfant et al., 1969; Baumann et al., 1971; Lenfant et al., 1971; Gerlach et al., 1972), it was expected that standard HbP₅₀ would increase with increasing altitude [on the basis of increased levels of 2,3-diphosphoglycerate, which rose from 1.7 ± 0.3 mmol/L at P_B = 760 mmHg to 3.8 ± 0.4 mmol/L at P_B = 282 mmHg (Suter et al., 1988)], but the degree of change and consequences for O₂ saturation of the blood were unpredictable at extreme altitude, especially in the face of counterbalancing effects from progressive hyperventilation and alkalosis.

It was found that standard HbP₅₀ increased substantially and significantly from 28.2 mmHg at sea level to 33.1 mmHg on the summit. This increase would be sufficient to cause a fall of 12 percentage points in O₂ saturation at a P_{O₂} of 30 mmHg (at constant pH and P_{CO₂}). However, because of the progressive hypocapnia and alkalosis during ascent, in vivo P₅₀ remained remarkably constant at about 27 mmHg at all simulated altitudes. Theoretical calculations were performed to assess the importance of the increase in standard HbP₅₀ on diffusive loading of O₂ in the lungs, diffusive

unloading of O₂ in the muscles, and on total O₂ transport. They showed that the increase in P₅₀ would not affect maximal O₂ transport, or alveolar, arterial, and venous P_{O₂}, but would substantially reduce arterial and venous O₂ saturations and increase fractional muscle O₂ extraction.

METHODS

Timing of measurements in relation to ascent profile

The ascent profile has been reported previously (Houston et al., 1987) and is somewhat faster than typical field ascents of Everest itself. After sea-level measurements, subjects were decompressed to the equivalent of 15,000 ft (4572 m, P_B = 429 mmHg), and measurements were made at this altitude at days 10 and 11. Subjects then moved to 20,000 ft (6096 m, P_B = 347 mmHg), with measurements over days 18 to 22. They were then taken higher, to 25,000 ft (7620 m, P_B = 282 mmHg) with measurements over days 36 to 40, and on the same day for each subject, further acute decompression to the Everest summit (8840 m, P_B = 253 mmHg) was achieved with a final set of measurements at that time. At every altitude, blood samples were collected at rest and during several incremental steady-state, constant-load exercise levels up to maximal. At all altitudes except 4572 m, samples were collected from both arterial and pulmonary arterial blood. At 4572 m, no pulmonary artery catheter was placed, and so data were available only from the radial artery. Blood samples were placed on ice and locked out to the laboratory adjacent to the chamber where P_{O₂}, P_{CO₂}, pH, O₂ saturation, Hb concentration, and hematocrit were all measured at 37° using standard blood gas electrodes (Radiometer model ABL3) and a co-oximeter (Instrumentation Laboratories model IL 282). Time from collection to measurement was less than 30 min.

Calculation of standard HbP₅₀

Standard P₅₀ is the P_{O₂} corresponding to a saturation of 50% when the blood temperature is 37°, blood P_{CO₂} is 40 mmHg, and blood

pH is 7.40. As expected in the present study, no sample during exercise and only resting arterial samples at sea level, reflected these standard conditions. Furthermore, the P_{O_2} 's of arterial and venous blood were generally far from those corresponding to the actual P_{50} . Accordingly, the following two methods were devised for determining the standard HbP_{50} .

Method 1. First, for each individual blood sample, Kelman's computer algorithms (Kelman, 1966; Kelman, 1967) were used to calculate the "virtual" P_{O_2} from the measured value. By this is meant the P_{O_2} that the actual sample would have shown had temperature been 37°, P_{CO_2} was 40 mmHg, and pH was 7.40. Then, for each sample, the O_2 saturation was calculated for a trial value of the standard HbP_{50} using the virtual P_{O_2} . All samples with O_2 saturations below 15% and above 85% were excluded from the subsequent analysis. This was done to avoid undue influence of errors in data points far from 50% saturation (and where the O_2Hb dissociation curve becomes less steep or even flat). These exclusion bounds were applied at all altitudes to all subjects without prior inspection of data.

The difference between this calculated saturation and the directly measured O_2 saturation was noted, squared, and summed over all samples included from an individual subject separately at each altitude (except at both 25,000 and 29,000 ft, where blood gas data were combined, because the samples at these two altitudes were taken within 1 h of each other). Then, still for each subject separately, the unique HbP_{50} was found that produced the best fit (least squares) between the measured and calculated O_2 saturations across the entire set of samples at each altitude.

Method 2. A conceptually similar approach was used except that, after using Kelman's algorithms to calculate virtual P_{O_2} values, the Hill equation was used to compute the best-fit standard P_{50} . The Hill equation is

$$\text{Saturation} = \frac{P_{O_2}^n}{P_{O_2}^n + P_{50}^n}$$

where n is the Hill coefficient. After rearranging terms and then taking logarithms, this equation can be expressed as follows:

$$\text{LOG} \frac{\text{saturation}}{(1 - \text{saturation})} = n \times \text{LOG}[P_{O_2}] - n \times \text{LOG}[P_{50}]$$

Using standard linear regression, where $Y = \text{LOG}[\text{saturation}/(1 - \text{saturation})]$ and $X = \text{LOG}[P_{O_2}]$, the values of n and P_{50} are readily obtained from the slope and intercept of the regression line between Y and X .

Calculation of in vivo HbP_{50} and of the effects of changes in P_{50} on O_2 saturation

Using the standard P_{50} determined as above, Kelman's algorithms were invoked to determine the P_{O_2} that would produce 50% O_2 saturation of arterial blood under the actual, measured conditions (i.e., actual temperature, P_{CO_2} , and pH) at rest for each subject at each altitude.

Hypothetical effects of the changes in standard P_{50} with altitude on arterial and venous O_2 saturation were then evaluated at the measured blood P_{O_2} values to assess the physiological importance of P_{50} at altitude. Again, Kelman's algorithms were used, comparing calculated saturations using the sea-level standard P_{50} and the summit standard P_{50} . In these calculations, the actual temperature, P_{CO_2} , and pH values were utilized.

Finally, using reported numerical analysis methods (Wagner, 1996), the consequences of the changes in HbP_{50} were examined for diffusive uptake of O_2 in the lungs and diffusive unloading of O_2 in the muscles, for systemic O_2 delivery, and for maximal \dot{V}_{O_2} . The data needed to conduct these calculations were the same as used in the above analysis (Wagner, 1996) from OE II.

Statistical analysis

Data are presented as mean \pm standard error of the mean. Standard error was chosen rather than standard deviation because the interest is in group mean outcomes. With 5 subjects, standard deviation is 2.236 greater than standard error. Repeated measures ANOVA was used to determine the significance of the

changes in P₅₀ with altitude, and a *p* value of 0.05 or less was taken to reflect significance.

RESULTS

Although 8 subjects began the ascent, only 5 provided data at all altitudes. Since the present analysis focuses on changes in P₅₀ from sea level to altitude, it is important to include data from only those subjects who provided sam-

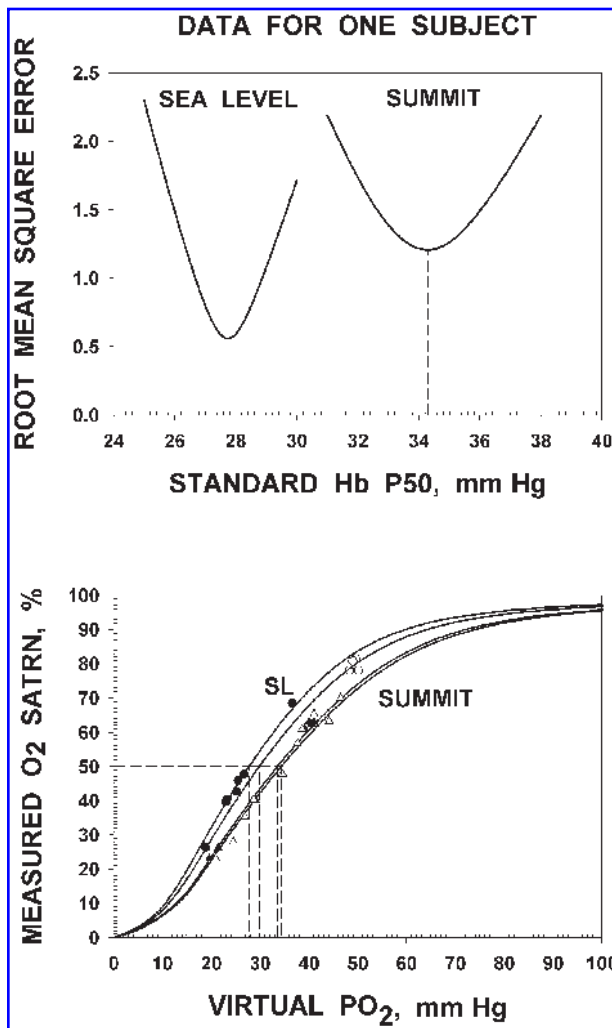


FIG. 1. Top: Identification of standard P₅₀ by the P_{O₂} associated with minimal root mean square error between measured and calculated O₂ saturations in one subject at two altitudes. Bottom: Data at four altitudes from the same subject as in the top panel, plotted as saturation-P_{O₂} pairs, showing the dissociation curve of best fit at each altitude: sea level, closed circles; 429 mmHg, open circles; 347 mmHg, closed triangles; 282/253 mmHg, open triangles. Dashed lines show P₅₀ increasing from 27.7 to 34.3 mmHg.

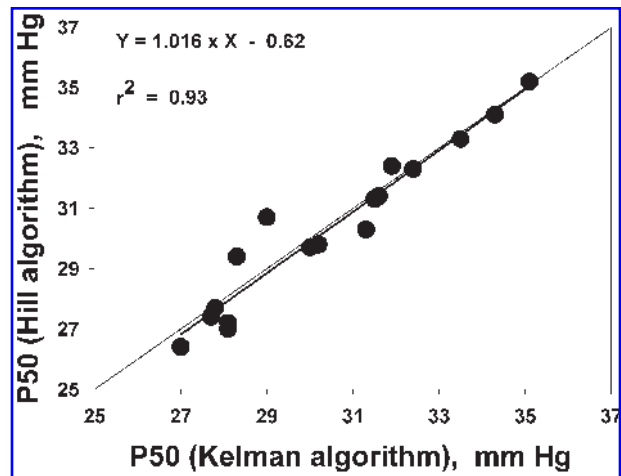


FIG. 2. Agreement ($r^2 = 0.93$, data on the line of identity) between the two methods for estimating standard P₅₀, one using the Hill equation (ordinate) and the other using the Kelman algorithms. Each data point represents one subject at one altitude.

ples at all altitudes. Thus the number of subjects whose data form the basis of this paper is 5.

Method of determination of standard HbP₅₀

The upper panel of Fig. 1 shows the relationship between the sum of squares of the differences between measured and calculated saturations for one representative subject at sea level and at the summit. The ordinate actually expresses the square root of the sum of squares divided by the number of samples used in the calculation and indicates the robust nature of this approach. The lower panel of this figure plots the individual data points for the same subject across all altitudes against O₂Hb dissociation curves of the corresponding least-squares best-fit P₅₀ values. As the lower panel shows, the potential effects of this rightward shift are substantial. For example, at an arterial P_{O₂} of 30 mmHg, similar to values measured on the summit (Sutton et al., 1988), a rightward shift of this magnitude would lead to a 12 percentage point reduction in saturation, from ~55% to ~43% in this case. This would in turn reduce arterial O₂ concentration by about 22%.

Figure 2 shows the relationship between standard P₅₀ values determined by the two different methods. While 5 subjects were studied at five altitudes, only 17 points appear in the figure. This is for two separate reasons. First,

at 15,000 ft, in the absence of venous blood samples, data sets from 3 subjects were associated with saturations that differed so little between rest and exercise within each subject that the Hill linear regression method failed to work. Second, the data at 25,000 and 29,000 ft were combined because the blood samples at these two altitudes were taken within about 1 h of each other. The figure shows that the two methods produced the same values, with the regression producing a relationship not different from the identity line and with an r^2 value of 0.93.

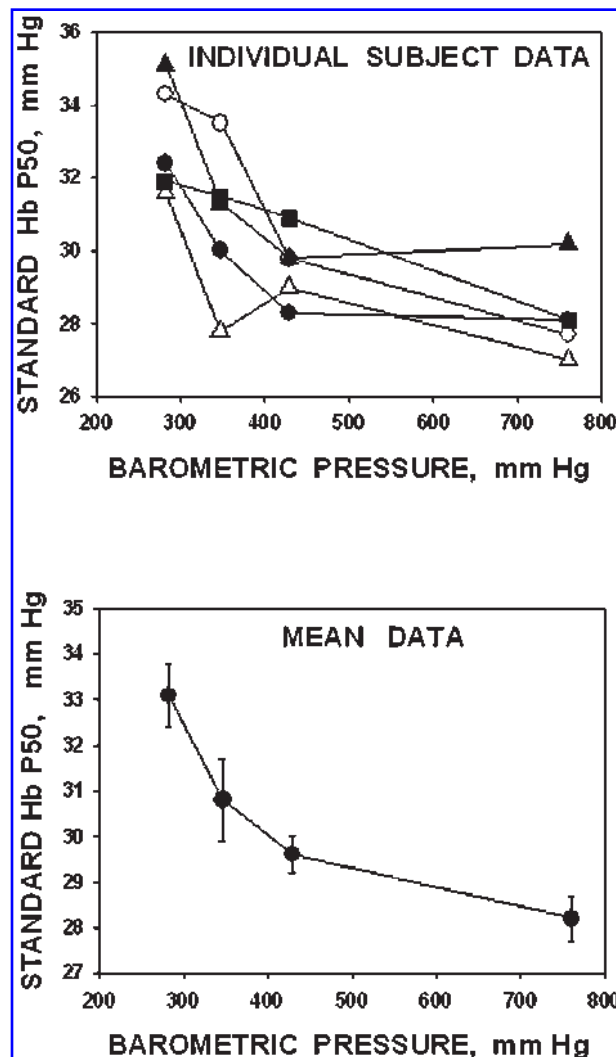


FIG. 3. Standard P_{50} increasing with progressive simulated ascent indicated by barometric pressure. Top panel shows individual data; bottom panel group shows mean data, with P_{50} increasing from 28.2 to 33.1 mmHg, $p < 0.001$.

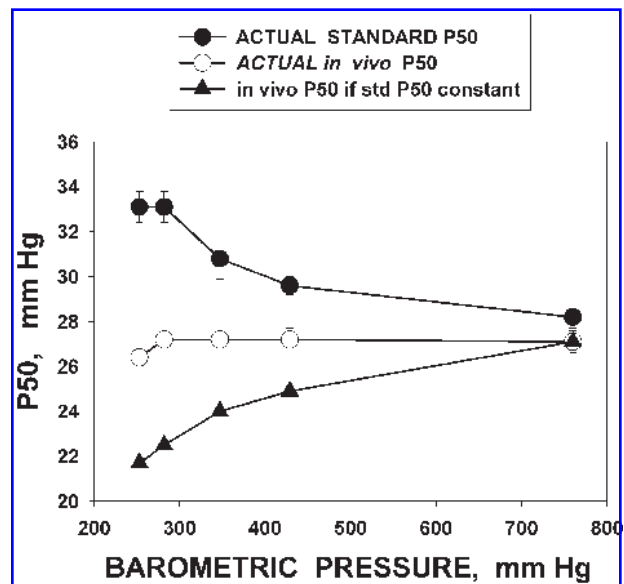


FIG. 4. P_{50} changes with altitude: Closed circles show standard P_{50} increasing with ascent; open circles show in vivo P_{50} that takes into account the progressive alkalosis and hypocapnia with continuing ascent. In vivo P_{50} is independent of altitude due to the counterbalancing effects of rightward- and leftward-shifting influences. Closed triangles show how in vivo P_{50} would change if standard P_{50} had not increased with ascent.

Standard Hb P_{50} results

Figure 3 shows the standard Hb P_{50} values (method 1) for all 5 subjects at all altitudes. The upper panel displays individual data and the lower panel the mean data for the 5 subjects. There is a progressive and substantial rise in standard P_{50} from 28.2 to 33.1 mmHg as barometric pressure falls from 760 to 282 mmHg ($F = 16.0$, $df = (3, 12)$, $p < 0.001$).

In vivo Hb P_{50} results

Figure 4 compares the standard Hb P_{50} (from Fig. 3, closed circles) to the in vivo P_{50} values that take into account temperature, P_{CO_2} and pH (open circles). In vivo P_{50} remains independent of altitude at between 26 and 27 mmHg. Also presented in Fig. 4 are hypothetical values for in vivo P_{50} had the standard P_{50} not risen during ascent (closed triangles). There would have been a substantial fall to about 22 mmHg at the highest altitudes caused by the progressive alkalosis and hypocapnia.

Figure 5 shows resting arterial P_{CO_2} and pH expressed at body temperature for the 5 sub-

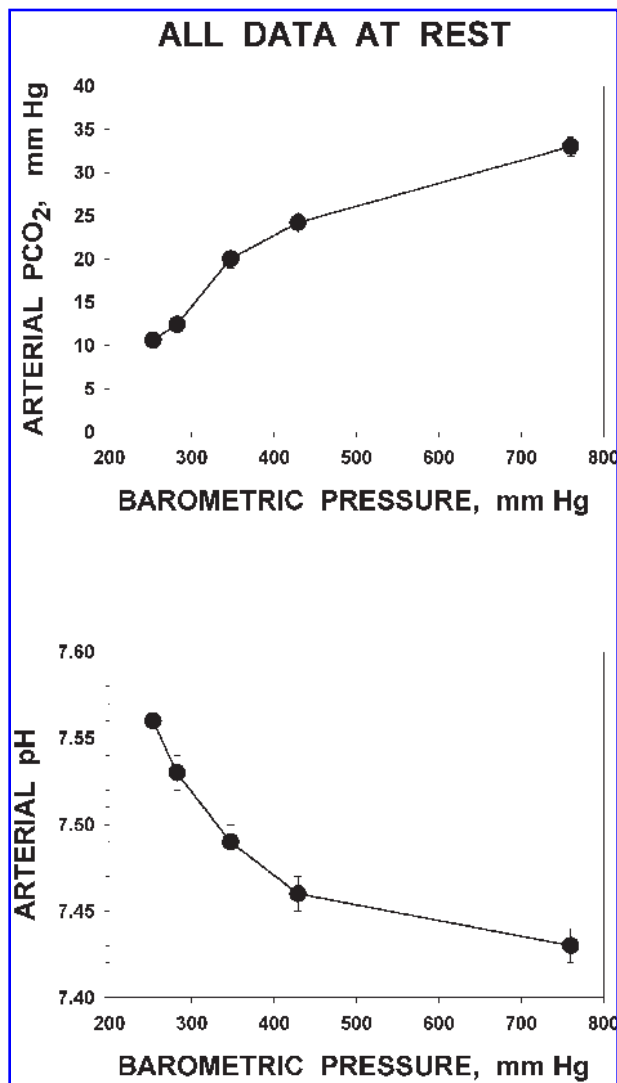


FIG. 5. Progressive fall in arterial P_{CO_2} and rise in arterial pH with continued ascent underlying the rightward-shifting effects of hyperventilation at altitude.

jects at all altitudes to explain the substantial effect of hyperventilation and alkalosis on in vivo P_{50} . The changes with altitude are, as expected, highly significant ($F > 29$, $df = (4, 16)$, $p < 0.0001$).

For conditions of maximal exercise on the summit, Table 1 shows the calculated values for alveolar, arterial, and venous blood gas variables under two circumstances: with the actual sea-level standard P_{50} of 28.2 mmHg and with the actual summit standard P_{50} of 33.1 mmHg. For these calculations, arterial P_{CO_2} was taken to be 9 mmHg and base excess to be -10 meq/L. Blood [Hb] was 18.0 g/dL, cardiac output 16 L/min, alveolar ventilation 165

L/min BTPS, lung diffusing capacity 100 mL/(min/mmHg) and muscle diffusing capacity 60 mL/(min/mmHg), all as listed previously (Wagner, 1996).

Several differences and some similarities can be seen. The similarities are in alveolar, arterial, and venous O_2 tensions and in maximal \dot{V}_{O_2} . The differences are in O_2 saturation of arterial and venous blood, in systemic O_2 delivery, and in muscle O_2 extraction. In essence, lower arterial O_2 saturation is compensated for by greater muscle O_2 extraction.

DISCUSSION

Methodology for determining P_{50}

There are several ways of measuring Hb P_{50} . One involves inscribing the entire O_2 dissociation curve under standard conditions (temperature 37°, P_{CO_2} 40 mmHg, pH 7.40). This was achieved by Imai et al (Imai et al., 1970), but the device was not available during OE II in 1985 when the blood samples were taken. Another method is to mix equal volumes of completely deoxygenated and completely oxygenated blood prepared from a single divided sample from the same subject, keeping both aliquots at standard temperature, P_{CO_2} , and pH. The P_{O_2} of the mixed blood is the P_{50} because the saturation must, by mass conservation, be 50%. This requires tonometry with special gas mixtures to both oxygenate and deoxygenate blood and keep P_{CO_2} at 40 mmHg and pH at 7.40. It is hard to completely rid blood of all O_2 , and fully oxygenated blood has significant amounts of dissolved O_2 that has to be taken into account. Both of these approaches use single blood samples.

The alternative approach, as used here, is to apply accepted algorithms (Kelman, 1966; Kelman, 1967) that correct P_{O_2} for temperature, P_{CO_2} and pH when different from standard conditions. Then, by using all available already-measured blood samples analyzed together, on the hypothesis that on a given day standard P_{50} is constant between rest and exercise, between arterial and venous blood, and as temperature, pH, and P_{CO_2} change, a least-squares, best-fit approach can identify

TABLE 1. CALCULATED DIFFERENCES IN O₂ TRANSPORT AND UTILIZATION AT MAXIMAL EXERCISE ON THE SUMMIT AS A FUNCTION OF P₅₀

| Variable | Standard P ₅₀ (28.2 mmHg) | Standard P ₅₀ (33.1 mmHg) | Ratio |
|---|---|---|-------|
| Alveolar PO ₂ , mmHg | 34.6 | 34.7 | 1.00 |
| Arterial PO ₂ , mmHg | 25.4 | 25.8 | 1.02 |
| Venous PO ₂ , mmHg | 17.9 | 17.1 | 0.96 |
| Arterial O ₂ saturation, % | 64.5 | 56.1 | 0.87 |
| Venous O ₂ saturation, % | 36.9 | 19.5 | 0.53 |
| Systemic O ₂ delivery, L/min | 2.59 | 2.26 | 0.87 |
| Systemic O ₂ extraction, % | 57.1 | 65.0 | 1.13 |
| Maximal $\dot{V}O_2$, L/min | 1.48 | 1.47 | 0.99 |

the single P₅₀ for which the calculated O₂ saturations differ least from the measured values from the same blood samples. By using information from many samples, random error is intrinsically less than in either of the above two approaches, which depend on a single sample. Figure 1 shows the robustness of this method and the reasonableness of the assumptions laid out above. At each altitude, the P_{O₂}-saturation pairs do lie closely on a single dissociation curve. The minimum value of the parabolic relationships in the top panel represent the per sample mean difference between calculated and measured saturation. In these two examples, that difference is just 0.5 percentage point of saturation at sea level and 1.1 percentage points at altitude. Thus, this method adds something valuable to the analysis of blood gas measurements in general—an estimate of reproducibility of the integrated electrode plus co-oximeter system.

Why is standard HbP₅₀ increased with altitude?

This question is posed at two levels: What is the biochemical basis of the rightward shift, and what is the (presumed) physiological advantage to the organism from a rightward shift on ascent to altitude?

The biochemical basis is most likely the result of an increase in 2,3-diphosphoglycerate [2,3-DPG] levels induced by hypoxia (Chanutin and Curnish, 1967; Lenfant et al., 1969; Boushel et al., 2000). The 2,3-DPG levels were measured at three elevations in OE II: P_B = 760, 347, and 282 mmHg; and the values were 1.7 ± 0.3, 3.0 ± 0.2, and 3.8 ± 0.4 mmol/L, respectively

(Sutton et al., 1988). Our results therefore add to the already strong evidence that 2,3-DPG levels increase with altitude and are responsible for the increase in standard P₅₀ (Lenfant et al., 1968; Lenfant et al., 1969; Lenfant et al., 1971). Other than from increased temperature, increased P_{CO₂}, or reduced pH, rightward shifting of the O₂ dissociation curve is known to occur only from increased levels of [2,3-DPG]. Because it is standard P₅₀ that increased in the present study, changes in temperature, P_{CO₂}, or pH cannot be invoked as an explanation.

The question of what biological advantage the substantial (5 mmHg) increase in standard P₅₀ might convey to the organism is more vexing. Conventional arguments state that at altitude a leftward shift enhances diffusive loading of O₂ from alveolar gas into the pulmonary capillaries in the lungs (Bencowitz et al., 1982). This is because during the diffusive process a more hyperbolic, less linear dissociation curve enables large increases in blood O₂ content without large increases in P_{O₂}. Maintaining a low blood P_{O₂} in turn favors diffusive loading by preserving the gradient in P_{O₂} between alveolar gas and the capillaries. Thus a rightward shift would impair O₂ uptake by the lungs.

Similar conventional reasoning suggests that in the skeletal muscles a rightward shift enhances O₂ unloading (West, 1995). Here a similar argument is used in reverse: blood P_{O₂} can be kept high, enhancing diffusion to mitochondria, despite reducing saturation (i.e., increasing off-loading) as the curve shifts rightward. The logical dilemma here is that standard P₅₀ does not oscillate between arterial and venous blood. As a result, what is

“gained” in the muscles is “lost” in the lungs as the dissociation curves shifts rightward (and vice versa should the standard P_{50} be reduced). Indeed, previous calculations (Wagner, 1997) have shown this counterbalance, and the specific application of these principles to the present situation confirms the futility of a change in standard P_{50} as a tactic for enhancing overall, integrated O_2 transport through the whole O_2 pathway (lungs, heart and circulation, blood and tissues). In the present situation, the increase in standard P_{50} has led to lower saturation of arterial and venous blood and greater fractional O_2 extraction by the muscles, such that maximal O_2 transport overall remained unaffected (Table 1).

Perhaps the increase in 2,3-DPG levels on ascent is one of several other possible examples of well-known physiological responses to cellular hypoxia whose primary purpose may be to respond to a different stress. More explicitly, the polycythemia of altitude has most commonly been shown not to materially enhance exercise capacity (Winslow et al., 1981). It would be easier to argue that this erythropoietic hypoxic response evolved over time to stimulate red cell production in response to cellular hypoxia following anemia (nutritional or traumatic) at low altitude, rather than as a response to hypoxia at altitude. Rebuilding red cell mass that has been diminished by nutritional or traumatic factors would seem to be a rational and specific restorative response. The altitude response may therefore be essentially an epiphenomenon. Similar arguments have been made about hypoxic pulmonary vasoconstriction: its primary purpose may be to diminish pulmonary blood flow during fetal development (West, 1995). Its postnatal activity may be again an epiphenomenon. Here this is even easier to imagine because the effect of vasoconstriction is of course pulmonary hypertension, which places a load on the right heart, hardly an advantage to the organism. We already know that the ability of hypoxic vasoconstriction to restore ventilation–perfusion relationships in the lung, another suggested reason for having this response, is far from perfect. Studies comparing \dot{V}_A/\dot{Q} relationships breathing air and O_2 do show an effect of vasoconstriction that improves these relationships, but

the effect is small, and the fact remains that most patients with lung disease show severe \dot{V}_A/\dot{Q} inequality despite the presence of hypoxic vasoconstriction (Wagner et al., 1975; Wagner, 1977).

To return to the possible evolutionary role for [2,3-DPG], diffusion in the lung at rest in normoxia is a process with a large amount of excess capacity. Thus, capillary P_{O_2} equilibrates with that in alveolar gas in just one-third of the available transit time in normal lungs (Wagner and West, 1972). Even during maximal exercise, normal subjects, other than trained athletes, are usually not diffusion limited in the lungs, and even then only about 50% of trained athletes have evident exercise-induced hypoxemia (Dempsey et al., 1984; Powers et al., 1988). In contrast, there is good evidence that muscle O_2 unloading during normoxic exercise is usually limited by muscle O_2 diffusive conductance (Roca et al., 1989), since intracellular P_{O_2} is far below that of even muscle venous blood. Thus, enhancing muscle O_2 unloading during normoxic exercise by right-shifting the dissociation curve (when anemia develops from nutritional or traumatic causes) may well enhance $\dot{V}_{O_{2MAX}}$ because there is more diffusive reserve in the lungs to compensate for the negative effects of such a shift on diffusive uptake. It is certainly known that 2,3-DPG levels are increased in anemias (Valeri and Fortier, 1969; Torrance et al., 1970; Lichtman et al., 1971; Boning and Enciso, 1987; Wallis et al., 2005).

Standard versus in vivo P_{50} : Interplay between standard P_{50} , alkalosis, and hypocapnia

It is curious that the progressive rightward shift we saw in the standard dissociation curve with altitude was precisely counterbalanced at every altitude by an equally progressive leftward shift from increasing alkalosis and hypocapnia, as shown in Fig. 4. That these opposing forces are in play has been known for many years (see Winslow et al., 1981; Mairbaurl et al., 1990). Such precise homeostasis usually suggests a regulatory process, which would in this case somehow be mediated by O_2 levels somewhere in the O_2 pathway; but OE II was clearly not designed to investigate whether such is the case or not. Thus, the constancy of

in vivo P_{50} may be either an active result of some regulatory process, or it may simply be a chance observation with little if any functional significance. Given our discussion of the lack of importance of P_{50} on maximal O_2 transport at altitude, one is inclined to think the latter, but the present data cannot help decide the issue.

Comparison with AMREE from sea level to 6300 m

Winslow et al. (1984) made measurements of P_{50} during the 1981 American Medical Research Expedition to Everest (AMREE). It appears that when P_{50} was corrected for pH (that is, corrected from actual pH to 7.40), the corrected values increased by 1.7 mmHg from 28.1 mmHg at sea level to 29.8 mmHg at 6300 m. This is similar to the present findings in which standard P_{50} , 28.2 mmHg at sea level, increased by 2.6 mmHg to 30.8 mmHg at 6100-m equivalent altitude. Both Winslow's and the present data further suggest that this increase in standard P_{50} is counterbalanced by progressive hyperventilation and alkalosis so that in vivo P_{50} remains essentially independent of altitude. Therefore, up to 6300 m, our findings appear essentially similar to those reported by Winslow. This is further supported by the similarity in levels of [2,3-DPG]. Up to 6100 to 6300 m, the highest altitudes for which there are both AMREE and OE II [2,3-DPG] data, there is no difference. Thus, in AMREE, the [2,3-DPG] concentration at sea level was 0.84 mol/mol Hb and at 6300 m it was 1.04 mol/mol Hb (Winslow et al., 1984). Converting to mmol/L blood using Winslow's [Hb] data allows a comparison with OE II values. In these units, the AMREE values become 1.9 and 3.0 mmol/L, respectively. Corresponding values from OE II were 1.7 and 3.0 mmol/L (Sutton et al., 1988).

Comparison with AMREE above 6300 m

Above 6300 m, Winslow found that in the one available subject, estimated summit P_{50} expressed at a pH of 7.4 was only 27.4 mmHg. It is not clear if this estimate is also expressed at a P_{CO_2} of 40 mmHg or corrects only for pH. Given the estimated summit arterial P_{CO_2} of 7.5

mmHg, the P_{CO_2} correction to 40 mmHg (without change in pH) would raise the estimate of P_{50} by 2.6 mmHg. Furthermore, this subject may be an extreme case, since his P_{50} expressed at pH 7.4 was more than 4 mmHg lower than that of the only other subject assessed above 6300 m. It is also substantially lower than the group mean data at 6300 m. If these factors are taken into account, it is possible that the average AMREE subject would have had a standard P_{50} some 6 mmHg higher than 27.4 mmHg, in the region of 33 mmHg, as we found in the present study. Unfortunately, above 6300 m, Winslow's results come from just one (or two) subjects and without arterial blood sampling. Our data come from 5 subjects, each providing multiple arterial and venous samples at all altitudes, and must be considered more reliable and representative.

Winslow also calculated for the single subject that the estimated severe degrees of hypocapnia and alkalosis (P_{CO_2} 7.5 mmHg, pH 7.78) would cause in vivo P_{50} on the summit to fall substantially, to about 19 mmHg. On the other hand, we found no significant change of in vivo P_{50} with altitude, even at the summit (Fig. 4). This is because (1) the standard P_{50} continued to increase in our subjects above 6100 m, and (2) the measured degrees of hypocapnia and alkalosis (P_{CO_2} 10.6 mmHg, pH 7.56) were not as extreme as estimated in AMREE.

The differences between Winslow's and our studies above 6300 m may therefore be explained by several possible factors. First, Winslow's single subject may not have been a typical subject. Second, if correction was made only for pH and not also for P_{CO_2} , standard P_{50} would have been underestimated by Winslow. Third, the ascent profiles of AMREE and OE II were different, with OE II subjects "ascending" more quickly than those in AMREE. AMREE subjects appear to have achieved greater P_{CO_2} reduction and were more alkalotic, which would have together caused a relative leftward shift of the dissociation curve, all other factors equal. This could explain part of the in vivo, but not the standard, P_{50} differences. However, the difference in in vivo P_{50} according to whether P_{CO_2} was 7.5 as in AMREE or 10.6 mmHg as in OE II is only 0.5 mmHg.

Consequences for exercise at altitude

This paper confirms through theoretical analysis that the observed changes in P₅₀ at altitude do not confer increased ability to exercise. It would be difficult, especially in humans, to conduct an interventional study to directly test this hypothesis. Thus the present conclusions await experimental confirmation. However, isolated perfused muscle studies have been conducted wherein standard P₅₀ was increased (Richardson et al., 1998) or decreased (Hogan et al., 1991), and predictable increases and decreases in maximal \dot{V}_{O_2} were recorded at constant blood flow and arterial O₂ delivery. These studies in part confirm the present concept of how shifts in the dissociation curve affect O₂ transport and fit directionally with the OE II data presented herein.

CONCLUSIONS

The principal message would seem to be that, while traditional arguments about how the dissociation curve shifts to the left favor pulmonary O₂ uptake, whereas rightward shifts favor tissue O₂ unloading may be true, any given curve shift will produce counterbalancing effects in the lungs and tissues that lead to little net effect on maximal \dot{V}_{O_2} , especially at altitude. This is the case in the present situation, where the 5-mmHg increase in standard P₅₀ found as subjects ascended from sea level to the equivalent of the Everest summit was calculated to reduce arterial and venous O₂ saturations substantially, but at the same time increase muscle O₂ extraction to preserve maximal O₂ transport.

The individual values for arterial and venous P_{O₂}, P_{CO₂}, pH, O₂ saturation, Hb, and hematocrit are available from the corresponding author by request.

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