J Appl Physiol 100: 1238–1248, 2006;
doi:10.1152/japplphysiol.00742.2005.

Exercise training in normobaric hypoxia in endurance runners. I. Improvement in aerobic performance capacity

Stéphane P. Dufour,[1] Elodie Ponsot,[1] Joffrey Zoll,[2] Stéphane Doutreleau,[1] Evelyne Lonsdorfer-Wolf,[1] Bernard Geny,[1] Eliane Lampert,[1] Martin Flück,[2] Hans Hoppeler,[2] Véronique Billat,[3] Bertrand Mettauer,[1,4] Ruddy Richard,[1] and Jean Lonsdorfer[1]

[1]*Département de Physiologie et des Explorations Fonctionnelles, Hôpital Civil, and Faculté de Médicine, Institut de Physiologie, Unité Propre de Recherche de l'Enseignement Supérieur Équipe d'Accueil 3072, Strasbourg, France;* [2]*Institute of Anatomy, University of Bern, Bern, Switzerland;* [3]*Laboratoire d'Etudes Physiologiques à l'Exercice, Département des Sciences du Sport et de l'Exercice, Équipe d'Accueil 3872, Université d'Evry Val d'Essonne, Evry, France; and* [4]*Service de Cardiologie, Hôpitaux Civils de Colmar, Colmar, France*

Submitted 22 June 2005; accepted in final form 27 July 2005

Dufour, Stéphane P., Elodie Ponsot, Joffrey Zoll, Stéphane Doutreleau, Evelyne Lonsdorfer-Wolf, Bernard Geny, Eliane Lampert, Martin Fluck, Hans Hoppeler, Véronique Billat, Bertrand Mettauer, Ruddy Richard, and Jean Lonsdorfer. Exercise training in normobaric hypoxia in endurance runners. I. Improvement in aerobic performance capacity. *J Appl Physiol* 100: 1238–1248, 2006; doi:10.1152/japplphysiol.00742.2005.—This study investigates whether a 6-wk intermittent hypoxia training (IHT), designed to avoid reductions in training loads and intensities, improves the endurance performance capacity of competitive distance runners. Eighteen athletes were randomly assigned to train in normoxia [Nor group; $n = 9$; maximal oxygen uptake ($\dot{V}O_{2\,max}$) = 61.5 ± 1.1 ml·kg^{-1}·min^{-1}] or intermittently in hypoxia (Hyp group; $n = 9$; $\dot{V}O_{2\,max}$ = 64.2 ± 1.2 ml·kg^{-1}·min^{-1}). Into their usual normoxic training schedule, athletes included two weekly high-intensity (second ventilatory threshold) and moderate-duration (24–40 min) training sessions, performed either in normoxia [inspired O_2 fraction ($F_{I_{O_2}}$) = 20.9%] or in normobaric hypoxia ($F_{I_{O_2}}$ = 14.5%). Before and after training, all athletes realized *1*) a normoxic and hypoxic incremental test to determine $\dot{V}O_{2\,max}$ and ventilatory thresholds (first and second ventilatory threshold), and *2*) an all-out test at the pretraining minimal velocity eliciting $\dot{V}O_{2\,max}$ to determine their time to exhaustion (T_{lim}) and the parameters of O_2 uptake ($\dot{V}O_2$) kinetics. Only the Hyp group significantly improved $\dot{V}O_{2\,max}$ (+5% at both $F_{I_{O_2}}$, $P < 0.05$), without changes in blood O_2-carrying capacity. Moreover, T_{lim} lengthened in the Hyp group only (+35%, $P < 0.001$), without significant modifications of $\dot{V}O_2$ kinetics. Despite similar training load, the Nor group displayed no such improvements, with unchanged $\dot{V}O_{2\,max}$ (+1%, nonsignificant), T_{lim} (+10%, nonsignificant), and $\dot{V}O_2$ kinetics. In addition, T_{lim} improvements in the Hyp group were not correlated with concomitant modifications of other parameters, including $\dot{V}O_{2\,max}$ or $\dot{V}O_2$ kinetics. The present IHT model, involving specific high-intensity and moderate-duration hypoxic sessions, may potentialize the metabolic stimuli of training in already trained athletes and elicit peripheral muscle adaptations, resulting in increased endurance performance capacity.

maximal oxygen uptake; time to exhaustion; competitive endurance runners

AT SEA LEVEL, IT IS WELL KNOWN that the training-induced improvements in endurance performance progressively level off as the aerobic fitness progresses. Therefore, the use of the metabolic stimulus provided by living (i.e., living high-training low) or training at altitude (i.e., living low-training high) has gained popularity in athletes to further enhance endurance performance. In this context, methods that impose short-term altitude exposure while exercising have progressively emerged to cope with the growing evidence that long-term altitude exposure possesses several detrimental effects, including a limited aerobic power, reducing both the metabolic and mechanical components of the total training load (24). To conciliate altitude training with a maintained training load, it has recently been proposed to use altitude in several but not all training sessions, included into a training program otherwise performed in normoxia [intermittent hypoxia training (IHT)] (38, 44–46).

To date, these training programs have provided conflicting results in endurance athletes (38, 44, 46), which could be due to the various combinations of duration and intensity of the hypoxic training sessions employed (32). Accordingly, improvement of performance in competitive swimmers has not been observed after an IHT program, including very short high-intensity (30–60 s) hypoxic sessions (44), whereas longer periods of high-intensity hypoxic exercise (70 s to 3 min) improved maximal power output at sea level in professional cyclists (39). In addition, significant maximum oxygen uptake ($\dot{V}O_{2\,max}$) improvement at sea level has been reported in trained subjects after hypoxic exercise bouts of 2- to 12-min duration (38). Recent evidence also demonstrated no beneficial effects of IHT programs, when the hypoxic exercise intensity is set below 80% of normoxic $\dot{V}O_{2\,max}$ (44, 46). Collectively, these findings point to a pivotal role for a minimal hypoxic exercise duration and intensity in IHT models, especially in trained athletes. Based on these observations, we assumed that two successive hypoxic training bouts, of 12–20 min, performed at the second ventilatory threshold (VT_2) (~80% of normoxic $\dot{V}O_{2\,max}$) are likely to comply with the above information. Moreover, integrated within the usual normoxic training of competitive runners, the intermittent nature of such specific hypoxic sessions would allow maintaining high levels of total training load and may elicit significant improvement of endurance performance capacity.

Characterization of the endurance performance capacity in athletes involves incremental exercise testing, allowing for the determination of the ventilatory thresholds [first ventilatory

Address for reprint requests and other correspondence: J. Lonsdorfer, Hôpital de la Robertsau, 83 rue Himmerich, BP 426, 67091 Strasbourg Cedex, France (e-mail: jeanlonsdorfer@hotmail.fr).

The costs of publication of this article were defrayed in part by the payment of page charges. The article must therefore be hereby marked "*advertisement*" in accordance with 18 U.S.C. Section 1734 solely to indicate this fact.

threshold (VT_1) and VT_2], $\dot{V}O_{2\,max}$, as well as their associated minimal running velocities (vVT_1, vVT_2, and $v\dot{V}O_{2\,max}$). Additionally, since $v\dot{V}O_{2\,max}$ falls among the significant predictors of endurance performance (4, 5), the time to exhaustion at $v\dot{V}O_{2\,max}$ (T_{lim}) is thought to constitute an important determinant of the endurance performance capacity. Despite its athletic relevance, the effect of IHT program on T_{lim} in endurance athletes remains unknown. Since the maximal rate (i.e., $\dot{V}O_{2\,max}$ or $v\dot{V}O_{2\,max}$) (6, 23) and/or kinetic changes in the O_2 flux adjustment (13) are expected to contribute to T_{lim} performance, the possible influence of IHT on both of these respective properties of aerobic metabolism is also not elucidated.

Therefore, the purpose of this study was to test the hypotheses that an original IHT model, including two weekly moderate-duration (24–40 min) and high-intensity (VT_2) hypoxic sessions within the usual normoxic training of already trained athletes, *1*) improves running velocities at sea level due to amelioration of aerobic energy provision, including $\dot{V}O_{2\,max}$; and *2*) lengthens T_{lim} at sea level with concomittant adaptations of aerobic metabolism properties, mainly $\dot{V}O_{2\,max}$ and/or oxygen uptake ($\dot{V}O_2$) kinetics.

METHODS

Subjects

Eighteen highly trained male distance runners were recruited from local athletic teams and completed the study before the beginning of their competitive season. Their main physical and physiological characteristics are shown in Table 1. After all the potential risks were explained, the athletes gave a voluntary written consent to participate to the protocol, approved by our hospital and national review boards. In the weeks before and during the study, the subjects lived under the altitude of 300 m and were engaged in a regular training schedule comprising five training sessions per week, including two weekly training sessions performed specifically at VT_2 (49). Their respective individual training schedule remained unaltered during the experimental period. All were highly motivated to participate in the study, familiar with treadmill running, and with current 10,000 m or equivalent personal-best times of <35:00 (min:s).

Table 1. *Anthropometric data and performance capacity of the subjects*

	Hypoxic Group	Normoxic Group	*P* Value
Number of subjects	9	9	NS
Body weight, kg	70.6±2.2	71.3±2.2	NS
Height, cm	180±1	180±2	NS
Age, yr	30.3±6.3	30.3±6.1	NS
Body fat, %	11.8±0.8	12.1±1.2	NS
[Hb], g/l	15.3±0.2	15.3±0.3	NS
Hct, %	45.1±0.8	46.0±1.2	NS
$\dot{V}O_{2\,max}$, ml·kg^{-1}·min^{-1}	64.2±1.2	61.5±1.1	NS
$v\dot{V}O_{2\,max}$, km/h	19.6±0.2	19.0±0.4	NS
VT_2, % $\dot{V}O_{2\,max}$	89.7±1.5	88.7±1.2	NS

Values are means ± SE. Hypoxic and normoxic groups are groups that included only two training sessions at the velocity corresponding to the second ventilatory threshold (VT_2) in their usual weekly training schedule and performed under hypoxic or normoxic condition, respectively. %Body fat is the percentage of body fat determined according to Durnin and Womersley (16). [Hb], hemoglobin concentration; Hct, hematocrit; $\dot{V}O_{2\,max}$, maximal oxygen uptake determined in the normoxic incremental test; $v\dot{V}O_{2\,max}$, the lowest running speed associated with $\dot{V}O_{2\,max}$ in the normoxic incremental exercise test. VT_2 was determined during the normoxic incremental test. NS, no significant difference between hypoxic and normoxic groups.

Experimental Design

As shown in Fig. 1, the study was organized in four successive phases: a basal medical examination, the pretraining treadmill performance evaluation, the training process, and the posttraining treadmill performance evaluation.

Basal medical examination. Two weeks before the beginning of the training period, each subject came to the laboratory for anthropometric measurements, physical examination, resting electrocardiography, and echocardiography recordings. To verify their exercise and hypoxic tolerance under careful cardiac monitoring, all athletes also performed maximal graded cycle tests in normoxia and hypoxia. These tests did not reveal any abnormality that could prevent the subjects from being included in the experimental protocol.

Pre- and posttraining treadmill performance evaluation. In the week before and after the training intervention, all of the subjects performed three exercise tests on a motorized treadmill (Gymrol 2500 SP, Tecmachine), which were separated by at least 24 h of rest: *1*) a treadmill incremental exercise test (IET) to exhaustion in Nor [IET_N; inspired O_2 fraction (FI_{O_2}) = 20.9%]; *2*) a treadmill IET to exhaustion in Hyp (IET_H; FI_{O_2} = 14.5%, equivalent to an altitude of 3,000 m); and *3*) a normoxic all-out test at pretraining $v\dot{V}O_{2\,max}$. For a given subject, all tests were performed at the same time of day in a climate-controlled environment (21–23°C).

Training program. During the 6 wk of the study, both groups continued their usual training program (5 sessions/week), including their two weekly sessions at VT_2 that were performed in the laboratory. All of the laboratory training sessions were performed under careful supervision of an experimented physician. For the group who trained in normoxia (Nor group), VT_2 was determined during the IET_N, and for the group who trained in hypoxia (Hyp group), VT_2 was determined during the IET_H. Each VT_2 session began with a 10-min warm-up at 60% $\dot{V}O_{2\,max}$ (<VT_1), followed by two periods at VT_2 (time run at VT_2 specified in Fig. 1), separated by 5-min recovery at 60% $\dot{V}O_{2\,max}$. For the Hyp group, the subjects trained under hypoxic conditions only during the running periods at VT_2 by breathing through face masks connected to a mixing chamber via appropriate tubing. Warm-up and recoveries were performed under normoxia. The training load during the laboratory sessions was organized into two 3-wk periods in which the exercise duration at VT_2 increased progressively (Fig. 1). At the 4th wk, the training velocity was readjusted to maintain an exercise heart rate (HR) corresponding to the one achieved at the first training session. Throughout the study, each athlete underwent a total of 12 controlled laboratory training sessions. No athletes withdrew from the study before the achievement of the posttraining treadmill performance evaluation, and none complained of health complications throughout the study.

Procedures

Altitude simulation. Normobaric hypoxic conditions corresponding to an altitude of 3,000 m (FI_{O_2} = 14.5%) were simulated by diluting ambient air with nitrogen via a mixing chamber, with the dilution being constantly controlled by a P_{O_2} probe (Alti-Trainer$_{200}$, Sport and Medical Technology). This device allows the inspired P_{O_2} to be set at a predetermined value to simulate altitude. The precision of the P_{O_2} is of ±0.82 Torr. The respiratory effort induced by the device at 6 l/s was negligible (<0.01 W).

Treadmill tests. The IET_N or the IET_H were performed in random order on a motorized treadmill with 0% slope, to determine VT_1, VT_2, $\dot{V}O_{2\,max}$, the associated velocities, and the running economy (RE) in both conditions of oxygen availability. During each IET, the initial running speed was set at 10 km/h and increased by 1 km/h every 2-min until volitional exhaustion. Each subject was encouraged to give a maximum effort. Arterialized blood samples were obtained from the earlobe at rest, at exhaustion, as well as at the first and third minute of recovery to determine total blood lactate concentration ([La]).

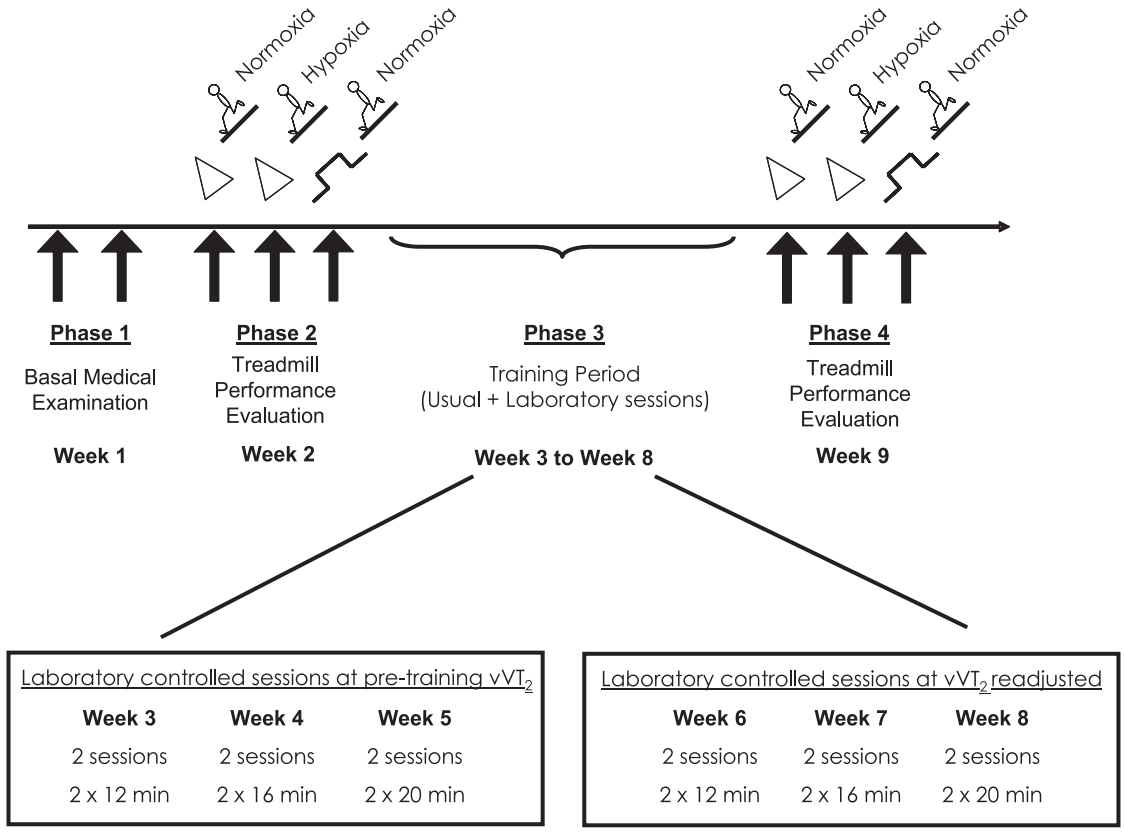

Fig. 1. Study design with the 4 phases of the experimental protocol. △, Incremental running tests; ⌐¬, all-out running tests in normoxia. Normoxia: inspired O_2 fraction (FI_{O_2}) = 20.9%. Hypoxia: FI_{O_2} = 14.5%. *Phases 1*, *2*, *3*, and *4* are the respective experimental phases (see text for details). vVT_2, running velocity corresponding to the second ventilatory threshold.

The all-out running test was performed in normoxia at pretraining $v\dot{V}O_{2\,max}$, i.e., the same absolute running speed before and after training. The test began by 10-min warm-up at 60% of the subject's $v\dot{V}O_{2\,max}$ (lower than vVT_1 in all subjects). The subjects were then connected to the test equipments during a 5-min period of rest and immediately asked to run at their individual $v\dot{V}O_{2\,max}$ for as long as possible. The transition from rest to $v\dot{V}O_{2\,max}$ occurs within a 20-s delay (range 17–23 s), necessary for the treadmill to reach the desired speed. No information about the time elapsed was provided to the athletes. During this test, arterialized blood samples were obtained from the earlobe at rest, at exhaustion, as well as at the 1st and 3rd min of recovery to determine total blood [La].

HR monitoring. During all of the running tests, as well as during the controlled training sessions, HR was continuously monitored by telemetry (Polar Vantage, Kempeley, Finland).

Gas exchange measurements. During all tests, inspiratory ($\dot{V}I$) and expiratory minute ventilation ($\dot{V}E$), $\dot{V}O_2$, and carbon dioxide output ($\dot{V}CO_2$) were measured breath by breath with an open-circuit metabolic cart with rapid O_2 and CO_2 analyzers (Sensor Medics MSE, Yorba Linda, CA). Before each individual exercise test, the pneumotachograph was calibrated with several strokes given by a 3-liter calibration syringe. The gas analyzers were calibrated by using reference gases with known O_2 and CO_2 concentrations (12% O_2, 5% CO_2). FI_{O_2} and fraction of O_2 in the expired air (FE_{O_2}) were analyzed continuously for each breath. Therefore, $\dot{V}O_2$ was calculated in normoxia and hypoxia by the following formula, where all parameters are expressed in STPD conditions: $\dot{V}O_2 = \dot{V}I \times FI_{O_2} - \dot{V}E \times FE_{O_2}$.

During the IET, each athlete was encouraged to give a maximal effort. Peak treadmill velocity was defined as the last achieved running speed sustained for at least 30 s. $\dot{V}O_{2\,max}$ was always defined as the highest 30-s averaged $\dot{V}O_2$ value. As previously described by Billat and Koralsztein (4), $v\dot{V}O_{2\,max}$ was defined as the minimal velocity at which $\dot{V}O_{2\,max}$ occurred. In detail, if $\dot{V}O_{2\,max}$ was reached during the last stage, which was maintained >90 s, that particular velocity was taken as $v\dot{V}O_{2\,max}$. If that velocity eliciting $\dot{V}O_{2\,max}$ was sustained <60 s, then $v\dot{V}O_{2\,max}$ was taken as the velocity at the previous stage. If that velocity eliciting $\dot{V}O_{2\,max}$ was maintained between 60 and 90 s, then $v\dot{V}O_{2\,max}$ was considered to be equal to the velocity during the previous stage plus the half velocity increase between the last two stages, i.e., (1 km/h)/2 = 0.5 km/h (29). Ventilatory thresholds were assessed by using established criteria (3, 49). VT_1 corresponds to the break point in the plot of $\dot{V}CO_2$ as a function of $\dot{V}O_2$. At that point, the ventilatory equivalent for O_2 ($\dot{V}E/\dot{V}O_2$) increases without an increase in the ventilatory equivalent for CO_2 ($\dot{V}E/\dot{V}CO_2$). VT_2 was located between VT_1 and $\dot{V}O_{2\,max}$, when $\dot{V}E/\dot{V}CO_2$ starts to increase while $\dot{V}E/\dot{V}O_2$ continues to increase. The oxygen pulse (O_2p) was calculated as the ratio between $\dot{V}O_2$ and HR, also representing stroke volume times arteriovenous oxygen difference [Δ(a-v)O_2] (30). RE was defined as the rate of $\dot{V}O_2$ for a given submaximal work rate (9). Therefore, RE corresponds to the 1-min average of the $\dot{V}O_2$ values recorded at the end of the 12 km/h stage during each IET. This speed was lower than VT_1 for all of the subjects in both environmental conditions and allows an estimation of RE for an exercise intensity expected to be mainly aerobic. To provide additional insights in the effect of IHT on RE, we also determined RE at 18 km/h in IET_N and at 15 km/h in IET_H. These running speeds amounted to ~92 and 90% of the respective normoxic and hypoxic $v\dot{V}O_{2\,max}$, corresponding to recommended speed for RE determination in athletes (10).

Blood O_2-carrying capacity and lactate. On the first day of the treadmill performance evaluation before and after training, blood was drawn from an antecubital vein in each group to immediately measure

hematocrit (Hct) and hemoglobin concentration. Earlobe blood samples obtained during all running tests were also immediately analyzed for total blood [La] by an enzymatic method.

Oxygen saturation. During each exercise test, hemoglobin saturation was monitored continuously by earlobe pulse oximetry (Oxypleth, Novametrix-Medical System).

$\dot{V}O_2$ Kinetics

Data modelization. To describe the $\dot{V}O_2$ kinetics [$\dot{V}O_2(t)$] during the all-out test, we used a mathematical model with two exponential functions (2):

$$\dot{V}O_2(t) = \dot{V}O_{2b} + A_1\{1 - e^{-[(t-td_1)/\tau_1]}\}U_1 \text{ [Phase 2 (fast component)]} + A_2\{1 - e^{-[(t-td_2)/\tau_2]}\}U_2 \text{ [Phase 3 (slow component)]} \quad (1)$$

where $U_1 = 0$ for $t < td_1$ and $U_1 = 1$ for $t \geq td_1$; $U_2 = 0$ for $t < td_2$ and $U_2 = 1$ for $t \geq td_2$; $\dot{V}O_{2b}$ is the rate of $\dot{V}O_2$ at rest before the start of the all-out test; A_1 and A_2 are the asymptotic amplitudes for the first and second exponential terms, respectively; τ_1 and τ_2 are the time constants and represent the time to reach 63% of the total amplitude of the respective fast and slow $\dot{V}O_2$ components; td_1 and td_2 represent the time delays for the fast and the slow components, respectively. As the initial cardiodynamic phase of the $\dot{V}O_2$ adjustment to a rest-to-exercise transition does not influence the fast component of $\dot{V}O_2$ (36) and because we focused on the fast and slow components of the $\dot{V}O_2$ response, the cardiodynamic phase was excluded from analysis by removing the data from the first 20 s of the all-out test. The parameters of the model were determined with an iterative procedure that minimizes the sum of the mean squares of the differences between the model $\dot{V}O_2$ estimates and the corresponding $\dot{V}O_2$ measurements. To exclude aberrant breaths from analysis, breath-by-breath $\dot{V}O_2$ values that were greater than three standard deviations from the modeled $\dot{V}O_2$ were removed and assumed to represent events unrelated to the physiological response of interest (31, 39). These values represented <1% of the total data.

Slow component of $\dot{V}O_2$ kinetics. Because the asymptotic value of the second exponential term is not necessarily reached at the subject's exhaustion, the amplitude of the slow component was computed as A_2' (7):

$$A_2' = A_2[1 - e^{-(T_{lim}-td_2)/\tau_2)}] \quad (2)$$

where T_{lim} is the time at the end of the all-out exercise test. Moreover, to compare the amplitude of the $\dot{V}O_2$ slow component at consistent time before and after training, we also calculated the amplitude of the $\dot{V}O_2$ slow component achieved posttraining when the subjects attained their pretraining T_{lim} value (A_2'old).

O_2 deficit calculation. According to Whipp and Ozyener (51), the fast component of the $\dot{V}O_2$ kinetics represents an "expected $\dot{V}O_2$," whereas the slow component is the manifestation of an "excess $\dot{V}O_2$" occurring later during exercise (i.e., after td_2). Consequently, the oxygen deficit (O_2def) is estimated from the area between the fast-component response curve and the fast-component asymptote (13):

$$O_2\text{def} = (td_1 \times A_1) + (\tau_1 \times A_1) \quad (3)$$

where O_2def is in milliliters, td_1 and τ_1 are in seconds, and A_1 is in milliliters per second.

Computation of the time sustained at pretraining $\dot{V}O_{2\,max}$. Besides T_{lim}, which could be considered as a mechanical parameter of endurance performance (reflecting the total mechanical work performed at $v\dot{V}O_{2\,max}$), we also calculated a metabolic correlate (*Eq. 4*), from the time sustained while the athlete ran at >95% of pretraining $\dot{V}O_{2\,max}$ (T_{lim} @ $\dot{V}O_{2\,max}$). This percentage was chosen to account for a 5% random error in the determination of $\dot{V}O_{2\,max}$ (33) and also because all athletes did not necessarily reach 100% $\dot{V}O_{2\,max}$ in T_{lim} testing (13).

$$T_{lim}@\dot{V}O_{2max}(s) = T_{lim} - TA\ \dot{V}O_{2max} \quad (4)$$

where T_{lim} is the time to exhaustion while the athletes ran at the pretraining minimal velocity associated with $\dot{V}O_{2\,max}$ (s), and the time to attain $\dot{V}O_{2\,max}$ (TA $\dot{V}O_{2\,max}$) corresponds to the time necessary to reach 95% of pretraining $\dot{V}O_{2\,max}$ (s). Depending on whether the $\dot{V}O_2$ kinetics were better described by a mono- or a double-exponential model, TA $\dot{V}O_{2\,max}$ was computed from the equations below.

1) For the monoexponential model (fast component in *Eq. 1*)

$$TA\ \dot{V}O_{2max} = td_1 - \tau_1 \times \{\ln[1 - (0.95 \times \dot{V}O_{2max} - \dot{V}O_{2b})/A_1]\}$$

2) For the double-exponential model (fast + slow component in *Eq. 1*)

$$TA\ \dot{V}O_{2max} = td_2 - \tau_1 \times \{\ln[1 - (0.95 \times \dot{V}O_{2max} - \dot{V}O_{2b} - A_1)/A_2]\}$$

Evaluation of Training

All athletes were asked to report their individual training schedule into detailed training logs, including duration, distance, and intensity of each training sessions. Laboratory as well as field work bouts were taken into account to provide both quantitative and qualitative characterization of the overall training load. Duration and intensity of the training sessions performed out of the laboratory were assessed based on the running velocity spread out in four intensity zones: low ($<v\dot{V}T_1$), moderate ($v\dot{V}T_1 - v\dot{V}T_2$), heavy ($v\dot{V}T_2 - v\dot{V}O_{2\,max}$), and severe intensity ($>v\dot{V}O_{2\,max}$).

Statistics

Whether a mono- or biexponential model better described the $\dot{V}O_2$ kinetics during the all-out tests was determined using a Fisher test. We used the bootstrap method to obtain an estimation of the accuracy of the parameters describing the $\dot{V}O_2$ kinetics (7, 8, 17). This method, creating 1,000 different samples of the same size than the original data set, allows the determination of a coefficient of variation for each mathematical parameter on an individual basis.

Data were first tested for distribution normality and variance homogeneity. Subsequently, the differences between groups before the training period were analyzed with the Mann-Whitney procedure. To test for both treatment (Hyp vs. Nor) and time (before vs. after) effects on each of the measurements during the training period, we used a two-way ANOVA for repeated measures. When significant modifications were found, the Student-Newman-Keuls post hoc procedure was performed to localize the difference. Pearson linear regression analysis was used to determine any potential linear relationship between variables. All statistical analyses were performed with the SigmaStat 3.0 software (SPSS, Chicago, IL), and the level of significance was chosen for $P < 0.05$. Values are means ± SE.

RESULTS

The anthropometric and treadmill performance characteristics of the athletes are shown in Table 1. No significant differences were reported between the two experimental groups before the training period. Moreover, in both groups, the training period did not modify anthropometric and blood parameters, including body mass [Hyp after: 70.5 ± 2.2 kg, nonsignificant (NS); Nor after: 71.3 ± 2.2 kg, NS], hemoglobin (Hyp after: 15.8 ± 0.5 g/dl, NS; Nor after: 15.7 ± 0.5 g/dl, NS), and Hct (Hyp after: 46.4 ± 1.5%, NS; Nor after: 46.9 ± 1.2%, NS).

Training Load

Laboratory training sessions. At the beginning of the study and according to the training environment, the Hyp group trained at a significantly lower running speed (Table 2). These different running speeds corresponded to the same exercise HR, whether expressed in absolute (Hyp: 166 ± 3 vs. Nor: 172 ± 3 beats/min; NS) or in relative value (Hyp: 96 ± 1 vs.

Table 2. *Training load characteristics*

	Hypoxic Group	Normoxic Group	*P* Value
Laboratory controlled sessions			
First 3 wk			
Running speed at VT_2, km/h	15.0±0.2	16.7±0.3	<0.01
Running speed absolute, %	77±1	88±1	<0.01
Running speed relative, %	89±2	88±1	NS
Last 3 wk			
Running speed at VT_2, km/h	15.4±0.2	16.7±0.3	<0.01
Running speed absolute, %	76±1	86±1	<0.01
Running speed relative, %	87±1	86±1	NS
Total training, % of total training time			
Low	72.7±1.8	68.7±4.2	NS
Moderate	4.9±1.4	9.7±1.9	NS
Heavy	21.0±1.0	21.4±2.8	NS
Severe	1.3±0.5	0.3±0.1	NS

Values are means ± SE. The running velocity of the controlled training sessions was readjusted after 3 wk of training, according to heart rate changes (See METHODS). Running speed absolute is running velocity expressed as a percentage of the pre- (first 3 wk) or posttraining (last 3 wk) velocity associated with $\dot{V}O_{2\,max}$ under normoxic conditions. Running speed relative is running velocity expressed as a percentage of the pre- (first 3 wk) or posttraining (last 3 wk) velocity associated with $\dot{V}O_{2max}$ in the group-specific environment. Intensity zones are as follow: Low < velocity associated with the first ventilatory threshold < Moderate < velocity associated with the second ventilatory threshold < Heavy < velocity associated with maximal oxygen uptake < Severe. *P* value, level of significance for the difference between groups.

Nor: 94 ± 1%; NS). During the first VT_2 training session in the Hyp group, blood oxygen saturation stabilized at a value of 80 ± 1%. At the 4th wk, the running speed of the VT_2 bouts was increased by 0.4 ± 0.1 km/h for the Hyp group, but not modified for the Nor group. As a result, HR values remained unaltered throughout the 6-wk intervention in both groups as it was the case for blood oxygen saturation in the Hyp group. For the Hyp group, the total duration of hypoxic exposure amounted to 384 min (i.e., *week 1 + week 2 +. . .+ week 6*) and was well tolerated.

Total training load. The total training load (i.e., field and laboratory training sessions) was comparable in both groups. During the 6-wk training, Hyp and Nor groups performed, respectively, 33.0 ± 0.6 and 31.2 ± 1.7 training sessions, leading to no difference in total training time and total training distance (Hyp: 2,013 ± 114 min and 478 ± 27 km vs. Nor: 2,095 ± 158 min and 498 ± 40 km). Consequently, the averaged running speed over the 6-wk training intervention was similar in both groups (Hyp: 14.3 ± 0.2 vs. Nor: 14.2 ± 0.2 km/h). No significant differences appeared either in total time or in total distance run in the respective intensity zones (Table 2).

Exercise Capacity in Hypoxia

IET. Table 3 and Fig. 2*A* report the effects of the 6-wk training on results of the IET_H. Only the Hyp group significantly improved submaximal and maximal running velocities (Table 3) under hypoxia. Indeed, vVT_1, vVT_2, and $v\dot{V}O_{2\,max}$ increased, respectively, by +7, +8, and +5% after IHT. The $\dot{V}O_2$ associated with these velocities improved in the same proportions by +7, +7, and +5%, respectively (Fig. 2*A*), but RE did not change. The maximum O_2p (O_2p_{max}) improved (+5%) only in the Hyp group after IHT (Table 3). Conversely, the Nor group demonstrated no improvement of all of these parameters under hypoxic conditions.

Exercise Capacity in Normoxia

IET. $v\dot{V}O_{2\,max}$ improved significantly by +4 and +3% and vVT_2 increased significantly by +5 and +3% in the Hyp and Nor groups, respectively ($P < 0.05$), under normoxic conditions (Table 3). However, only the Hyp group significantly enhanced $\dot{V}O_{2\,max}$ as well as $\dot{V}O_2$ at VT_2 by +5 and +7%, respectively ($P < 0.05$), with no modification of the RE (Fig.

Table 3. *Running velocities, running economy, and selected maximal physiological parameters measured in normoxic and hypoxic incremental tests before and after the 6-wk training period*

	Hypoxic Group		Normoxic Group	
	Pre	Post	Pre	Post
Running velocities				
V_{peak}, km/h				
Normoxia	20.5±0.2	20.9±0.2*	19.8±0.4	20.2±0.4*
Hypoxia	17.7±0.3	18.4±0.2†	17.2±0.4	17.6±0.4
$v\dot{V}O_{2\,max}$, km/h				
Normoxia	19.6±0.2	20.3±0.2†	19.0±0.4	19.6±0.3*
Hypoxia	17.0±0.4	17.8±0.3†	16.3±0.3	16.7±0.4
vVT_2, km/h				
Normoxia	18.0±0.2	18.9±0.1†	17.2±0.4	17.8±0.4*
Hypoxia	15.4±0.2	16.6±0.2†	15.1±0.3	15.6±0.4
vVT_1, km/h				
Normoxia	15.3±0.2	15.6±0.2	14.4±0.4	15.0±0.4*
Hypoxia	13.1±0.2	14.0±0.2†	12.9±0.3	13.2±0.4
Running economy, ml $O_2 \cdot min^{-1} \cdot kg^{-1}$				
Normoxia at 12 km/h	38.2±1.9	36.9±0.8	36.9±1.2	36.1±1.0
Hypoxia at 12 km/h	39.3±1.1	38.6±1.6	39.9±1.3	38.7±1.3
Normoxia at 18 km/h	57.9±1.6	57.8±1.4	57.2±1.8	55.6±1.1
Hypoxia at 15 km/h	50.2±1.3	50.6±2.1	51.4±1.8	49.8±1.2
Maximal physiological parameters				
O_2p_{max}, ml·beats^{-1}·min^{-1}				
Normoxia	24.7±0.8	26.2±1.0*	23.8±0.6	23.9±0.9
Hypoxia	23.5±1.0	24.8±1.2*	22.6±0.8	22.5±0.5
HR_{max}, beats/min				
Normoxia	183±2	182±4	184±4	185±3
Hypoxia	170±3	172±3	174±4	174±3
$\dot{V}E_{max}$, l/min				
Normoxia	157±7	162±8	147±7	144±5
Hypoxia	134±7	137±8	142±5	132±7
$[La]_{max}$, mmol/l				
Normoxia	6.9±0.8	6.5±0.5	7.3±0.5	7.9±0.7
Hypoxia	6.7±0.9	6.6±0.7	8.6±0.7	7.7±0.7
RER_{max}				
Normoxia	1.05±0.02	1.05±0.02	1.04±0.03	1.04±0.03
Hypoxia	1.04±0.02	1.07±0.04	1.06±0.01	1.06±0.03
$\dot{V}O_2$ leveling off (yes/no), no.				
Normoxia	7/2	6/3	6/3	8/1
Hypoxia	6/3	6/3	7/2	6/3

Values are means ± SE. Pre and Post, before and after the 6-wk training period; V_{peak}, $v\dot{V}O_{2\,max}$, vVT_2, vVT_1: running velocities achieved during the incremental exercise test at exhaustion, at $\dot{V}O_{2max}$, and at the second and first ventilatory threshold, respectively; O_2p_{max}, HR_{max}, $\dot{V}E_{max}$, $[La]_{max}$, and RER_{max}: maximal values for oxygen pulse, heart rate, ventilation, blood lactate, and respiratory exchange ratio, respectively; $\dot{V}O_2$ leveling off, number of subjects who have/have not reached a $\dot{V}O_2$ plateau at the end of the incremental test. Significant differences between Pre and Post values: *$P <$ 0.05, †$P < 0.01$.

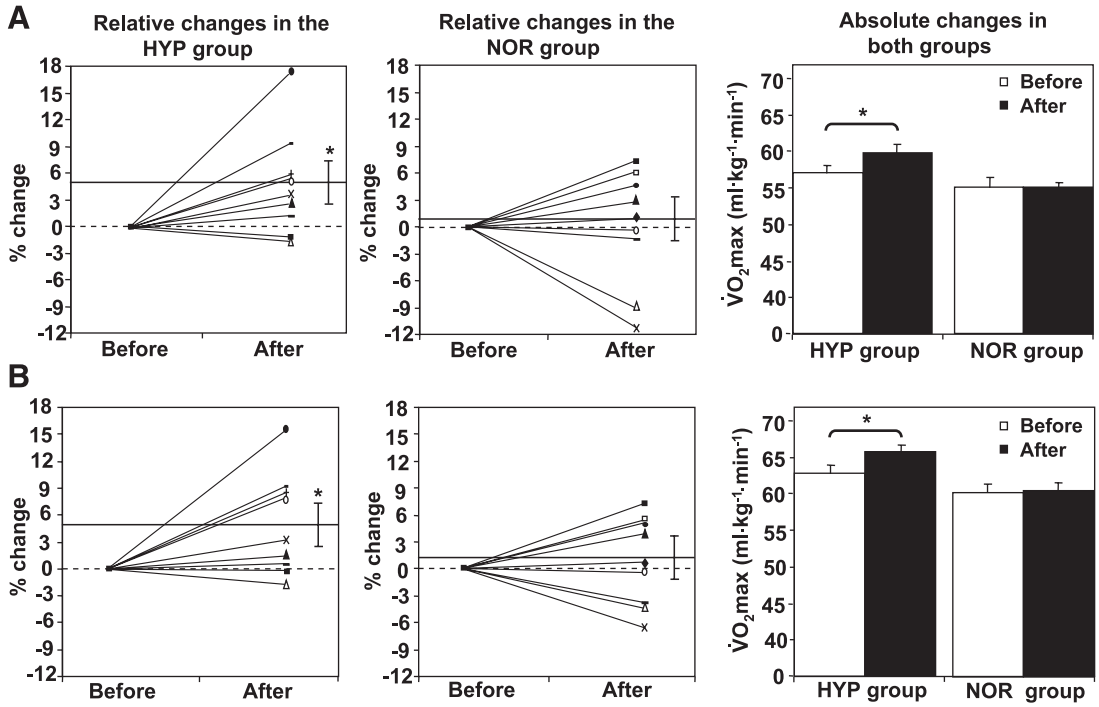

Fig. 2. Percent change in maximal oxygen uptake ($\dot{V}O_{2\,max}$) in hypoxia (*A*) and in normoxia (*B*) for each individual subject of the hypoxia group (Hyp; *left*) and normoxia group (Nor; *middle*), before and after the training program. Horizontal solid lines with vertical bars represent group changes. Horizontal dashed lines are the zero level. *Right*: absolute mean changes for all subjects from the Hyp and Nor groups. Hyp and Nor represent the groups of subjects that performed the laboratory-controlled training sessions under hypoxic or normoxic conditions, respectively. All values are presented as means ± SE. Significant differences before vs. after training, $^*P < 0.05$.

2*B*). Again, O_2p_{max} increased (+6%) only in the Hyp group after IHT (Table 3). The Nor group disclosed no significant changes, neither for exercise $\dot{V}O_2$ nor for RE.

All-out exercise test. The all-out exercise tests were performed in normoxia at the same absolute running velocity before and after training, i.e., pretraining $v\dot{V}O_{2\,max}$. After training, this speed amounted to 96 and 97% of the posttraining $v\dot{V}O_{2\,max}$ for the Hyp and Nor group, respectively, therefore corresponding to the same relative running speed in both groups. As shown in Fig. 3, training significantly enhanced T_{lim} in the Hyp but not in the Nor group (+35 vs. +10%, $P < 0.05$). Similar changes in the time sustained at pretraining $v\dot{V}O_{2\,max}$ were obtained when the transition period required for treadmill speed stabilization was subtracted from T_{lim} (+35 vs. +10%, $P < 0.05$). Concomitantly, the end-exercise $\dot{V}O_2$ achieved during the all-out test increased in the Hyp group only (+6%, $P < 0.05$), whereas the maximal [La] values remained unchanged after training (Table 4).

The kinetics of $\dot{V}O_2$ response of a typical subject from the Hyp and Nor group are shown in Fig. 4. Training did not modify parameters of the fast component of $\dot{V}O_2$ kinetics (Table 4) and O_2def remained unchanged (Hyp group: before 3,319 ± 266 vs. after 3,372 ± 469 ml O_2, NS; Nor group: before 2,793 ± 239 vs. after 2,563 ± 169 ml O_2, NS). A slow

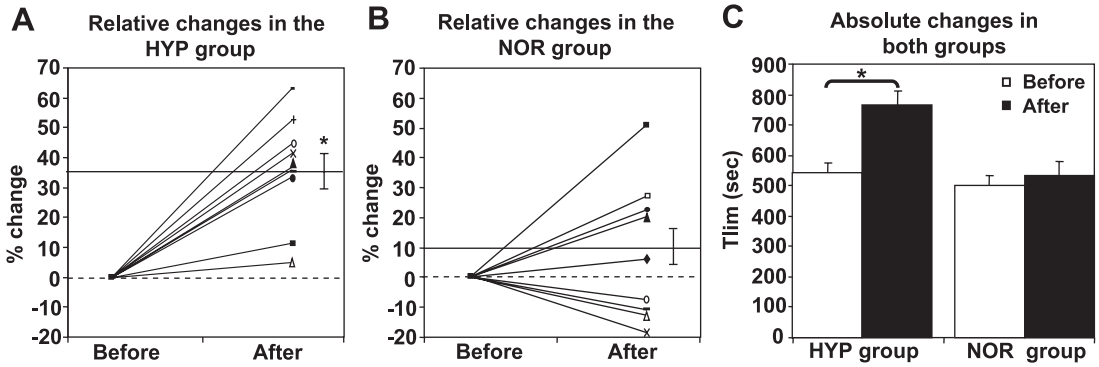

Fig. 3. Percent change in time to exhaustion for each individual subject before and after the training program in the Hyp (*A*) and Nor (*B*) groups. Horizontal solid lines with vertical bars represent group changes. Horizontal dashed lines are the zero level. *C*: absolute mean changes for all subjects from the Hyp and Nor groups. Values are presented as means ± SE. Hyp and Nor represent the group of subjects who performed the laboratory-controlled training sessions under hypoxic or normoxic conditions, respectively. T_{lim}, running time to exhaustion at the pretraining minimal velocity associated with $\dot{V}O_{2\,max}$. Significant differences before vs. after training, $^*P < 0.05$.

Table 4. *Training effects on the time until exhaustion and the parameters of the $\dot{V}O_2$ kinetics*

	Hypoxic Group				Normoxic Group			
	Pre		Post		Pre		Post	
	Values	CV mean, %	Values	CV mean, %	Values	CV mean, %	Values	CV mean, %
td_1, s	18.0 ± 1.7	13.9	22.6 ± 2.6	20.5	19.3 ± 2.2	23.0	16.2 ± 1.4	19.8
τ_1, s	31.0 ± 4.4	19.8	32.3 ± 2.5	22.4	29.7 ± 2.1	20.4	26.9 ± 2.1	24.3
A_1, ml/min	3,605 ± 159	2.0	3,825 ± 202	3.3	3,680 ± 138	2.3	3,565 ± 107	3.0
td_2, s	136.1 ± 15.3 (n = 5)	16.9	207.8 ± 29.1 (n = 5)	28.7	179.1 ± 13.7 (n = 6)	20.6	140.1 ± 21.1 (n = 6)	21.2
τ_2, s	157.3 ± 38.1 (n = 5)	25.4	148.8 ± 49.3 (n = 5)	44.5	163.4 ± 39.7 (n = 6)	17.4	108.6 ± 38.2 (n = 6)	29.3
A_2', ml/min	475 ± 101 (n = 5)	39.4	532 ± 92 (n = 5)	30.2	269 ± 65 (n = 6)	41.1	371 ± 89 (n = 6)	35.7
A_2' old, ml/min			475 ± 92 (n = 5)				333 ± 74 (n = 5)	
TA $\dot{V}O_{2\ max}$, s	344 ± 66		207 ± 34		187 ± 32		264 ± 61	
T_{lim} @ $\dot{V}O_{2\ max}$, s	228 ± 47		577 ± 75*		319 ± 46		281 ± 73	
EE $\dot{V}O_2$, ml·kg^{-1}·min^{-1}	62.7 ± 1.3		66.8 ± 1.5*		62.0 ± 0.4		61.6 ± 1.1	
EE HR, beats/min	176 ± 3		179 ± 3		177 ± 3		178 ± 4	
EE [La], mmol/l	7.7 ± 0.6		7.2 ± 0.7		9.5 ± 0.8		9 ± 1.1	

Values are means ± SE. CV mean coefficient of variation estimated by the bootstrap method; A_1 and A_2', amplitude terms for $\dot{V}O_2$; td_1 and td_2, time delays to onset of each component; τ_1 and τ_2, time constants of each component; A_2' old, amplitude of the posttraining slow component obtained when the subject reached his pretraining T_{lim} (only 5 and 6 subjects demonstrated a slow component of the $\dot{V}O_2$ kinetics in the hypoxia and normoxia groups, respectively); EE $\dot{V}O_2$, end-exercise oxygen uptake; TA $\dot{V}O_{2max}$, time to reach pretraining $\dot{V}O_{2max}$; T_{lim} at $\dot{V}O_{2max}$, time sustained at pretraining $\dot{V}O_{2max}$; EE HR and EE [La], end-exercise values for heart rate and blood lactate, respectively. Significant differences between Pre vs. Post values, *$P < 0.05$.

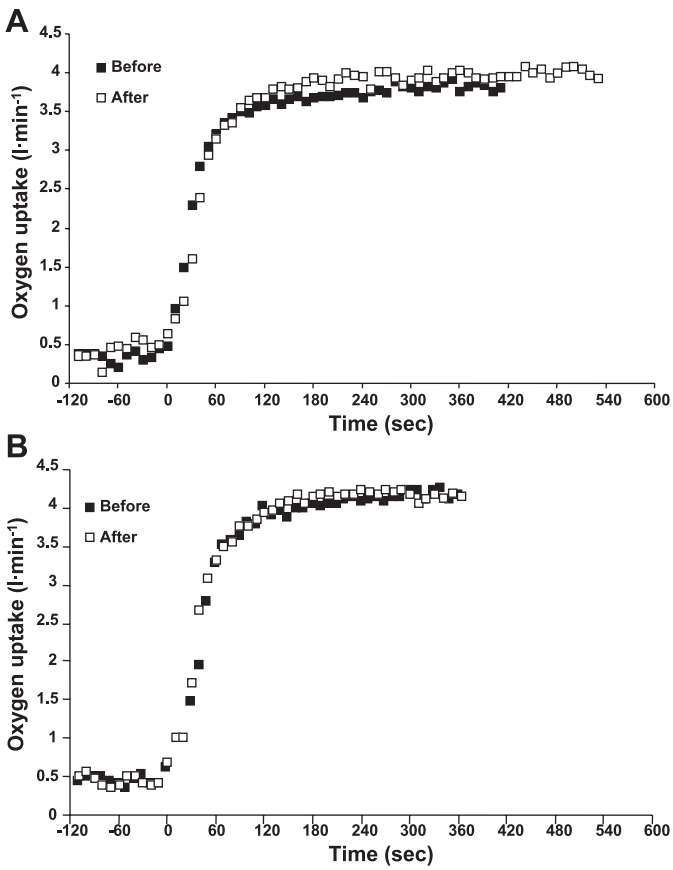

Fig. 4. Kinetics of the oxygen uptake ($\dot{V}O_2$) response of one representative individual from the Hyp group (*A*) and Nor group (*B*) during the all-out run at the pretraining minimal velocity associated with $\dot{V}O_{2\ max}$, before (■) and after (□) the 6 wk of intermittent hypoxia training program. Note that T_{lim} changes appeared, despite no modification in the kinetics of the $\dot{V}O_2$ response in both subjects.

component of $\dot{V}O_2$ kinetics was consistently observed in only five subjects in the Hyp and six subjects in the Nor group, respectively. Its computed amplitude (A_2') did not change after training, even when expressed at similar exercise time after vs. before training (A_2'old). Neither $\dot{V}O_2$ kinetics alterations nor IET_N-derived factors significantly correlated with the observed modifications in T_{lim}, either in absolute or in delta (after vs. before) values. There was no difference between groups in TA $\dot{V}O_{2\ max}$ or in T_{lim} at $\dot{V}O_{2\ max}$ before training (Table 4). However, T_{lim} at $\dot{V}O_{2\ max}$ significantly improved by 2.5 times only in the Hyp group after IHT, without modification of TA $\dot{V}O_{2\ max}$.

DISCUSSION

Major Findings

This study demonstrates that, when the hypoxic sessions of an IHT program features moderate duration (24–40 min) and high intensity (VT_2), significant improvements of $\dot{V}O_{2\ max}$ are obtained in already trained athletes, not only at altitude but also at sea level. Despite similar total training load (i.e., absolute and relative values), no such amelioration in the maximal rate of O_2 fluxes was observed in a control group exercising under permanent normoxia. The second finding of this work is that the present IHT program significantly lengthened T_{lim}, specifically in the Hyp group, without significant changes in $\dot{V}O_2$ kinetics. These results suggest that IHT did not change the control of O_2 flux adjustment to high-intensity exercise in competitive runners. Moreover, T_{lim} improvement in the Hyp group was correlated neither with $\dot{V}O_{2\ max}$ nor with ventilatory thresholds changes.

Maximal Aerobic Capacity and Ventilatory Thresholds

In hypoxia. This study demonstrates that the present IHT program elicits significant improvements of maximal and submaximal running velocities under hypoxia ($v\dot{V}O_{2\ max}$, vVT_2,

and $v\dot{V}T_1$). Accordingly, all of the athletes of the Hyp group required an increase of the training velocity under hypoxia (+0.4 km/h) to maintain the initial HR values throughout the 6-wk IHT program. Since no RE changes resulted from the training period, the improvements observed in running speeds are mainly due to significant increases in the associated O_2 flux rates in the Hyp group only ($\dot{V}O_{2\,max}$ and $\dot{V}O_2$ at the ventilatory thresholds). These findings expand the observations reported by Terrados et al. (43) in professional cyclists, demonstrating a specific increase of exercise capacity under hypoxia after altitude training only. Moreover, the present data also extend to already trained athletes the results obtained in untrained subjects, in which some consensus has been reach about the beneficial effect of hypoxic training on $\dot{V}O_{2\,max}$ at altitude (21, 47).

At sea level. The effects of IHT on the aerobic performance capacity at sea level remains highly debated, especially in trained subjects (32). Despite both groups improving their running velocities at sea level ($v\dot{V}O_{2\,max}$ and $v\dot{V}T_2$) in quite near proportions, the underlying physiological adaptations may well have been different. $v\dot{V}O_{2\,max}$ and $v\dot{V}T_2$ increased in the Nor group, through concomitant changes of $\dot{V}O_2$ and RE values (although not statistically significant). Conversely, one important result of this study is that the running speed improvements of the Hyp group were associated with increases in $\dot{V}O_{2\,max}$ and $\dot{V}O_2$ at VT_2, with no RE alterations. These findings suggest that a normoxic training effect was present in the Nor group over the 6-wk period and that this effect was further potentialized by IHT in the Hyp group, through an additional effect of IHT vs. normoxic training on aerobic power. This amelioration of aerobic power in the Hyp group is further exemplified by the increased $\dot{V}O_2$ at exhaustion during the all-out test. According to the specific intensity and duration of the present hypoxic training sessions, our results are in agreement with previous observations (38, 44, 46). Studies reporting no improvement in $\dot{V}O_{2\,max}$ after IHT either used lower hypoxic exercise intensity (at VT_1) (46) or shorter hypoxic exercise bouts (0.5–1 min) (44). On the other hand, similar increase in $\dot{V}O_{2\,max}$ has been recently reported with an IHT model, including longer periods of hypoxic exercise (2–12 min) (38). A specific oxygen-sensing transcription factor, the hypoxia-inducible factor-1α (HIF-1α), is expected to play a pivotal role for the functional adaptations to hypoxic training (1, 11, 47). Of note, the duration and intensity of the hypoxic exercise bouts included in the present IHT model are in good agreement with the properties of HIF-1α expression at the cellular level in humans. Not only does the half-time of the HIF-1α response to hypoxia fall in the range of 12–13 min (25), but also the magnitude of this response varies exponentially with the degree of Hyp in the physiological range (26). These observations further reinforce the necessity to combine a minimum duration and intensity of hypoxic exercise in IHT programs, to reduce oxygen pressure in the active muscle (37) and achieve a substantial HIF-1α response, resulting in peripheral muscle adaptations. Consequently, present and previous results suggest that the combination of sufficient hypoxic exercise intensity and duration within IHT programs is of paramount importance to obtain significant performance ameliorations in already trained athletes. An additional advantage of the hypoxic sessions in the present IHT design (i.e., 19% of the total training time in the present study) is the possibility to maintain the usual training load, which could also participate in the $\dot{V}O_{2\,max}$ improvement that we observed.

Some of our findings let us consider that peripheral adaptations might have been involved. We observed that O_2p_{max} improved in the Hyp group only after IHT. Because O_2p represents the product of stroke volume with $\Delta(a\text{-}v)O_2$, and because invasive experiments have shown that O_2p_{max} is largely determined by $\Delta(a\text{-}v)O_2$ (35, 42), O_2p_{max} is likely to have increased via an $\Delta(a\text{-}v)O_2$-mediated mechanism after IHT, suggesting an enhanced tissue O_2 extraction. Because our study was not designed to investigate O_2 extraction, further studies are needed to verify this hypothesis. Nevertheless, several muscle changes have already been observed after hypoxic training programs in endurance-trained subjects, such as larger deoxygenation in active muscles (28) and, although not reaching significance, a 36% increase in capillary density (43), supporting the concept of an improved O_2 extraction after IHT. Moreover, modelization studies have suggested that exercising in Hyp may increase the relative contribution of peripheral factors (i.e., muscle perfusion, peripheral diffusion, and mitochondrial capacity) to O_2 delivery and utilization (14, 15, 19, 48). We believe that the intensity and duration of the hypoxic exercise bouts included in the present IHT program are sufficient to induce the signaling cascade initiated by HIF-1α, leading to molecular and tissue changes within the exercising skeletal muscles of our Hyp subjects (34). The results disclosed in the two companion papers of our study, appearing in the present issue, also support this concept, at least in part. Conversely, as far as O_2 transport is concerned, we observed that hemoglobin and Hct were similar in both groups, before vs. after training, in agreement with previous reports (22, 28, 34, 38, 43). Together with the unchanged maximum HR (HR_{max}), these results suggest that O_2 delivery capacity is unlikely to represent a major cause of the $\dot{V}O_{2\,max}$ improvement of the Hyp group after IHT.

T_{lim} at $v\dot{V}O_{2\,max}$ and Oxygen Kinetics

A major finding of the present study is that T_{lim} is specifically improved after IHT (+35%) but unchanged after normoxic training. Due to the exponential shape of the running velocity/time-to-fatigue relationship, our observed 3.7-min lengthening of T_{lim} suggests that larger improvements of endurance time at lower velocities may have occurred. Thus this observation can be considered as a hallmark of an enhanced performance capacity in middle and long-distance running events. Consequently, T_{lim} lengthening in the present study extends previous findings, demonstrating that 3 wk of IHT dramatically delayed fatigue during a submaximal constant-load test in elite triathletes (45).

To date, the mechanisms leading to T_{lim} improvement remain poorly understood. It has been proposed that normoxic training may lengthen the endurance time at a given absolute running velocity, due to increases of $v\dot{V}O_{2\,max}$ and/or submaximal running velocity (velocity at the lactate threshold), reducing the relative running speed the subjects have to sustain (i.e., expressed in percentage of the posttraining $v\dot{V}O_{2\,max}$) (12, 23). In the present study, we did not find any correlation between T_{lim} changes and alterations of maximal ($\dot{V}O_{2\,max}$) and sub-

maximal (ventilatory thresholds) $\dot{V}O_2$ nor with their associated velocities ($v\dot{V}O_{2\,max}$, vVT_2, and vVT_1). Therefore, it is unlikely that changes in O_2 fluxes (i.e., $\dot{V}O_{2\,max}$) and/or running velocities (i.e., $v\dot{V}O_{2\,max}$) are the major causes of the T_{lim} improvement that we observed. However, as $\dot{V}O_{2\,max}$ and ventilatory thresholds improved concomitantly with T_{lim} in the Hyp group, we cannot rule out the possible relevance of these changes, and this point warrants further investigations.

Alternatively, $\dot{V}O_2$ kinetics have also been proposed as a determinant of T_{lim} that may be improved after normoxic training. To the best of our knowledge, the effect of hypoxic training on $\dot{V}O_2$ kinetics has never been reported, especially in already trained athletes. A speeding of $\dot{V}O_2$ adjustment has been proposed as a potential contributor of the delayed fatigue after high-intensity training at sea level (13). These changes are expected to reduce the reliance toward anaerobic metabolisms for energy provision, which have been reported to amount to ~15% of energy expenditure during such T_{lim} testing (18). Nevertheless, we failed to observe such a mechanism, as illustrated by an unchanged fast component of $\dot{V}O_2$ kinetics, leading to unaltered O_2def in both experimental groups. Additionally, sea level training was often demonstrated to reduce the amplitude of the $\dot{V}O_2$ slow component, thereby contributing to improve exercise tolerance and delay fatigue (20). Again, we recorded no alterations in the $\dot{V}O_2$ slow component, even when expressed at consistent exercise time before vs. after IHT (A'_2old). Taken together, the unchanged fast and slow components of $\dot{V}O_2$ kinetics suggest that the dynamic control of O_2 fluxes is not a likely contributor to T_{lim} changes after IHT in already trained athletes. Therefore, neither the rates of O_2 fluxes nor $\dot{V}O_2$ kinetics significantly account for the T_{lim} lengthening that we observed, suggesting that IHT may improve T_{lim} by specific, hypoxic-related adaptations.

A 2.5 times longer T_{lim} at $\dot{V}O_{2\,max}$ was observed in the Hyp group after IHT, indicating an improved capacity to sustain high levels of O_2 fluxes close to or above pretraining $\dot{V}O_{2\,max}$, before exhaustion occurs. This observation appears, despite unchanged [La] values recorded at exhaustion during the all-out test after vs. before IHT. Collectively, these findings suggest either a slower rate of blood lactate accumulation and/or a better tolerance of high levels of blood lactate after IHT. This might be associated with a concomitant amelioration of metabolite exchange and/or removal, contributing to enhance cellular homeostasis, thereby delaying the time at which fatigue occurs. This idea has already been suggested by a previous study, demonstrating that T_{lim} is related to the capacity of lactate exchange and removal. Due to its coupled transport with H^+ (27), an improved lactate exchange and removal could have contributed to slow down the progressive lowering of muscle pH while running at pretraining $v\dot{V}O_{2\,max}$. Although purely speculative in the present study, additional supports for the peripheral hypothesis underlying the improvement of endurance performance capacity after IHT are presented in the two following papers appearing in this issue. The second companion paper of the present study suggests that IHT induces qualitative mitochondrial changes leading to an enhanced channeling of energy within the muscle cell, whereas the third companion paper shows that IHT training induces transcriptional changes, potentially mediated by HIF-1α, leading to enhanced metabolite exchanges and improved aerobic metabolism within the skeletal muscle cell.

Limitations of the Study

A limitation of the present study is related to the IHT design and management of training intensities. First, we speculated that VT_2 might be more effective in IHT designs than lower (i.e., VT_1) or higher (i.e., $v\dot{V}O_{2\,max}$) training intensities, because of the achievement of a unique combination of intensity and duration of the hypoxic training stimulus. Moreover, this protocol was chosen as it allowed the usual training load of athletes to be unaltered (Table 2). Nevertheless, we did not test this hypothesis in the present study by including additional experimental groups training at either lower or higher intensity during the hypoxic sessions. Therefore, it remains to be determined whether different hypoxic training intensities and durations elicit similar beneficial effects on endurance performance capacity in already trained athletes. Especially including a group trained at, or close to, VT_1, would have been helpful and remains to be done.

Second, only the Hyp group required its laboratory running speed to be increased at the end of *week 3* to maintain the initial HR level, raising the question as to whether the Hyp group may have trained harder than the Nor group. We believed that this possibility is not supported by the unchanged VT_2 at the end vs. the beginning of training, when expressed in percentage of posttraining $v\dot{V}O_{2\,max}$, indicating that both groups trained at the same relative intensity during the laboratory sessions (Table 3). Nevertheless, a different time course of training speed and $v\dot{V}O_{2\,max}$ improvements may have led to a transient increase (i.e., *weeks 4* and *5*) in relative training intensity, thereby potentially acting as a confounding factor in our results. We believe that this possibility should have been counterbalanced by the transient lower relative intensity that could be expected in the Hyp group just before training speed adjustments (i.e., *weeks 2* and *3*). Therefore, differences in relative training intensity, if present, may have probably played a minor role in the present study. Nevertheless, future studies need to incorporate serial $\dot{V}O_{2\,max}$ testing to completely eliminate this possibility.

On the same token, an additional T_{lim} test performed at the new $v\dot{V}O_{2\,max}$ after training (same relative intensity before vs. after training) could have been helpful to clarify the role of $\dot{V}O_{2\,max}$ and $v\dot{V}O_{2\,max}$ in the improvement of T_{lim} that we observed in the Hyp group (+35%). However, since both groups improved $v\dot{V}O_{2\,max}$ in quite near proportions, the posttraining T_{lim} was performed at a similar relative intensity in both groups (96 vs. 97% of the posttraining $v\dot{V}O_{2\,max}$ in the Hyp and Nor group, respectively). Therefore, although not performed at 100% of posttraining $v\dot{V}O_{2\,max}$, the changes in relative testing intensity are unlikely to account for the T_{lim} improvements that we observed.

On a methodological standpoint, it can be argued from our relatively low maximum respiratory exchange ratio and maximum [La] values (Table 3) that $\dot{V}O_{2\,max}$ might have been underestimated. Nevertheless, 67–89% of the subjects reached a true $\dot{V}O_2$ plateau (i.e., always at least 6 of 9 subjects in each test), and the HR_{max} were close to (97%) the theoretical HR_{max}. Moreover, $\dot{V}O_{2\,max}$ as well as HR_{max} were significantly higher

on the treadmill than the ones previously obtained on the cycle ergometer at the time of subjects' basal medical examination. Conversely, these parameters were similar between IET and T_{lim} testing. Therefore, we believe that true $\dot{V}O_{2\,max}$ has been at least closely approached. On the other hand, our RE values were estimated at moderate mainly aerobic (12 km/h) and high (18 and 15 km/h in normoxia and hypoxia, respectively) running speeds, yielding results consistent with previous reports (40, 41, 50). Nevertheless, since they were not measured at steady state during constant load exercise, these RE values must be interpreted with caution, until appropriate RE testing is done by further investigations.

In conclusion, the present study investigates the effects of a carefully calibrated IHT program, designed to avoid reduction in training load, by including high-intensity (VT_2) and moderate-duration (24–40 min) hypoxic sessions, into the usual normoxic training of already trained athletes. Such an IHT model provides an original framework, in which the metabolic stimulus is enhanced through hypoxic sessions, without altering the mechanical component of the usual training load. Significant improvements of several indexes of aerobic performance capacity were observed not only at altitude but also at sea level, including $\dot{V}O_{2\,max}$ and T_{lim}. Additionally, IHT did not significantly modify $\dot{V}O_2$ kinetics such that T_{lim} lengthening was correlated neither with changes in the rate of $\dot{V}O_2$ adjustment nor with $\dot{V}O_{2\,max}$ and ventilatory thresholds. Collectively, these findings suggest that the enhanced endurance performance capacity obtained with IHT might be due to specific muscle adaptations to hypoxic training. This hypothesis is further explored in the two following companion papers of our study appearing in the present issue.

ACKNOWLEDGMENTS

The authors thank all of the athletes for enthusiastic participation; the whole laboratory staff from the Département de Physiologie et des Explorations Fonctionnelles and the Equipe d'Accueil 3072 for daily technical support; M. François Piquard for statistical advices; and Valérie Bougault and Frédéric Daussin for contribution during the training sessions. The help of M. Fabio Borrani was greatly appreciated for the application of the bootstrap method to the statistical treatment of the $\dot{V}O_2$ kinetics.

GRANTS

This project was supported by grants from the International Olympic Committee and the Ministère Français de la Jeunesse et des Sports. The scientific and sport coordination were, respectively, assumed by Prof. Jean-Paul Richalet and M. Laurent Schmitt, to whom we express our sincere gratitude.

REFERENCES

1. **Ameln H, Gustafsson T, Sundberg CJ, Okamoto K, Jansson E, Poellinger L, and Makino Y.** Physiological activation of hypoxia inducible factor-1 in human skeletal muscle. *FASEB J* 19: 1009–1011, 2005.
2. **Barstow TJ and Mole PA.** Linear and nonlinear characteristics of oxygen uptake kinetics during heavy exercise. *J Appl Physiol* 71: 2099–2106, 1991.
3. **Beaver WL, Wasserman K, and Whipp BJ.** A new method for detecting anaerobic threshold by gas exchange. *J Appl Physiol* 60: 2020–2027, 1986.
4. **Billat LV and Koralsztein JP.** Significance of the velocity at $\dot{V}O_{2\,max}$ and time to exhaustion at this velocity. *Sports Med* 22: 90–108, 1996.
5. **Billat V, Lepretre PM, Heugas AM, Laurence MH, Salim D, and Koralsztein JP.** Training and bioenergetic characteristics in elite male and female Kenyan runners. *Med Sci Sports Exerc* 35: 297–304, 2003.
6. **Billat VL, Lepretre PM, Heubert RP, Koralsztein JP, and Gazeau FP.** Influence of acute moderate hypoxia on time to exhaustion at $v\dot{V}O_{2\,max}$ in unacclimatized runners. *Int J Sports Med* 24: 9–14, 2003.
7. **Borrani F, Candau R, Millet GY, Perrey S, Fuchslocher J, and Rouillon JD.** Is the $\dot{V}O_2$ slow component dependent on progressive recruitment of fast-twitch fibers in trained runners? *J Appl Physiol* 90: 2212–2220, 2001.
8. **Borrani F, Candau R, Perrey S, Millet GY, Millet GP, and Rouillon JD.** Does the mechanical work in running change during the $\dot{V}O_2$ slow component? *Med Sci Sports Exerc* 35: 50–57, 2003.
9. **Cavanagh PR and Williams KR.** The effect of stride length variation on oxygen uptake during distance running. *Med Sci Sports Exerc* 14: 30–35, 1982.
10. **Daniels J and Daniels N.** Running economy of elite male and elite female runners. *Med Sci Sports Exerc* 24: 483–489, 1992.
11. **Däpp C, Gassmann M, Hoppeler H, and Flück M.** Hypoxia-induced gene activity in disused oxidative muscle. In: *Hypoxia and Exercise*, edited by Roach R. New York: Springer, 2005, chapt. 16.
12. **Demarle AP, Heugas AM, Slawinski JJ, Tricot VM, Koralsztein JP, and Billat VL.** Whichever the initial training status, any increase in velocity at lactate threshold appears as a major factor in improved time to exhaustion at the same severe velocity after training. *Arch Physiol Biochem* 111: 167–176, 2003.
13. **Demarle AP, Slawinski JJ, Laffite LP, Bocquet VG, Koralsztein JP, and Billat VL.** Decrease of O_2 deficit is a potential factor in increased time to exhaustion after specific endurance training. *J Appl Physiol* 90: 947–953, 2001.
14. **Di Prampero PE.** Metabolic and circulatory limitations to $\dot{V}O_{2\,max}$ at the whole animal level. *J Exp Biol* 115: 319–331, 1985.
15. **Di Prampero PE.** A brief comment on the factors limiting maximal oxygen consumption in humans. *Eur J Appl Physiol* 80: 516–517, 1999.
16. **Durnin JV and Womersley J.** Body fat assessed from total body density and its estimation from skinfold thickness: measurements on 481 men and women aged from 16 to 72 years. *Br J Nutr* 32: 77–97, 1974.
17. **Effron B and Tibshirani RJ.** *An Introduction to the Bootstrap*. New York: Chapman and Hall, 1993.
18. **Faina M, Billat V, Squadrone R, De Angelis M, Koralsztein JP, and Dal Monte A.** Anaerobic contribution to the time to exhaustion at the minimal exercise intensity at which maximal oxygen uptake occurs in elite cyclists, kayakists and swimmers. *Eur J Appl Physiol* 76: 13–20, 1997.
19. **Ferretti G and di Prampero PE.** Factors limiting maximal O_2 consumption: effects of acute changes in ventilation. *Respir Physiol* 99: 259–271, 1995.
20. **Gaesser GA and Poole DC.** The slow component of oxygen uptake kinetics in humans. *Exerc Sport Sci Rev* 24: 35–71, 1996.
21. **Geiser J, Vogt M, Billeter R, Zuleger C, Belforti F, and Hoppeler H.** Training high–living low: changes of aerobic performance and muscle structure with training at simulated altitude. *Int J Sports Med* 22: 579–585, 2001.
22. **Hendriksen IJ and Meeuwsen T.** The effect of intermittent training in hypobaric hypoxia on sea-level exercise: a cross-over study in humans. *Eur J Appl Physiol* 88: 396–403, 2003.
23. **Heubert R, Bocquet V, Koralsztein JP, and Billat V.** Effect of 4 weeks of training on the limit time at $\dot{V}O_{2\,max}$. *Can J Appl Physiol* 28: 717–736, 2003.
24. **Hornbein TF and Schoene RB.** *High Altitude: An Exploration of Human Adaptation*. New York: Dekker, 2001.
25. **Jewell UR, Kvietikova I, Scheid A, Bauer C, Wenger RH, and Gassmann M.** Induction of HIF-1α in response to hypoxia is instantaneous. *FASEB J* 15: 1312–1314, 2001.
26. **Jiang BH, Semenza GL, Bauer C, and Marti HH.** Hypoxia-inducible factor 1 levels vary exponentially over a physiologically relevant range of O_2 tension. *Am J Physiol Cell Physiol* 271: C1172–C1180, 1996.
27. **Juel C.** Muscle pH regulation: role of training. *Acta Physiol Scand* 162: 359–366, 1998.
28. **Kime R, Karlsen T, Nioka S, Lech G, Madsen O, Saeterdal R, Im J, Chance B, and Stray-Gundersen J.** Discrepancy between cardiorespiratory system and skeletal muscle in elite cyclists after hypoxic training. *Dyn Med* 2: 4, 2003.
29. **Kuipers H, Verstappen FT, Keizer HA, Geurten P, and Van Kranenburg G.** Variability of aerobic performance in the laboratory and its physiological correlates. *Int J Sports Med* 6: 197–201, 1985.
30. **Laffite LP, Mille-Hamard L, Koralsztein JP, and Billat VL.** The effects of interval training on oxygen pulse and performance in supra-threshold runs. *Arch Physiol Biochem* 111: 202–210, 2003.

31. **Lamarra N, Whipp BJ, Ward SA, and Wasserman K.** Effect of interbreath fluctuations on characterizing exercise gas exchange kinetics. *J Appl Physiol* 62: 2003–2012, 1987.
32. **Levine BD.** Intermittent hypoxic training: fact and fancy. *High Alt Med Biol* 3: 177–193, 2002.
33. **Macfarlane DJ.** Automated metabolic gas analysis systems: a review. *Sports Med* 31: 841–861, 2001.
34. **Meeuwsen T, Hendriksen IJ, and Holewijn M.** Training-induced increases in sea-level performance are enhanced by acute intermittent hypobaric hypoxia. *Eur J Appl Physiol* 84: 283–290, 2001.
35. **Mortensen SP, Dawson EA, Yoshiga CC, Dalsgaard MK, Damsgaard R, Secher NH, and Gonzalez-Alonso J.** Limitations to systemic and locomotor limb muscle oxygen delivery and uptake during maximal exercise in humans. *J Physiol* 566: 273–285, 2005.
36. **Paterson DH and Whipp BJ.** Asymmetries of oxygen uptake transients at the on- and offset of heavy exercise in humans. *J Physiol* 443: 575–586, 1991.
37. **Richardson RS, Noyszewski EA, Kendrick KF, Leigh JS, and Wagner PD.** Myoglobin O_2 desaturation during exercise. Evidence of limited O_2 transport. *J Clin Invest* 96: 1916–1926, 1995.
38. **Roels B, Millet GP, Marcoux CJ, Coste O, Bentley DJ, and Candau RB.** Effects of hypoxic interval training on cycling performance. *Med Sci Sports Exerc* 37: 138–146, 2005.
39. **Rossiter HB, Howe FA, Ward SA, Kowalchuk JM, Griffiths JR, and Whipp BJ.** Intersample fluctuations in phosphocreatine concentration determined by ^{31}P-magnetic resonance spectroscopy and parameter estimation of metabolic responses to exercise in humans. *J Physiol* 528: 359–369, 2000.
40. **Saunders PU, Pyne DB, Telford RD, and Hawley JA.** Reliability and variability of running economy in elite distance runners. *Med Sci Sports Exerc* 36: 1972–1976, 2004.
41. **Saunders PU, Telford RD, Pyne DB, Cunningham RB, Gore CJ, Hahn AG, and Hawley JA.** Improved running economy in elite runners after 20 days of simulated moderate-altitude exposure. *J Appl Physiol* 96: 931–937, 2004.
42. **Stringer WW, Hansen JE, and Wasserman K.** Cardiac output estimated noninvasively from oxygen uptake during exercise. *J Appl Physiol* 82: 908–912, 1997.
43. **Terrados N, Melichna J, Sylven C, Jansson E, and Kaijser L.** Effects of training at simulated altitude on performance and muscle metabolic capacity in competitive road cyclists. *Eur J Appl Physiol* 57: 203–209, 1988.
44. **Truijens MJ, Toussaint HM, Dow J, and Levine BD.** Effect of high-intensity hypoxic training on sea-level swimming performances. *J Appl Physiol* 94: 733–743, 2003.
45. **Vallier JM, Chateau P, and Guezennec CY.** Effects of physical training in a hypobaric chamber on the physical performance of competitive triathletes. *Eur J Appl Physiol* 73: 471–478, 1996.
46. **Ventura N, Hoppeler H, Seiler R, Binggeli A, Mullis P, and Vogt M.** The response of trained athletes to six weeks of endurance training in hypoxia or normoxia. *Int J Sports Med* 24: 166–172, 2003.
47. **Vogt M, Puntschart A, Geiser J, Zuleger C, Billeter R, and Hoppeler H.** Molecular adaptations in human skeletal muscle to endurance training under simulated hypoxic conditions. *J Appl Physiol* 91: 173–182, 2001.
48. **Wagner PD.** A theoretical analysis of factors determining $\dot{V}O_{2\,max}$ at sea level and altitude. *Respir Physiol* 106: 329–343, 1996.
49. **Wasserman K, Hansen JE, Sue DY, Whipp BJ, and Casaburi R.** *Principles of Exercise Testing and Interpretation*. Philadelphia, PA: Williams & Wilkins, 1994.
50. **Weston AR, Mbambo Z, and Myburgh KH.** Running economy of African and Caucasian distance runners. *Med Sci Sports Exerc* 32: 1130–1134, 2000.
51. **Whipp BJ and Ozyener F.** The kinetics of exertional oxygen uptake: assumptions and inferences. *Med Sport* 51: 139–149, 1998.