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DOES ERYTHROCYTE INFUSION IMPROVE TWO-MILE RUN PERFORMANCE AT
HIGH ALTITUDE?

BY

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ABSTRACT

Effects of autologous erythrocyte infusion (ER) on improving exercise performance at high altitude have not been studied. ER effects on two-mile (3.2 km) run performance were evaluated during acute (AA, 3 days) and chronic (CA, 14 days) exposure to high altitude (4300m) in infused (ER) and control (CON) subjects (Ss) initially matched ($P>0.05$; $n=8$ each group) for age, body size and aerobic fitness. After sea level runs (SL, 50m), unacclimated male Ss received either 700 ml of saline and autologous erythrocytes (42% Hct) (ER) or saline alone (CON). Two-mile run times (min:sec) did not differ ($P>0.05$) between groups at SL ($\bar{X}\pm\text{SE}$: ER, 13:14 \pm 00:19; CON, 13:39 \pm 00:32), or during AA (ER, 19:02 \pm 00:18; CON, 19:44 \pm 00:43) and CA (ER, 17:44 \pm 00:27; CON, 18:45 \pm 00:55), but times were faster ($P<0.05$) comparing SL to AA or CA. Heart rates (HR, $\text{bts}\cdot\text{min}^{-1}$) did not differ between groups at SL (ER, 188 \pm 3; CON, 191 \pm 3), or during AA (ER, 170 \pm 4; CON, 178 \pm 4) and CA (ER, 162 \pm 6; CON, 169 \pm 5), but HR were higher ($P<0.05$) comparing SL to AA or CA. Perceived exertion (local, central, overall) did not differ between groups at SL, AA or CA, but local was lower ($P<0.05$) at SL compared to AA and CA, and overall was lower for SL than AA. ANCOVA (adjusted for SL group run times) revealed 00:14 (AA) and 00:28 (CA) mean improvement trends ($P>0.05$) for ER compared to CON. When compared to the ER literature reporting run time improvement for similar distances at SL or relatively low altitude, the improvement noted during AA was the same, but run time improvement doubled for CA.

Key Words: blood doping; exercise endurance; heart rate; hypoxia; mountainous environment; perceived exertion; 3.2 km run time

INTRODUCTION

Autologous erythrocyte infusion or "blood doping" enhances the capacity for aerobic-type exercise at sea level (21, 22). During competitive races ranging from 1500 m to 10 km, the post-infusion improvement in run time at sea level or relatively low altitude increases as a function of the distance run (2, 3, 7, 27). For instance, the expected improvement in run time at 2, 6, and 10 km is approximately 7, 30, and 68 sec, respectively, following autologous erythrocyte infusion from two units of freeze-preserved blood (22).

When sea-level residents travel to high altitude, arterial oxygen pressure decreases causing oxygen saturation of hemoglobin to fall, in turn, reducing the amount of oxygen in the blood (5, 8). Exercise endurance and maximal aerobic power are degraded at high altitude in proportion to this reduction in arterial oxygen content (9, 21). The ergogenic effects of infusing autologous erythrocytes primarily result from the greater hemoglobin concentration produced which increases arterial oxygen content for any given arterial oxygen pressure and hemoglobin saturation (21, 22). Thus, autologous erythrocyte infusion might be even more ergogenic at high altitude than sea level. The increased hemoglobin concentration resulting from autologous erythrocyte infusion could offset, at least in part, the reduction in oxygen saturation of hemoglobin on arterial oxygen content (21), possibly ameliorating the deficit in physical exercise performance seen when sea-level residents ascend to high altitude.

No studies have evaluated the ergogenic effects of autologous erythrocyte infusion at high altitude. This study compared two-mile run (3.2 km) performance at sea level

(50 m) to performance of this same event after autologous erythrocyte infusion during acute (3 days) and chronic (14 days) exposure to high altitude (4300 m). Comparisons were made between infused and control subjects initially matched for age, body size and aerobic fitness. We hypothesized that autologous erythrocyte infusion would significantly improve two-mile run performance for infused compared to control subjects at high altitude particularly after chronic exposure.

METHODS

Subjects. Sixteen healthy, young Caucasian men who were members of two A teams of the Tenth Special Forces Group at Fort Devens, Ayer, MA served as volunteer subjects after medical screening. After sea-level testing, these men were divided into two groups (n=8 each) with one group (ER) to undergo autologous erythrocyte infusion from two units of freeze-preserved blood and the other group to serve as saline controls (CON). The two groups were matched ($P>0.05$) for age, body size and maximal aerobic power. These two groups of subjects ($P>0.05$, all comparisons) had mean (\pm SD) ages of 31.0 ± 3.1 yr (CON), 29.7 ± 2.1 yr (ER); heights 177.1 ± 6.2 cm (CON), 179.8 ± 8.8 cm (ER); weights 83.2 ± 7.2 kg (CON), 82.1 ± 9.9 kg (ER); DuBois surface area 2.0 ± 0.1 m² (CON), 2.0 ± 0.2 m² (ER); body fats $20.1 \pm 4.9\%$ (CON), $17.2 \pm 3.3\%$ (ER); and, maximal aerobic powers 4.34 ± 0.39 l·min⁻¹ (CON), 4.62 ± 0.70 l·min⁻¹ (ER), or 52.07 ± 4.76 ml·kg⁻¹·min⁻¹ (CON), 54.08 ± 3.46 ml·kg⁻¹·min⁻¹ (ER). All subjects were informed of the potential risks and gave their voluntary written consent. All subjects were life-long sea-level residents and had not been exposed to high altitude for at least a month before these experiments. Blood removal was completed during March and April while the

determination of physical characteristics and actual experiments were conducted during June and July. The experimental protocol and many of the procedures have been described in detail earlier (28).

Protocol. Each subject's percent body fat was determined by hydrostatic weighing and maximal aerobic power ($\dot{V}O_2\text{max}$) determined by a treadmill-running test at sea level (SL, 50 m). The maximal treadmill protocol was progressive in intensity and continuous in nature. The initial treadmill grade was 5% at SL and 0% at high altitude; and, increased by 2.5% increments every 100 sec while subjects ran at $2.68 \text{ m}\cdot\text{s}^{-1}$. Documented criteria (19, 20) were employed for determination of $\dot{V}O_2\text{max}$.

On two separate occasions before experimental testing, subjects had one unit of blood removed by venesection. At least five weeks separated collection of the first and second unit of blood, and at least eight weeks were allowed between the collection of the second unit of blood and the beginning of the experiments. The blood was processed and stored as previously described (24, 25). After the two-mile (3.2 km) runs at SL and twenty-four hours before ascent to high altitude, the ER group received 700 ml of saline and autologous erythrocytes (42% Hct) while the CON group received 700 ml of saline solution. The principal investigator and chief medical monitor were the only staff at this Institute informed as to the type of infusion each subject received.

The two-mile run at SL was performed outdoors on a level, quarter mile (0.4 km) cinder surface track at Fort Devens. Subjects ran in small groups dressed in running clothes and running shoes. All subjects were asked to exert a maximal effort in completing this distance in the fastest possible time. Investigators and staff made

measurements at each half mile (0.8 km) run and provided encouragement to all subjects. Each subject ran twice at SL with the best two-mile run time used for statistical analysis. At high altitude (4300 m), the two-mile run was completed on a fairly level, near quarter mile dirt surface roadway on the summit of Pikes Peak. Subjects were dressed, instructed and encouraged for the altitude runs similar to those at SL. Each subject was evaluated for two-mile performance during acute (AA, 3 days) and chronic (CA, 14 days) exposure to high altitude. Ambient temperature and relative humidity were determined during all runs.

Performance, physiological and perceptual variables. During $\dot{V}O_2$ max tests, an automated system (Sensormedics Horizon MMC) was used to measure $\dot{V}O_2$ employing a Hans Rudolph (no.2700) breathing valve. Performance times for each subject were recorded to the nearest second using calibrated digital stopwatches at the half mile (0.8 km), mile (1.6 km), mile and a half (2.4 km), and two-mile (3.2 km) points. Each subject wore and monitored his heart rate (HR) using a Polar Vantage (XL) wristwatch which displayed HR to the nearest beat. Differentiated ratings (local, central and overall) of perceived exertion (RPE) using the Borg scale (1, 14) were recorded for each subject. In addition, thermal sensation (TS) using a category rating scale was also evaluated (16, 29). Subjects called out their HR, RPE and TS to investigators as they passed each of the points where performance time was recorded.

Statistical analysis. A repeated-measures analysis of variance (ANOVA) was used to compare the performance, physiological and perceptual values. If significant F values were found, critical differences were determined by Tukey's procedure. In some

instances, analysis of covariance (ANCOVA) was employed. Statistical significance was accepted at the $P < 0.05$ level. The data reported represent means \pm SE.

RESULTS

No significant differences ($P > 0.05$) were observed between the ER and CON groups for age, height, weight, surface area, body fat and maximal aerobic power ($l \cdot \text{min}^{-1}$ or $\text{ml} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$). Mean (\pm SE) ambient temperature (T_a) and/or relative humidity (rh) differed ($P < 0.05$) between SL ($T_a = 20.4 \pm 1.0^\circ\text{C}$, $\text{rh} = 69.1 \pm 3.1\%$), AA ($T_a = 11.0 \pm 0.4^\circ\text{C}$, $\text{rh} = 57.7 \pm 3.0\%$), and CA ($T_a = 9.1 \pm 0.6^\circ\text{C}$, $\text{rh} = 71.5 \pm 1.8\%$) two-mile runs. All T_a differences between SL, AA or CA were significant while all rh differences were significant except between SL and CA ($P > 0.05$).

INSERT TABLE 1 ABOUT HERE

Table 1 presents the two-mile run times for the ER and CON groups at SL, AA and CA. Two-mile run times (min: sec) did not differ significantly ($P > 0.05$) between the ER and CON groups at SL, or during AA and CA. However, trends ($P > 0.05$) for faster run times in the ER compared to CON group can be seen at AA and CA. Two-mile run times were faster ($P < 0.05$) at SL than AA or CA. No significant differences were found between groups for half mile (0.8 km), mile (1.6 km), and mile and a half (2.4 km) run times at SL, AA or CA (data not shown). Again, ER ran faster than CON for all three intervals at SL, AA and CA (\bar{X} range, 12 sec at SL to 51 sec faster at CA).

INSERT TABLE 2 ABOUT HERE

Table 2 displays the two-mile HR for the ER and CON groups at SL, AA and CA. Two-mile run HR ($\text{beats} \cdot \text{min}^{-1}$) did not differ ($P > 0.05$) between groups at SL, AA or CA.

However, two-mile run HR were higher ($P < 0.05$) for SL compared to AA or CA. While not shown formally, no differences ($P > 0.05$) in HR were observed between groups at SL, AA or CA for the intermediate distances (1/2 mile, 1 mile or 1-1/2 mile).

INSERT TABLE 3 ABOUT HERE

Table 3 shows the two-mile RPE (local, central and overall) for the ER and CON groups at SL, AA and CA. Local, central and overall RPE did not differ ($P > 0.05$) between ER and CON groups at SL, AA or CA. However, local RPE was lower ($P < 0.05$) at SL compared to AA or CA, overall RPE was lower ($P < 0.05$) at SL contrasted to AA only, and central RPE did not differ ($P > 0.05$) between SL, AA or CA. No differences ($P > 0.05$) in local, central and overall RPE were seen between groups for SL, AA or CA at the other three run intervals.

Thermal sensation did not differ ($P > 0.05$) between the ER and CON groups at SL, AA or CA for any distance. Two-mile run TS (mean \pm SE) also did not differ ($P > 0.05$) between SL (5.9 ± 0.2), AA (5.7 ± 0.3) or CA (5.3 ± 0.3). However, TS was significantly warmer ($P < 0.05$) for SL than CA at the other distances (1/2 mile, 1 mile or 1-1/2 mile).

INSERT FIGURE 1 ABOUT HERE

Figure 1 illustrates the relationship between post-infusion (RBCs from 2 units) cumulative improvement in run time (sec) and various run distances completed (km) at SL or relatively low altitude from four previously published studies, to which we have added our findings at high altitude. In all of the previous studies, subjects served as their own controls in repeated-measures designs. The predicted post-infusion cumulative improvement in run time at 2, 4, 6, 8 and 10 km from these four studies was about 7,

15, 30, 45 and 68 sec, respectively. Our run time data were further analyzed using ANCOVA to adjust for the pre-infusion SL group run time differences. The adjusted mean run times following ANCOVA revealed that the ER compared to CON group was always faster ($P>0.05$) for both AA and CA at 0.8 km (1/2 mile), 1.6 km (1 mile), 2.4 km (1-1/2 mile), and 3.2 km (2 mile). As shown in Figure 1, the mean improvement in run time for the ER compared to CON group during AA was about 8 sec at 0.8 km, 10 sec at 1.6 km, 9 sec at 2.4 km, and 14 sec at 3.2 km, while for CA the improvement between groups was 0.4 sec at 0.8 km, 8 sec at 1.6 km, 16 sec at 2.4 km, and 28 sec at 3.2 km. In general, the trend for improvement in run time between groups was greater for CA than AA particularly as distance increased.

DISCUSSION

Experimental design. All other published studies (2, 3, 7, 27) concerning blood doping and run performance at sea level or relatively low altitude employed some form of a double-blind placebo crossover experimental design. Logistical problems associated with a 14-day sojourn to high altitude precluded that and the present study utilized an experimental design involving infused and control groups ($n=8$ each) of men who were matched ($P>0.05$) for age, body size, morphology and $\dot{V}O_2\text{max}$. Our design allowed us to evenly distribute and control for the effects of these important confounding variables between the two groups. This is the first study to report the performance and physiological effects of autologous erythrocyte infusion on run performance at high altitude.

Run distance selected. The two-mile (3.2 km) run was selected as our

performance measure because it is currently part of the Army Physical Fitness Test (APFT) (10). All U.S. Army soldiers are evaluated against this measure at least semi-annually as part of their APFT; thus, our Special Forces test subjects were quite familiar with our performance test. Two-mile run times have been shown to be highly correlated with $\dot{V}O_2\text{max}$ for both male ($r=-0.91$) and female ($r=-0.89$) soldiers (11). Special Forces soldiers are the most aerobically fit U.S. military unit reported to date (12), and their motivation is without question.

Ergogenic effect. Autologous erythrocyte infusion has been shown to increase hematocrit (Hct), hemoglobin concentration (Hb) and arterial O_2 content (C_aO_2) (4, 13, 17, 18). In the present study, infusion of 700 ml of saline with autologous erythrocytes (42% Hct) resulted in higher ($P<0.05$) arterial Hct (+9%), arterial Hb (+10%), and a smaller decrease ($P<0.05$) in C_aO_2 for the ER compared to CON group during acute exposure to high altitude. These observations are discussed in a companion paper (28). The present findings concerning autologous erythrocyte infusion and run performance at high altitude shall be discussed from two standpoints: (a) the effects of the severe level of hypoxia, and (b) the distance run.

Level of hypoxic stress. This is the first study on the effects of blood doping on persons actually sojourning at high altitude, and only a few studies have evaluated the effects of blood doping on individuals exposed to hypoxia. Pace et al. (13) showed that young men infused with 1000 ml of erythrocytes (Hct +20%) displayed lower HR while treadmill walking in hypoxic conditions (hypobaric chamber at 4712m) when compared to controls walking under the same conditions. These authors (13) concluded that blood

doping could lower one's "physiological" altitude. More recently, Robertson et al. (18) demonstrated that hypoxic-gas breathing simulating exposure to 3566m lowered $\dot{V}O_2$ max half as much in men infused with 750 ml of autologous erythrocytes compared to the reduction in $\dot{V}O_2$ max seen before infusion. Subsequently, Robertson et al. (17) reported that erythrocyte infusion also prevented the reduction in $\dot{V}O_2$ max induced by hypoxic-gas breathing in women.

All those studies (13, 17, 18) simulated altitude conditions using short periods (< 1 hr) in a hypobaric chamber or breathing hypoxic-gas mixtures. In contrast, we studied the effects of autologous erythrocyte infusion on sea-level residents who rapidly ascended to the summit of Pikes Peak (4300m) and remained there for two weeks. On the first day after ascending, our ER subjects exhibited a 25% decline in $\dot{V}O_2$ max compared to SL which did not differ ($P>0.05$) from the 28% decrease seen in the CON (28). While our ER subjects tended to show greater improvement in run time following two weeks of altitude acclimation, two-mile run time at 4300m did not differ ($P>0.05$) between ER and CON groups.

Several interpretations emerged when the findings from these studies are compared. The ergogenic effects of blood doping may diminish as altitude increases. Thus, at sea level, blood doping increases $\dot{V}O_2$ max and improves run time, while at relatively low altitudes (< 2500m) blood doping may prevent the hypoxic-related decrements in $\dot{V}O_2$ max and run time (17). When individuals ascend to moderate altitudes (< 3800m), blood doping may lessen but not completely ameliorate the decline in $\dot{V}O_2$ max and endurance performance (18); whereas, the effects of blood doping on

these same measures may be negligible at higher altitudes. Another possible explanation is that the ergogenic effects are apparent only with very acute hypoxic exposure, and that acclimatization occurring with exposures lasting more than a few hours somehow obviates ergogenic effects. Another interpretation is that the increase in Hb at 4300m for our ER group was smaller than in the studies at lower simulated altitudes where ergogenic effects were shown (17, 18). While this possibility can not be dismissed, inducing larger increments (>10%) in Hct and Hb could be unsafe for individuals ascending to high altitudes due to the hemoconcentration normally occurring with altitude acclimatization.

A final explanation involves limitations to oxygen transport at high altitude which blood doping can not counteract. At high altitude, pulmonary gas exchange is impaired due to the appearance of an alveolar-end-capillary diffusion limitation and a ventilation-perfusion (\dot{V}_A/\dot{Q}) mismatch resulting in an elevated PAO_2-PaO_2 gradient or greater alveolar-arterial PO_2 difference ($AaDO_2$) (6, 23, 26). Wagner et al. (26) reported that at altitudes of 3048 and 4572m (~10,000 and 15,000 ft), the widening of the $AaDO_2$ with increasing exercise intensity was primarily related to a progressive increase of the diffusion limitation which was detectable even during light exercise ($\dot{V}O_2 \sim 15 \text{ ml} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$). These same authors concluded that alveolar-end-capillary diffusion limitation is the most important pulmonary factor interfering with gas exchange during exercise at these high altitudes, and may help explain why blood doping was not more effective at our altitude.

Run distances studied. A second concern involves the effects of distance run on autologous erythrocyte infusion induced improvements in run performance (see Figure

1). Williams et al. (27) were the first to evaluate the improvement in five-mile (8.0 km) run performance at sea level associated with the infusion of 920 ml autologous blood in 12 experienced male distance runners. When compared to a pre-saline trial, a post-saline trial after infusion of 920 ml saline and a pre-blood infusion trial, the mean performance time for the five-mile run was about 45 sec faster ($P < 0.05$) following autologous erythrocyte infusion (Hct +6%, Hb +8%). However, the effects of blood doping on run time were not significant ($P > 0.05$) for miles 1-3 when compared to the other trials. Next, Goforth et al. (7) studied six trained distance runners at sea level after infusion of 760 ml autologous red blood cells (Hct +10%) and reported that the mean three-mile run time "decreased" 23.7 sec (statistical significance not given).

In two published reports, Brien and colleagues (2, 3) investigated the effects of blood doping on the improvement in run performance at either 10 km (6.25 miles) or 1.5 km (~0.9 mile). Both studies were conducted in Albuquerque, NM at an elevation of 1800m and all subjects were acclimatized residents of this relatively low altitude. At 10 km, Brien and Simon (3) evaluated six male distance runners before infusion, after infusion of 100 ml of saline, and after infusion of 400 ml of autologous erythrocytes (Hct +5%). After blood doping, the mean performance time was 69 sec faster at 10 km compared to the other two trials ($P < 0.05$). For performance over one metric mile, Brien et al. (2) also studied four male distance runners for similar trials as outlined above (blood doping: Hct +10%, Hb +7%) and reported a significant 4.5 sec improvement in run time associated with infusion ($P < 0.05$). Their data was available in the report; however, our Institute statistician could not duplicate their statistical findings. We conclude that

run performance clearly improved following autologous erythrocyte infusion from two units of freeze-preserved blood at distances equal to or greater than five miles (8 km) but the improvements reported at shorter distances are questionable.

Our data show 14 and 28 sec faster mean two-mile run times for the ER compared to CON group during acute and chronic exposure to high altitude, respectively. In addition, the ER compared to CON group always ran faster on the average for all half-mile intervals of this two-mile run during both acute and chronic trials. However, none of the differences between groups were statistically significant. Even though trends for improvements associated with blood doping at altitude are evident, the distance run may have been too short to demonstrate differences significant for statistical purposes. In comparing our data with those from other studies involving blood doping and improvement in run performance (2, 3, 7, 27), the residual errors from the various ANOVAs were seen to decrease in comparison to the mean squared deviations as run distance increased. Thus, longer distances run tended to become statistically different in terms of the ergogenic effects of blood doping.

Environmental effects. A final concern involves the differences in the environmental conditions (T_a and rh) observed between the sea level and two altitude runs. While statistically significant, we do not believe that these environmental differences profoundly affected overall performance during these runs or our interpretations of the findings. Most important, the environmental conditions and thermal sensations were the same between groups for each run and for all of the distance intervals. Secondly, the avenues of heat exchange associated with thermoregulation are

not severely challenged until T_a rises from temperate ($\sim 22^\circ\text{C}$) to hotter conditions at the exercise intensity involved in these runs (15).

Conclusions. When compared to the autologous erythrocyte infusion literature on run time improvement for similar distances at sea level and relatively low altitude, run time improvement during acute altitude exposure was the same, but run time improvement doubled after chronic altitude exposure in the present study. ANCOVA (adjusted for sea level run times) showed 14 (acute) and 28 sec (chronic altitude) mean improvement trends ($P > 0.05$) for our ER compared to CON subjects. In conclusion, we suggest that blood doping effects may be found significant with either a longer running distance (> 5 miles) and/or a less severe altitude ($< 10,000$ ft).

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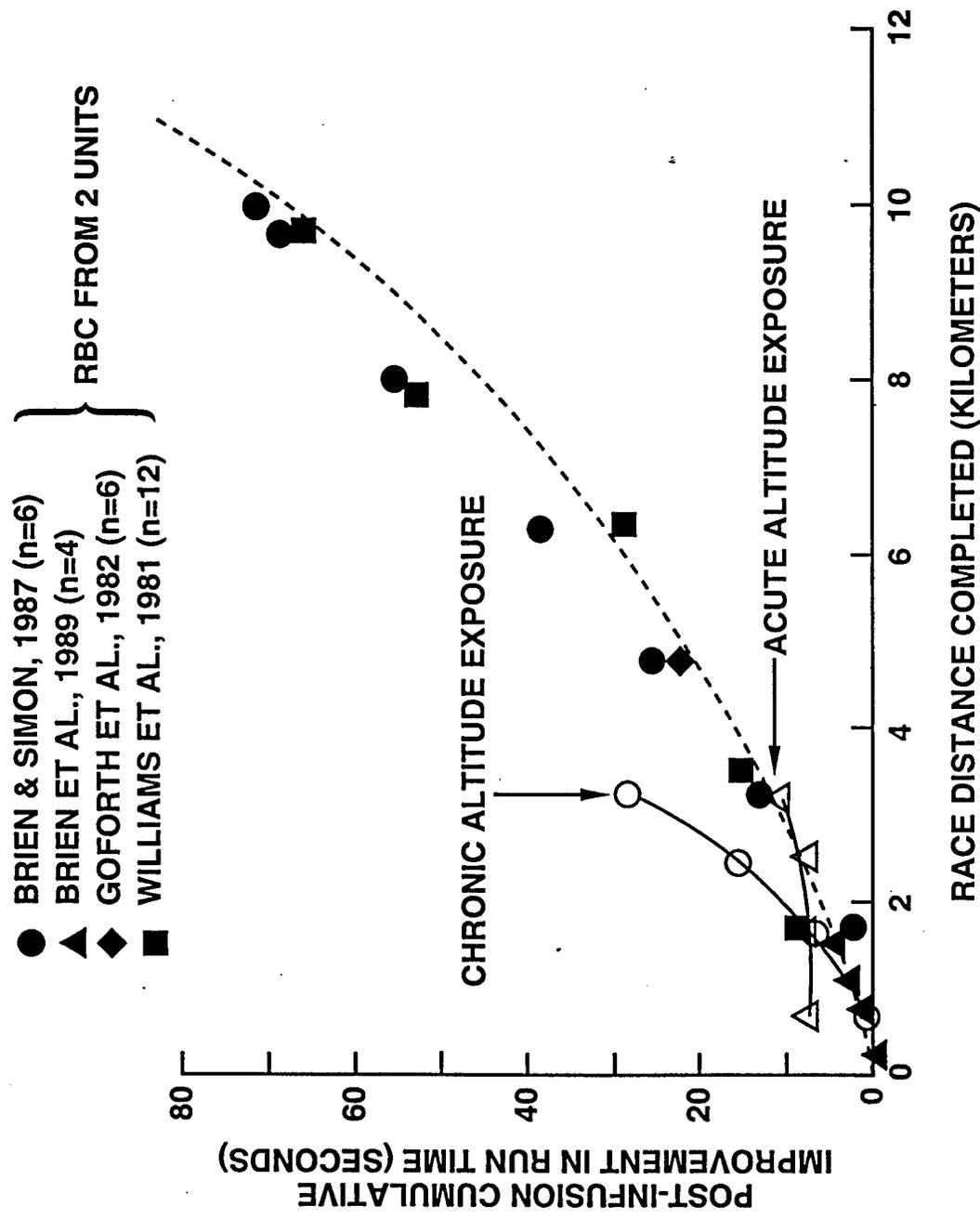
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FIGURE LEGEND

FIG. 1. Cumulative improvement in run time (sec) following the infusion of two units of freeze-preserved blood as a function of the distance (km) completed comparing the data from four previously published studies (2, 3, 7, 27) conducted at sea level or relatively low altitude to the present findings during acute and chronic exposure to high altitude.



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TABLE 1. TWO MILE RUN TIMES FOR THE INFUSED AND CONTROL GROUPS AT SEA LEVEL, AND DURING ACUTE AND CHRONIC EXPOSURE TO HIGH ALTITUDE (4300m)

GROUP	Sea Level (50m)	4300m Acute	4300m Chronic
Control Mean	13:39	19:44	18:45
S.E.	00:32	00:43	00:55
Infused Mean	13:14	19:02	17:44
S.E.	00:19	00:18	00:27

Control (CON) group, n=8; infused (ER) group, n=8; two mile run times, min:sec; ER vs. CON (P>0.05); sea level vs. acute and chronic high altitude (P<0.05).

TABLE 2. TWO MILE HEART RATES FOR THE INFUSED AND CONTROL GROUPS AT SEA LEVEL, AND DURING ACUTE AND CHRONIC EXPOSURE TO HIGH ALTITUDE (4300m)

GROUP	Sea Level (50m)	4300m Acute	4300m Chronic
Control Mean	191	178	169
S.E.	3	4	5
Infused Mean	188	170	162
S.E.	3	4	6

Control (CON) group, n=8; infused (ER) group, n=8; heart rate, beats·min⁻¹; ER vs. CON (P>0.05); sea level vs. acute and chronic high altitude (P<0.05).

TABLE 3. RATED PERCEIVED EXERTION FOR THE INFUSED AND CONTROL GROUPS AT SEA LEVEL, AND DURING ACUTE AND CHRONIC EXPOSURE TO HIGH ALTITUDE (4300m)

GROUP	Sea Level (50m)			4300m Acute			4300m Chronic		
	L	C	O	L	C	O	L	C	O
Control Mean	16.1	16.6	16.4	18.0	17.5	17.8	17.5	17.4	17.4
S.E.	.8	.6	.7	.5	.7	.6	.5	.5	.4
Infused Mean	15.9	16.0	15.5	16.9	17.4	17.5	17.4	17.3	17.1
S.E.	.5	.6	.7	.5	.4	.3	.5	.5	.5

Control (CON) group, n=8; infused (ER) group, n=8; rated perceived exertion: L, local; C, central; O, overall rating; ER vs. CON (P>0.05); L, sea level vs. acute and chronic high altitude (P<0.05); C, no altitude effect (P>0.05); O, sea level vs. acute high altitude (P<0.05).