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ORIGINAL ARTICLE

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Early effects of exercise training on $\dot{V}O_2$ on- and off-kinetics in 50-year-old subjects

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Abstract We tested the hypothesis that, in healthy middle-aged subjects (n=11, age 51.0±3.0 years, x ±SD), the effects of exercise training on pulmonary O_2 uptake ($\dot{V}O_2$) on- and off-kinetics would appear earlier than those on peak $\dot{V}O_2$. The subjects underwent a standard training program (combined endurance and resistance training) in a health club, and were evaluated before training (''time 0'', T0), and after 7 (T7), 15 (T15), 30 (T30), 60 (T60) and 90 (T90) days of training. Breath-by-breath pulmonary O_2 uptake ($\dot{V}O_2$), heart rate (HR), systolic (SBP) and diastolic blood pressure, and capillary blood lactate concentration ($[La]_b$) were determined at rest and at each workload ($\dot{w}$ during a cycle ergometer incremental exercise test. The ''heart rate · blood pressure product'' was calculated as (HR·SBP). The day following the incremental test, the subjects performed three repetitions of a square-wave exercise at 50% of $\dot{V}O_{2peak}$, for the determination of pulmonary $\dot{V}O_2$ on- and off-kinetics. $\dot{V}O_{2peak}$ and $[La]_{bpeak}$ tended to increase with training; the increases became significant at T60 or T90. HR_{peak} and $(HR \cdot SBP)_{peak}$ were unaffected by training. The time constant of the ''primary'' component of the $\dot{V}O_2$ on-kinetics (τ_2) was 46.9±17.3 s (T0), 38.1±14.2 s (T7), 34.4±12.6 s (T15), 28.8±6.8 s (T30), 30.2±8.0 s (T60), and 30.4±12.4 s (T90); a significant difference compared to T0 was observed from T15 onward. From T15 onward, τ_2 were not significantly different from values obtained (29.2±5.3 s) from a group of healthy untrained young controls (n=7, 21.6±0.5 years). The same pattern of change as a function of training was described for the $\dot{V}O_2$ off-kinetics. It is concluded that in 50-year-old subjects $\dot{V}O_2$ on- and off-kinetics are more sensitive to exercise training than other physiological variables determined at peak exercise.

Keywords Aging · Oxygen uptake kinetics · Training

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Introduction

Upon a step transition from rest to exercise, the rate of increase of oxygen uptake ($\dot{V}O_2$) is slower than that of power output, and follows a time-course often termed $\dot{V}O_2$ on-kinetics. Pulmonary $\dot{V}O_2$ on-kinetics, as usually determined in a clinical setting or in the exercise physiology laboratory, matches rather closely the on-kinetics of $\dot{V}O_2$ directly determined across exercising limbs [14]. More specifically, the ''primary'' component, or ''phase 2'', of pulmonary $\dot{V}O_2$ on-kinetics should closely reflect the kinetics of adjustment of oxidative metabolism at the skeletal muscle level [24]. It is generally agreed that a ''metabolic inertia'' within muscles during transitions to low-intensity exercise is the main limiting factor for $\dot{V}O_2$ on-kinetics [7, 13, 23]. $\dot{V}O_2$ on-kinetics during cycling exercise is known to get progressively slower with age [3, 4, 8, 9, 11]. Decreases in the rate of adjustment of skeletal muscle oxidative metabolism to increases in work rate increase the need for substrate-level phosphorylation and cause a greater disturbance of cellular and organ homeostasis; this has obvious implications for exercise capacity and muscle fatigue. The effects of endurance training on $\dot{V}O_2$ on-kinetics in young subjects are known to occur very early (after 4 days of training), well before changes in maximal oxygen uptake ($\dot{V}O_{2max}$) are observed [18]. It is also known that a 6-month endurance training program in aged subjects (≅70 years old) results in faster $\dot{V}O_2$ on-kinetics [2]. To

our knowledge, no data are available on the time-course of the effects of exercise training on $\dot{V}O_2$ kinetics in middle-aged and old subjects. Should the response be similar to that of young subjects [18], the positive effects of exercise training in middle-aged and old subjects would also become manifest after relatively short training programs, before changes in other physiological variables (such as $\dot{V}O_{2max}$) could appear. We tested this hypothesis in the present study. This was conducted on a group of middle-aged (≅50-year-old) previously untrained healthy subjects, undergoing a 3-month standard training program (including both ''aerobic'' and ''resistance'' training) in a health club, following the guidelines recommended by the American College of Sports Medicine (ACSM) [1]. The subjects were tested at different times during the program, in order to allow the investigators to detect possible early effects of exercise training on $\dot{V}O_2$ on- and off-kinetics, as well as on other physiological variables determined during submaximal constant-load bicycle exercise and during incremental bicycle exercise to exhaustion.

Materials and methods

Subjects

The study was conducted on 11 healthy previously untrained middle-aged males [age 51.0±3.0 years (x ±SD), height 176.9±5.6 cm, body mass 86.0±11.8 kg, BMI 27.3±2.5]. For reference, a group of young healthy untrained males (YG) (medical school students, n=7; age 21.6±0.5 years, height 176.1±7.8 cm, body mass 72.1±5.4 kg, BMI 23.3±1.5) was also tested. The subjects were fully informed of any risks and discomforts associated with these experiments before giving their written, informed consent to participate in this study, which was approved by the ethics committee of the Institutional Review Board of Istituto di Tecnologie Biomediche Avanzate, Consiglio Nazionale delle Ricerche, Milan, Italy.

Training program

Middle-aged subjects underwent a 90-day standard training program. The subjects trained three times a week, and each training session included, after some warm-up exercises, 30 min of cycle exercise at a workload corresponding to ≅50% of the difference between resting heart rate and peak heart rate (determined during the incremental exercise, see below), as well as 30 min of ''resistance'' (strength) training [six different exercises involving the major muscle groups of the upper and lower limbs, with 12–15 repetitions of each exercise at ≅70% of one repetition maximum (1RM) determined serially during the training period], according to a standard protocol utilized in the club and recommendations by ACSM [1]. Training sessions were supervised closely by experienced personnel. YG did not undergo any training program.

Exercise protocols

All tests were carried out under close medical supervision, and the subjects were continuously monitored by 12-lead electrocardiography (ECG). The tests were carried out in the afternoon, a few hours after a light meal. Ambient temperature in the laboratory was kept at ≅20°C (relative humidity 55–60%). Before training (T0) and 7 (T7), 15 (T15), 30 (T30), 60 (T60) and 90 (T90) days after the beginning of the training program the subjects underwent an incremental bicycle exercise (*incremental exercise*: starting from rest, 30 W added every 3 min) to voluntary exhaustion, which was defined as the inability to sustain the recommended pedaling frequency of ≅60 revolutions/min despite vigorous encouragement by the operators. An electromagnetically braked cycle ergometer (Cardioline STS 3) was utilized. Pedaling frequency was digitally displayed to the subjects throughout the tests.

The day following the incremental test, the subjects performed three repetitions of a square-wave exercise (*constant load exercise*) on the same bicycle ergometer, at a workload corresponding to 50% of the ''peak'' power output (see below) determined the day before. A constant load corresponding to 50% of $\dot{V}O_{2peak}$ was utilized in order to avoid complexities associated with the so-called ''slow component'' of $\dot{V}O_2$ on-kinetics [12], which should become manifest at higher percentages of $\dot{V}O_{2peak}$. Pedaling frequency was kept at ≅60 revolutions/min. At least 10–15 min of rest was observed between repetitions. On-transitions were from rest (subjects sitting on the cycle ergometer, with feet tightly strapped to the pedals) to the imposed load, which was attained in ≅3 s. Off-transitions were from the imposed load to rest. Orders to start and stop pedaling were given by voice to the subjects, without warning.

Investigated variables

Pulmonary ventilation ($\dot{V}_E$), $\dot{V}O_2$ and CO_2 output ($\dot{V}CO_2$) were determined breath-by-breath by a computerized metabolic cart (Sensor Medics Vmax29c). Expiratory flow measurement was performed by a mass flow sensor (hot wire anemometer), calibrated before each experiment by a 3-liter syringe at three different flow rates. Tidal volume (V_T) and $\dot{V}_E$ were calculated by integration of the flow tracings recorded at the mouth of the subject. $\dot{V}O_2$ and $\dot{V}CO_2$ were determined by continuously monitoring PO_2 and PCO_2 at the mouth of the subject throughout the respiratory cycle and from established mass balance equations, after alignment of the expiratory volume and expiratory gases tracings and A/D conversion. Calibration of the O_2 and CO_2 analyzers was performed before each experiment by utilizing gas mixtures of known composition. Digital data were transmitted to a personal computer and stored on disk. $\dot{V}O_2$ and $\dot{V}CO_2$ were expressed in STPD, $\dot{V}_E$ in BTPS. The gas exchange ratio (R) was calculated as $\dot{V}CO_2/\dot{V}O_2$.

Heart rate (HR) was determined beat-by-beat from the R–R intervals by a cardiotachometer coupler (PE4000, Polar Electro). Systolic (SBP) and diastolic blood pressures were determined during the last 30 s of each step of the incremental test by a sphygmomanometer. The ''HR·SBP product'' (taken as an indication of cardiac work) was also calculated. The YG group did not have their blood pressure measured. Arterial blood O_2 saturation (SaO_2) was continuously monitored by pulse oxymetry (Biox 3740 Pulse Oximeter, Ohmeda) at the earlobe.

At rest, during the last 30 s of each step of the incremental test and at different times during the first minutes of recovery after the incremental and the constant load tests, 20 µl of earlobe capillary blood was obtained for the determination of lactate concentration ($[La]_b$) by an enzymatic method (ESAT 6661 Lactat, Eppendorf).

Data analysis

Steady-state values of $\dot{V}_E$, $\dot{V}O_2$, $\dot{V}CO_2$, SaO_2, and HR for each workload of the incremental test were obtained by calculating averages of breath-by-breath or beat-by-beat values over the last 30–40 s of each workload. Resting values were obtained by calculating averages during ≅1 min of rest. Values obtained during the last 30 s of the incremental exercise were considered ''peak'' values.

As for the constant load exercise, breath-by-breath $\dot{V}O_2$ data and beat-by-beat HR data obtained during the three repetitions were time-aligned and superimposed for each subject. Resting and steady-state values were calculated over a 1-min interval at rest, during the last minute of exercise, and between the 4th and 5th

minutes of recovery. For on-kinetics (rest-to-exercise) and off-kinetics (exercise-to-rest) $\dot{V}O_2$ and HR kinetics analysis, data smoothing was obtained by calculating a five-point moving average.

As expected, $\dot{V}O_2$ rose by a small amount (phase 1) as soon as exercise started, followed by a predominant exponential rise (phase 2) to the new steady state (phase 3) (see [24]). Phase 1 is usually termed ''cardiodynamic'', and represents the circulatory transit delay between the exercising muscles and the lungs. During this phase the increase in pulmonary $\dot{V}O_2$ is due to the increased pulmonary blood flow. The onset of phase 2 was determined according to standard criteria [24]. $\dot{V}O_2$ on-kinetics during phase 2 (taken as an indication of the rate of adjustment of oxidative metabolism at the skeletal muscle level [24]) was evaluated by fitting an exponential function of the type

$$y=a+b\left[1-e^{-(t-c)/d}\right] \quad (1)$$

and parameter values (c and d) were determined that yielded the lowest sum of squared residuals. In Eq. 1, a indicates the baseline value (i.e. the $\dot{V}O_2$ value at the beginning of phase 2), b the amplitude ($ampl_2$) between a and the new steady-state value ($a+b$), c the time delay (TD_2) and d the time constant (τ_2) of the variable. An exponential function was not fitted to $\dot{V}O_2$ data during phase 1, due to the limited number of data points. As for phase 1, only the amplitude ($ampl_1$) of the response was determined. $\dot{V}O_2$ off-kinetics was evaluated by utilizing a mono-exponential function equivalent to that presented as Eq. 1.

As for HR on-kinetics, they were evaluated by fitting a polynomial function to the HR versus time data, and the half-time ($t_{1/2}$) of the response was calculated by solving the function for the time value at which the function reached 50% of the difference between the baseline and the new asymptotic value ($t_{1/2}$ HR on-). $\dot{V}O_2$ on- and HR on-kinetics were evaluated differently because the different phases of the two variables have different physiological significance. As for $\dot{V}O_2$, as mentioned above, the monoexponential increase during phase 2 is thought to be the one relevant to the functional evaluation of the rate of adjustment of oxidative metabolism at the skeletal muscle level (see e.g., [24]). As for HR, the on-kinetics can be subdivided into sequential phases because of the sequential involvement of parasympathetic withdrawal and sympathetic stimulation at the onset of exercise [21]. For the purposes of the present study we were not interested in discriminating between these different components. Thus, we decided to utilize a parameter that allows the overall rate of adjustment of the variable to be evaluated, such as the $t_{1/2}$ of the response.

Statistical analysis

Data were analyzed using a two-way analysis of variance, between/within split-plot design, with repeated measures. Training status was the between variable, and time was the within variable. When significant differences were found, a Tukey's post-hoc test was used to discriminate when significant differences occurred. Significance was set at the 0.05 level. Data are presented as arithmetic means ± standard deviation (x ±SD).

Results

Incremental exercise

Peak values of some cardiovascular and metabolic variables before and during the training program are shown in Fig. 1. Reference values obtained in YG are also shown in the figure (as mentioned above, blood pressure measurements from this group were not obtained). $\dot{V}O_{2peak}$ and $[La]_{bpeak}$ showed a tendency to increase with training; these increases became statistically significant at T60 (for $\dot{V}O_{2peak}$) or T90 (for $[La]_{bpeak}$). HR_{peak} and $(HR \cdot SBP)_{peak}$ were unaffected by training. At the end of the training program (T90), $\dot{V}O_{2peak}$ and HR_{peak} were still significantly lower than the corresponding values obtained in YG. No subject showed arterial oxyhemoglobin desaturation during the tests, the SaO_2 values ranging between 96% and 99%.

Steady-state values of some cardiovascular and metabolic variables were also determined at 90 W, i.e., at the

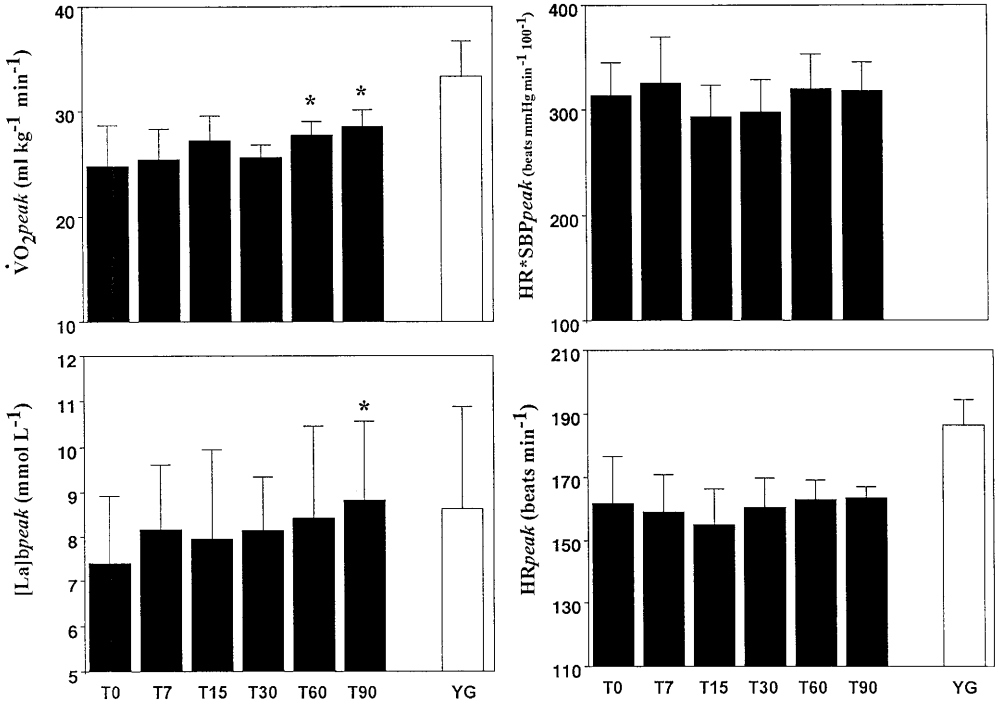

Fig. 1 Peak values (x ±SD) of some cardiovascular and metabolic variables obtained from middle-aged subjects during the 3-month training program. Reference values obtained from healthy young untrained subjects (*YG*) are also shown. *,**Significantly different from values at T0, $P<0.05$, 0.01. See text for further details. (*T0* Before training, *T7* 7 days of training, *T15* 15 days of training, *T30* 30 days of training, *T60* 60 days of training, *T90* 90 days of training)

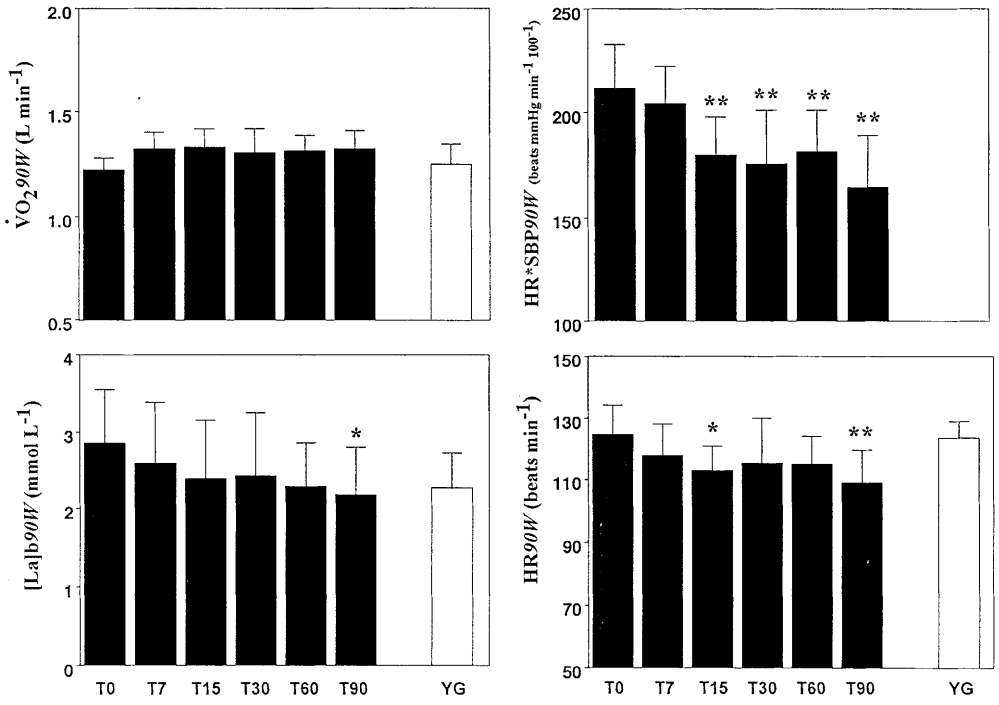

Fig. 2 Values (x ±SD) of some ventilatory, cardiovascular, and metabolic variables obtained at submaximal workload (90 W) during the incremental exercise. Reference values obtained from YG are also shown (except for HR·SBP). *,**Significantly different from values at T0, P<0.05, 0.01. See text for further details

Table 1 Steady state values of the main ventilatory, cardiovascular and metabolic variables during the constant-workload test (50% of $\dot{V}O_{2peak}$). Data are shown as x ±SD. (*HR* Heart rate, *R* gas exchange ratio, $\dot{V}CO_2$ CO_2 output, $\dot{V}_E$ pulmonary ventilation, $\dot{V}O_2$ O_2 uptake)

		Workload (W)	HR (beats · min^{-1})	$\dot{V}O_2$ (l · min^{-1})	$\dot{V}CO_2$ (l · min^{-1})	R	$\dot{V}_E$ (l · min^{-1})
T0	x	80	120	1.23	1.20	0.97	36.7
	SD	10	5	0.13	0.14	0.04	4.4
T7	x	87	112*	1.25	1.23	0.99	38.7
	SD	12	5	0.10	0.10	0.05	3.4
T15	x	92*	113*	1.32	1.31	1.00	40.3
	SD	9	4	0.11	0.13	0.03	4.5
T30	x	94*	114*	1.25	1.21	0.97	37.8
	SD	10	5	0.12	0.11	0.03	4.4
T60	x	99*	117	1.43**	1.44*	1.01	44.6*
	SD	15	5	0.19	0.21	0.03	7.8
T90	x	100**	114*	1.41**	1.40*	0.99	43.2*
	SD	13	4	0.15	0.15	0.04	5.1
YG	x	109	135	1.58	1.48	0.95	38.0
	SD	10	7	0.17	0.11	0.06	2.2

*,** Significantly different from value of each variable at T0 (P<0.05, 0.01). See text for further details

highest submaximal load carried out by all subjects in the different tests. The data are shown in Fig. 2, together with the corresponding values obtained in YG. $\dot{V}O_{290W}$ was unaffected by training, and the values obtained from the middle-aged subjects were not different from those obtained from YG. $[La]_{b90W}$, HR_{90W} and $(HR{\cdot}SBP)_{90W}$ showed a tendency to decrease with training; such decreases became statistically significant at T15 (for HR_{90W} and $[HR{\cdot}SBP]_{90W}$) or at T90 (for $[La]_{b90W}$). $[La]_{b90W}$ at T90 was not different from the value obtained from YG.

Constant-load exercise

Steady-state values of the main ventilatory, cardiovascular and metabolic variables during the constant-load tests are shown in Table 1. Figure 3 shows breath-by-breath $\dot{V}O_2$ values at rest, during 5 min at 50% of $\dot{w}_{peak}$ and during recovery in a typical middle-aged subject, at T0 and at T15: 15 days of training resulted in faster $\dot{V}O_2$ on- and off-kinetics. In none of the subjects was a slow component of $\dot{V}O_2$ on-kinetics [12] observed. The absence of a $\dot{V}O_2$ slow component was checked by visually inspecting the breath-by-breath data, and by calculating average $\dot{V}O_2$ values during the last 30 s of the 3rd and the 5th minutes of exercise: no significant differences were observed, indicating that the subjects had substantially reached a steady-state by the end of the 3rd minute.

As for parameters related to the $\dot{V}O_2$ on-kinetics, the amplitude ($ampl_1$) of phase 1, together with amplitude ($ampl_2$), time delay (TD_2) and time-constants (τ_2) of phase 2 are shown for middle-aged subjects and for YG

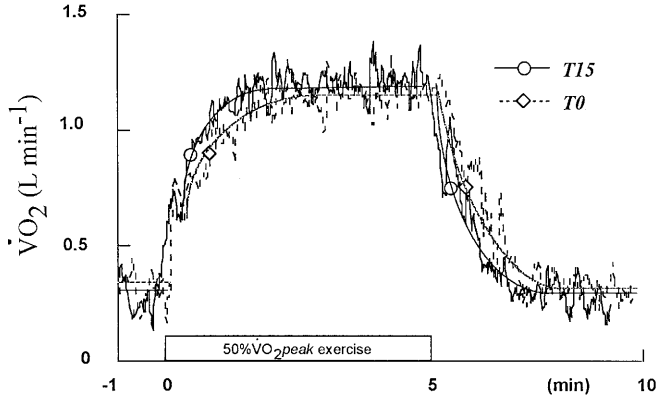

Fig. 3 Breath-by-breath oxygen uptake ($\dot{V}O_2$) at rest, during 5 min at 50% of peak $\dot{V}O_2$ and during recovery, in a typical middle-aged subject, before training (T0) and after 15 days of training (T15). The monoexponential functions (*solid lines*) fitted through the data are also shown. The time constants for the on- (τ_2) and off- (τ_{off}) functions are indicated by the symbols (○, ◇) see text for further details

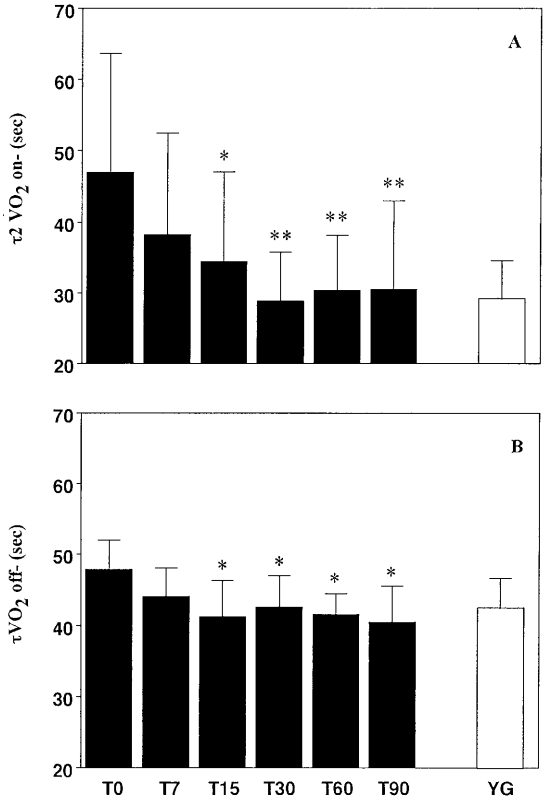

Fig. 4A, B Time constants (x ±SD) determined during phase 2 of $\dot{V}O_2$ on-kinetics (τ_2), and during $\dot{V}O_2$ off-kinetics ($\tau_{off\text{-}}$), as a function of training. *,**Significantly different from the value of $\dot{V}O_2$ on- and off- at T0, P<0.05, 0.01. See text for further details

in Table 2. $ampl_1$ and TD_2 were unaffected by training, and the values obtained from middle-aged subjects were not significantly different from those obtained from YG. As a consequence of differences of $\dot{w}$ during the con-

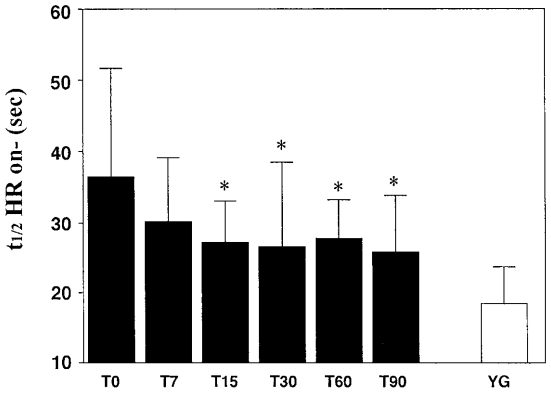

Fig. 5 Half-times ($t_{1/2}$) (x ±SD) calculated for heart rate (*HR*) on-kinetics in middle-aged subjects, as a function of training. *,**Significantly different from value of HR on- at T0, P<0.05, 0.01. See text for further details

Table 2 Parameters of $\dot{V}O_2$ on-kinetics as a function of training. ($Ampl_1$ Amplitude of phase 1, TD_2 duration of phase 1, $Ampl_2$ amplitude of phase 2, τ_2 time constant of phase 2)

		T0	T7	T15	T30	T60	T90	YG
$Ampl_1$	x	0.31	0.36	0.42	0.39	0.35	0.37	0.49
(l · min^{-1})	SD	0.11	0.10	0.08	0.08	0.11	0.11	0.14
TD_2 (s)	x	24.0	27.3	28.8	29.9	24.6	25.9	25.2
	SD	20.6	18.1	16.3	23.4	24.7	26.7	17.4
$Ampl_2$	x	0.60	0.58	0.58	0.55	0.74	0.71	0.74
(l · min^{-1})	SD	0.12	0.14	0.14	0.12	0.17	0.18	0.17
τ_2 (s)	x	46.9	38.1	34.4*	28.8**	30.2**	30.4**	29.2
	SD	18.3	14.2	12.6	6.8	8.0	12.4	5.3

*,**Significantly different from value of each parameter at T0 (P<0.05, 0.01). See text for further details

stant-load tests (see Table 1), $ampl_2$ values were lower in middle-aged subjects than in YG, with the exception of T90. τ_2 for the $\dot{V}O_2$ on-kinetics are also shown in Fig. 4A. τ_2 became lower (i.e., the $\dot{V}O_2$ on-kinetics during phase 2 became faster) with training, beginning from T7. A statistically significant difference, compared to T0, was observed from T15 onward. The decrease in t_2 showed a plateau from T30 onward. From T15 onward, τ_2 values obtained from middle-aged subjects were not significantly different from those obtained from YG. The τ values for the monoexponential $\dot{V}O_2$ decrease during the off-phase ($\tau_{off\text{-}}$) are shown in Fig. 4B. The pattern of change of $\tau_{off\text{-}}$, as a function of training, was the same as that described for τ_2 on-: a decrease (i.e., a faster kinetics) from T7 onward; a statistically significant difference, compared to T0, from T15 onward; a plateau from T15 onward; and no significant difference, compared to the value obtained from YG, from T15 onward.

$t_{1/2}$ HR on- values are shown in Fig. 5, for middle-aged subjects and YG. $t_{1/2}$ HR on- became lower with training (indicating faster kinetics), and a statistically significant difference compared to T0 was observed beginning from T15. Also for $t_{1/2}$ HR on-, the decrease

reached a plateau from T15 onward. $t_{1/2}$ HR on- values from middle-aged subjects, however, were always significantly higher than those observed in YG.

Discussion

The main finding of the present study is that, in previously untrained middle-aged (≅50-year-old) subjects, a standard training program adopted in health clubs results very quickly (i.e., after 1–2 weeks) in adaptations of physiological variables determined during submaximal (≅50% of $\dot{V}O_{2peak}$) exercises, such as faster $\dot{V}O_2$ on- and off-kinetics, faster HR on-kinetics, and lower HR and (HR·SBP) at steady-state. These adaptations, occurring much earlier than others described at peak workload (such as increase in $\dot{V}O_{2peak}$), may significantly and positively affect exercise tolerance, and may have important implications for the prevention of cardiovascular diseases.

Of particular interest, also from a physiological standpoint, is the finding of the faster $\dot{V}O_2$ on- and off-kinetics during the training program, occurring after only 1–2 weeks of training (i.e., three to six training sessions, corresponding to only 1.5–3 h of relatively low-intensity ''aerobic'' training). These observations extend previous results obtained by Phillips et al. [18], in their study of young subjects, to middle-aged subjects. These authors demonstrated that, in ≅20 year-old subjects, a training-induced acceleration of $\dot{V}O_2$ on-kinetics can be found as early as after 4 days of training, well before changes in $\dot{V}O_{2peak}$ are observed. Up till at least 50 years of age, therefore, $\dot{V}O_2$ on-kinetics appears to be particularly sensitive to exercise training. Its determination might therefore allow us to ''pick up'' subtle or early effects of training (and possibly also of detraining) on skeletal muscle oxidative metabolism. The present study demonstrates that this also applies to the $\dot{V}O_2$ off-kinetics, a variable often neglected by exercise physiologists, but nonetheless one that allows functional evaluation of the rate of adjustment of skeletal muscle oxidative metabolism to the regulating signal represented by the decreased skeletal muscle PCr stores, once allowance is made for the replenishment of the muscle O_2 stores at the very onset of recovery. PCr splitting is thought to be involved in the regulation of skeletal muscle oxidative metabolism, both during exercise transients and steady-states [5, 6, 17, 20, 23]. Analysis of $\dot{V}O_2$ off-kinetics offers the additional advantage (compared to the $\dot{V}O_2$ on-) that breath-by-breath $\dot{V}O_2$ ''noise'' or variability during the off-phase is significantly lower, thereby allowing greater reliability in parameter estimation.

In the present study the effects of exercise training on the $\dot{V}O_2$ on- and off-kinetics showed a plateau after ≅1 month of training (i.e., at T30). At that point, the $\dot{V}O_2$ on- and off- parameters of the middle-aged subjects were not different from those obtained from the reference group of ≅20-year-old untrained subjects. As is well known, a faster adjustment of oxidative metabolism during increases of power output reduces the need for PCr hydrolysis and substrate-level phosphorylation, with less disturbance of cellular and organ homeostasis, less muscle fatigue and improved exercise tolerance. The observation that, after ≅1 month of training, middle-aged subjects show $\dot{V}O_2$ on-kinetics that are not different from those observed in 20-year-old untrained subjects appears particularly interesting, since it demonstrates that even a short training period is enough, in middle-aged subjects, to counterbalance the well described slowing of $\dot{V}O_2$ on-kinetics associated with aging [3, 4, 8, 9]. Further studies could clarify if this also applies to older subjects. The results of the present study appear to agree with those obtained by Chilibeck et al. [8], according to whom a slowing of $\dot{V}O_2$ on-kinetics with age is evident during forms of exercise (such as cycling) to which the subjects are not accustomed, whereas the slowing is much less evident during other types of exercise (such as walking) which are usually performed during activities of everyday life. Our data also underscore the need to carefully standardize for training status when analyzing $\dot{V}O_2$ on-kinetics in subjects of different age groups.

As mentioned above, $t_{1/2}$ HR on- decreased as a function of training. This may indicate that the faster $\dot{V}O_2$ on-kinetics with training could be attributed, at least in part, to a faster O_2 delivery to the exercising muscles. However, correlation analysis between $t_{1/2}$ HR on- and τ_2 $\dot{V}O_2$ on-kinetics indicate a r^2 of 0.10, showing that only about 10% of τ_2 $\dot{V}O_2$ on-kinetics variability could be explained in terms of $t_{1/2}$ HR on-. Also against the hypothesis of a cause-and-effect relationship between the faster HR on- and faster $\dot{V}O_2$ on-kinetics is the observation that, in the present study, from T30 onward $\dot{V}O_2$ on-kinetics in middle-aged subjects was not different than in YG, despite the significantly higher $t_{1/2}$ HR on- in the former group. It must also be remembered that HR on-kinetics is only a very ''distant'' and indirect estimation of the relevant variable, i.e., the on-kinetics of O_2 delivery to skeletal muscle cells. For example, Shoemaker et al. [22] observed that the blood flow response to exercise depends not only on changes in cardiac output ($\dot{Q}$), but also on the effects of the muscle pump and various vasoactive metabolites and hormones produced both locally and systemically, all of which affect femoral artery vascular conductance and O_2 delivery, and all of which could in theory be affected by exercise training independently of the effects on $\dot{Q}$. Although Green et al. [15] reported that, in young subjects, the capillary-to-fiber area ratio increased after only 10–12 days of training, and Coggan et al. [10] observed increased capillary density in old subjects after several months of training, the consequences of a short-term training protocol on muscle microcirculation in middle-aged and elderly subjects are not known. Moreover, Chilibeck et al. [9] did not find a significant correlation between capillary-to-fiber number ratio and pulmonary $\dot{V}O_2$ kinetics in old subjects.

Another possible limitation to $\dot{V}O_2$ on-kinetics in the middle-aged subjects could be reduced arterial O_2 partial pressure (PaO_2) [16]. We did not measure PaO_2 in the

present study. In our subjects, however, SaO_2 values at rest and during exercise were not different in middle-aged subjects compared to YG. Moreover, previous studies have demonstrated that hyperoxic breathing in old subjects does not determine a faster $\dot{V}O_2$ on-kinetics [4].

Since during low-intensity exercise $\dot{V}O_2$ on-kinetics is thought to be determined mainly by intrinsic ''metabolic inertia'' of skeletal muscle oxidative metabolism [7, 13, 23], the positive effects of exercise training on $\dot{V}O_2$ on-kinetics, as observed in the present study, can be ascribed to some reduction of this inertia (level of metabolic controllers, enzyme activation, etc.). On the basis of our results, however, no further insight into these mechanisms can be obtained.

Interesting observations may also be derived from the analysis of other physiological variables determined at submaximal loads. To begin with, $\dot{V}O_{290W}$ was unaffected by training, and the values obtained from middle-aged subjects were not different from those obtained from YG. This indicates that the efficiency of exercise (as estimated by the ratio between $\dot{V}O_2$ and $\dot{w}$) at submaximal loads was independent of age, and was unaffected by training in middle-aged subjects. HR_{90W} and $(HR \cdot SBP)_{90W}$ in middle-aged subjects showed a tendency to decrease with training, and such decreases became statistically significant after only 15 days of training. Since (HR·SBP) allows an indirect evaluation of cardiac work, the observation of a decreased $(HR \cdot SBP)_{90W}$ after only 2 weeks of exercise training has obvious interest and important implications for the prevention of cardiovascular diseases, particularly since it relates to submaximal workloads, i.e., to exercise intensities normally encountered during activities of everyday life.

The training program also had effects on ''peak'' physiological variables determined during the incremental exercise. These effects, however, occurred later during the training program compared to those related to submaximal loads. $\dot{V}O_{2peak}$ in middle-aged subjects tended to increase with training, but differences compared to T0 were significant only after 2 months. However, at the end of the training program, $\dot{V}O_{2peak}$ in middle-aged subjects was still ≅17% lower than the values observed in YG. Training did not have any significant effect on HR_{peak}, and the values obtained from middle-aged subjects were significantly lower than those obtained from YG. This observation confirms that the well-known reduction of maximal HR with aging is not affected by exercise training. Other cardiovascular variables, such as $(HR \cdot SBP)_{peak}$, were also unaffected by exercise training. $[La]_{bpeak}$ increased as a function of training in middle-aged subjects, and after 2 months of training the values were not significantly different compared to those of YG.

In accordance with the American College of Sports Medicine guidelines [1] the recommended intensity for exercise training in older adults is 50–70% of the ''HR reserve'' determined during aerobic exercise. In this respect, our exercise intensity was at the low-end of the recommended spectrum (i.e., subjects trained at ~50% of their HR reserve, determined serially during the training program). The results of the present study demonstrate that even a relatively low training intensity (which is usually adopted for middle-aged and old subjects in most health clubs, such as that where these training sessions were carried out) can elicit significant and early modifications of relevant physiological variables determined during submaximal exercise and, to a lesser extent, some changes of physiological variables determined during ''peak'' exercise.

As for resistance (strength) training, the ACSM guidelines [1] suggest that the subjects perform one set of 8–10 different exercises involving all the major muscle groups, and 8–12 repetitions of each exercise is executed in one set. The resistance training program adopted in the present study agreed fully with these guidelines. To our knowledge, no previous studies on the effects of resistance training on $\dot{V}O_2$ on- and off-kinetics have been performed. Thus, we cannot speculate on how resistance training ''per se'' could have affected these variables in the present study. According to a recent position paper by the American Heart Association [19], however, a ''complementary resistance training program, when appropriately prescribed and supervised, has favorable effects on muscular strength and endurance, cardiovascular function, metabolism, coronary risk factors, …''. Thus, it appears that the resistance training program could have contributed to the improved exercise performance, particularly during submaximal exercise, described in the present study.

Conclusion

In previously untrained middle-aged (≅50 year-old) subjects, a standard training program adopted in health clubs very quickly leads to (i.e., after 1–2 weeks) adaptations of physiological variables determined during submaximal exercises; for example, faster $\dot{V}O_2$ on- and off-kinetics, faster HR on-kinetics, and lower HR and (HR·SBP) at steady-state. These adaptations occur much earlier than others described at peak workload (such as increases in $\dot{V}O_{2peak}$), may significantly and positively affect exercise tolerance, and might also have important implications in the prevention of cardiovascular diseases. Also in middle-aged subjects $\dot{V}O_2$ on- and off-kinetics appear particularly sensitive to exercise training. Evaluation of the kinetics might allow us to ''pick up'' subtle or early effects of training on skeletal muscle oxidative metabolism.

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References

1. ACSM (1998) Position stand on the recommended quantity and quality of exercise for developing and maintaining cardiorespiratory and muscular fitness, and flexibility in adults. Med Sci Sports Exerc 30:975–991
2. Babcock MA, Paterson DH, Cunningham DA (1994) Effects of aerobic endurance training on gas exchange kinetics of older men. Med Sci Sports Exerc 26:447–452
3. Babcock MA, Paterson DH, Cunningham DA, Dickinson JR (1994) Exercise on-transient gas exchange kinetics are slowed as a function of age. Med Sci Sports Exerc 26:440–446
4. Bell C, Paterson DH, Kowalchuk JM, Cunningham DA (1999) Oxygen uptake kinetics of older humans are slowed with age but are unaffected by hyperoxia. Exp Physiol 84:747–759
5. Binzoni T, Cerretelli P (1994) A bioenergetic approach to ''transfer function'' of the human skeletal muscle. J Appl Physiol 77:1784–1789
6. Cerretelli P, di Prampero PE (1987) Gas exchange in exercise. In: Handbook of physiology, section 3, vol. IV. The respiratory system. Gas exchange. American Physioogical Society, Bethesda, Md, pp 297–339
7. Cerretelli P, Rennie DW, Pendergast DR (1980) Kinetics of metabolic transients during exercise. In: Cerretelli P, Whipp BJ (eds) Exercise bioenergetics and gas exchange. Elsevier, Amsterdam, pp 187–209
8. Chilibeck PD, Paterson DH, Smith WDF, Cunningham DA (1996) Cardiorespiratory kinetics during exercises of different muscle groups and mass in old and young. J Appl Physiol 81:1388–1394
9. Chilibeck PD, Paterson DH, Cunningham DA, Taylor AW, Noble EG (1997) Muscle capillarization, O_2 diffusion distance, and $\dot{V}O_2$ kinetics in old and young individuals. J Appl Physiol 82:63–69
10. Coggan AR, Spina RJ, King DS, Rogers MA, Brown M, Nemeth PM, Holloszy JO (1992) Skeletal muscle adaptations to endurance training in 60- to 70-yr-old men and women. J Appl Physiol 72:1780–1786
11. Cunningham DA, Himann JE, Paterson DH, Dickinson JR (1992) Gas exchange dynamics with sinusoidal work in young and elderly women. Respir Physiol 91:43–56
12. Gaeeser GA, Poole DC (1996) The slow component of oxygen uptake kinetics in humans. In: Holloszy JO (ed) Exercise and sport sciences reviews, vol. 24. Williams and Wilkins, Baltimore, pp 35–71
13. Grassi B (2001) Regulation of oxygen consumption at exercise onset: is it really controversial? Exerc Sport Sci Rev 29:134–138
14. Grassi B, Poole DC, Richardson RS, Knight DR, Erickson BK, Wagner PD (1996) Muscle O_2 uptake kinetics: implications for metabolic control. J Appl Physiol 80:988–998
15. Green HJ, Jones S, Ball-Burnett ME, Smith D, Livesey J, Farrance BW (1991) Early muscular and metabolic adaptations to prolonged exercise training in humans. J Appl Physiol 70:2032–2038
16. Levitzky MG (1984) Effects of aging on the respiratory system. Physiologist 27:102–107
17. Meyer RA, Foley JM (1996) Cellular processes integrating the metabolic response to exercise. In: Handbook of physiology, Section 12. Exercise: regulation and integration of multiple systems. Oxford University Press, Oxford, pp 842–869
18. Phillips SM, Green HJ, MacDonald MJ, Hughson RL (1995) Progressive effect of endurance training on $\dot{V}O_2$ kinetics at the onset of submaximal exercise. J Appl Physiol 79:1914–1920
19. Pollock ML, Franklin BA, Balady GJ, Chaitman BL, Fleg JL, Fletcher B, Limacher M, Pi–a IL, Stein RA, Williams M, Bazzarre T (2000) Resistance exercise in individuals with and without cardiovascular disease. Benefits, rationale, safety and prescription. An advisory from the Committee on Exercise, Rehabilitation and Prevention, Council on Clinical Cardiology, American Heart Association. Circulation 101:828–833
20. Rossiter HB, Ward SA, Doyle VL, Howe FA, Griffiths JR, Whipp BJ (1999) Influence from pulmonary O_2 uptake with respect to intramuscular [phosphocreatine] kinetics during moderate exercise in human. J Physiol (Lond) 518:921–932
21. Rowell LB (1993) Human cardiovascular control. Oxford University Press, New York
22. Shoemaker JK, Phillips SM, Green HJ, Hughson RL (1996) Faster femoral artery blood velocity kinetics at the onset of exercise following short-term training. Cardiovasc Res 31:278–286
23. Whipp BJ, Mahler M (1980) Dynamics of pulmonary gas exchange during exercise. In: West JB (ed) Pulmonary gas exchange, vol. I. Oxford University Press, Oxford, pp 361–370
24. Whipp BJ, Ward SA, Lamarra N, Davis JA, Wasserman K (1982) Parameters of ventilatory and gas exchange dynamics during exercise. J Appl Physiol 52:1506–1513

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