# Age and Activity Status Affect Muscle Reoxygenation Time after Maximal Cycling Exercise

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#### ABSTRACT

ICHIMURA S., N. MURASE, T. OSADA, R. KIME, T. HOMMA, C. UEDA, T. NAGASAWA, M. MOTOBE, T. HAMAOKA, and T. KATSUMURA. Age and Activity Status Affect Muscle Reoxygenation Time after Maximal Cycling Exercise. Med. Sci. Sports Exerc., Vol. 38, No. 7, pp. 1277-1281, 2006. Purpose: The purpose of this study was to determine the interaction of age and habitual physical activity on recovery time of muscle oxygenation following maximal cycling exercise (CycEXmax). Methods: Twelve sedentary middle-aged (50  $\pm$  6), 13 sedentary elderly (66  $\pm$  3), 13 active middle-aged (53  $\pm$  5), and 20 active elderly (67  $\pm$  5) women participated in this study. We evaluated the peak pulmonary oxygen uptake (VO2peak) during CycEXmax and the half-recovery time of muscle oxygenation (T1/2reoxy time) using near-infrared spectroscopy at the vastus lateralis (VL) during the recovery phase after CycEXmax. Results: T1/2reoxy time was significantly greater in the elderly subjects than in the middle-aged subjects in both sedentary (P < 0.05) and active groups (P < 0.01). T1/2reoxy time of the active group was lower (P < 0.01) than that of the sedentary group regardless of age. Age was significantly correlated to T1/2reoxy time in both sedentary and active groups (in both sedentary and active groups: P < 0.01). The slope of T1/2reoxy time against age in the sedentary group was significantly greater (VL: P < 0.05) than that of the active group. VO<sub>2peak</sub> showed significant inverse correlation with T1/2reoxy time at the VL in both sedentary and active groups. The slope of VO<sub>2peak</sub> against T1/2reoxy time showed no significant differences between middle-aged and elderly subjects. Conclusion: The results of this study suggest that T1/2reoxy time was prolonged with aging, regardless of habitual physical activity levels. However, habitual physical activity may prevent the age-related prolongation in T1/2reoxy time after CycEXmax. VO<sub>2peak</sub> appears to be one of the major factors determining T1/2reoxy time, not age. Key Words: AGING, HABITUAL PHYSICAL ACTIVITY, SEDENTARY WOMEN, ACTIVE WOMEN, NEAR-INFRARED SPECTROSCOPY, VO2peak

uscle volume (5), muscle oxidative capacity (5,13), and blood flow to working muscle (1,21) decline with aging. Recently, it has been shown that muscle oxidative capacity and muscle blood flow after exercise to fatigue is higher in trained elderly people than in sedentary peers (4,12). Endurance training has restored or improved the muscle oxidative capacity and blood flow to working muscles even in elderly people (1,3). The age/ activity-related changes in these muscle functions contribute to the changes in systemic aerobic capacity.

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Recently, muscle oxygenation during exercise has been measured noninvasively by near-infrared spectroscopy (15). Muscle oxygenation measured by near-infrared spectroscopy reflects the balance of O<sub>2</sub> delivery to working muscles and muscle  $O_2$  consumption in capillary beds (15). Recovery time of muscle oxygenation following the completion of exercise is used as an index of deficits of O<sub>2</sub> delivery to the muscle in relation to  $O_2$  demand of the working muscle (2). It is thought that recovery time of muscle oxygenation is an indicator for evaluating muscle aerobic function. In previous studies, recovery time of muscle oxygenation after submaximal exercise has successfully been used to noninvasively and qualitatively assess muscle oxygenation kinetics in patients with peripheral vascular disease (14). These data suggested that recovery time of muscle oxygenation after submaximal exercise is considered mainly dependent on O2 delivery to muscles (10,14).

Kutsuzawa et al. (10) demonstrated that recovery rate of muscle oxygenation rate after submaximal handgrip exercise was delayed with increased age. Furthermore, Hamaoka et al. (8) suggested that recovery time of muscle oxygenation after

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submaximal cycling exercise was shorter in athletes than in their sedentary peers. To our knowledge, however, interaction of age and habitual physical activity on recovery time of muscle oxygenation after maximal cycling exercise (CycEXmax) has not been established. Further, the relation between systemic aerobic capacity and recovery time of muscle oxygenation after CycExmax is not clear. Therefore, the purpose of this study was to determine the interaction of age and habitual physical activity on recovery time of muscle oxygenation and the relation of systemic aerobic capacity and recovery time of muscle oxygenation following CycEXmax. We hypothesized that habitual physical activity may attenuate the age-related prolongation in recovery time of muscle oxygenation and that systemic aerobic capacity is related to recovery time of muscle oxygenation immediately after CycEXmax.

## METHODS

**Subjects.** Thirty-three healthy active women (40–67 yr old) and 25 healthy sedentary women (40-76 yr old) participated in this study. For at least the previous 6 months, subjects were either sedentary (no regular physical activity) or active (light- to moderate-intensity water exercise once or twice per week). In addition, these subjects were divided into sedentary middle-aged (N = 12), sedentary elderly (N = 13), active middle-aged (N = 13), and active elderly (N = 20)subgroups. All subjects were healthy and free of hypertension, diabetes, and cardiovascular disease as assessed by their medical history. Additionally, subjects currently on pharmacotherapies with known vascular effects were excluded. All subjects gave their written informed consent using the Tokyo Medical University Hospital institutional review board-approved forms to participate. All procedures were performed according to the Declaration of Helsinki.

**Exercise test.** Subjects performed a maximal aerobic exercise test using a bicycle ergometer (Corval Load). The physician determined an exercise load of 10 or 15 W·min<sup>-1</sup> by ramp method in consideration of the subject's age and weight. Pedal frequency was 60 rpm. The exercise test end point was determined as the point when the rhythm of the metronome and the leg movement were not synchronized.

**Measurements.** During the exercise test, pulmonary oxygen uptake ( $\dot{VO}_{2peak}$  AE-300 Minato, JAPAN) using breath-by-breath basis, heart rate, and blood pressure were measured. Additionally, oxygenated hemoglobin/myoglobin (Hb/MbO<sub>2</sub>) kinetics of the vastus lateralis (VL) were measured at the position two thirds of the way from the superior end patella to greater trochanter of the thigh using near-infrared continuous-wave spectroscopy (NIRcws; HEO-200 Omron, JAPAN). During the recovery phase immediately after exhaustion, the half-recovery time of Hb/MbO<sub>2</sub> (T1/2reoxy time) (2,14) was measured. T1/2reoxy time was determined as the time for 50% reoxygenation of Hb/MbO<sub>2</sub> from the exhaustion level to the peak level. We showed the calculation method of T1/2reoxy time in Figure 1.

Hb/MbO<sub>2</sub> measured by NIRcws can be described by the mathematical equation using Fick's Law (22). Hb/MbO<sub>2</sub>,



FIGURE 1—Typical kinetics of Hb/MbO<sub>2</sub> and method of evaluating T1/2reoxy time. Method to evaluate T1/2reoxy time, the time required to reach the value halfway between the Hb/MbO<sub>2</sub> level immediately after exercise and that at peak hyperemia during recovery.

measured by NIRcws, reflects the balance of  $O_2$  delivery to working muscles and muscle  $O_2$  consumption and indicates the  $O_2$  content in the capillary bed (15). Thus, Hb/MbO<sub>2</sub> measured by NIRcws can be expressed with the following simple mathematical equation:

$$Hb/MbO_2(CvO_2) = CaO_2 - VO_2 mus/O_2$$
 delivery

where CaO<sub>2</sub> is the O<sub>2</sub> content at the beginning of the capillary and CvO<sub>2</sub> is the O<sub>2</sub> content at the venous side of the capillary (measurement site by NIRcws).  $\dot{V}O_2$ mus and O<sub>2</sub> delivery are muscle O<sub>2</sub> consumption and O<sub>2</sub> delivery to muscle, respectively. Therefore, faster Hb/MbO<sub>2</sub> recovery measured by NIRcws during recovery from exercise should be the consequence of a faster recovery in  $\dot{V}O_2$ mus, or a higher O<sub>2</sub> delivery and/or slower recovery in  $\dot{V}O_2$ mus or a lower O<sub>2</sub> delivery and/or faster recovery in  $\dot{V}O_2$ mus or a lower O<sub>2</sub> delivery and/or faster recovery in O<sub>2</sub> delivery following exercise should elicit the delay of T1/2reoxy time during the recovery phase following exercise. Thus, T1/2reoxy time was used as an indicator for muscle aerobic function (2,10,14,22).

The theoretical basis and principle of the NIRcws device were discussed extensively in previous studies (15). In this study, we used the NIRcws device with wavelengths of 760 and 850 nm. This device consists of a probe and a computerized control unit, and the probe consists of a light source and an optical detector. The distance between the light source and optical detector was 3 cm. The data sampling rate was 2 Hz.

**Statistics.** Descriptive data were presented as the mean and SD for each group. A two-way analysis of variance (ANOVA) with *post hoc* comparisons was used to assess the effects of age and habitual physical activity on the measured variables. Linear correlation analysis or exponential curve correlation analysis was used to assess the relationship between the variables. When a significant exponential curve correlation was found, a logarithmic conversion was executed on the variables to examine an analysis of covariance (ANCOVA). ANCOVA was performed to test the difference between the middle-aged and elderly subjects or between active and sedentary groups in terms of dependent variable adjusted by independent variable when a significant linear correlation was found between the variables. Statistical significance was set *a priori* at P < 0.05.

## RESULTS

**Comparison between middle-aged and elderly subjects.** Peak workload and  $\dot{VO}_{2peak}$  were significantly lower (P < 0.01) in the elderly subjects than those of the middle-aged subjects in the active group, but not significantly different between ages in the sedentary group. HRpeak was significantly lower in the elderly subjects compared with the middle-aged subjects in both sedentary and active groups (sedentary group: P < 0.05; active group: P < 0.01). T1/2reoxy time was significantly greater in the elderly subjects compared with the middle-aged subjects within the same physical activity group (sedentary groups: P < 0.05; active group: P < 0.01).

**Comparison between sedentary and active groups.** Peak workload in the sedentary group was significantly lower than that of the active group only in the middle-aged subjects (P < 0.01). The differences in habitual physical activity did not influence HRpeak within subjects of the same age.  $VO_{2peak}$  in the sedentary group was significantly lower compared with the active group, regardless of age (both elderly and middle-aged subjects: P < 0.01). The sedentary group showed a significantly greater T1/2reoxy time than the active group of the same generation (both elderly and middle-aged subjects: P < 0.01).

The relationship between age and t1/2 reoxy time. Figure 2 shows the relationship between age and T1/2 reoxy time. The T1/2 reoxy time was positively related to age in both sedentary (r = 0.663, P < 0.001) and active (r = 0.498, P < 0.01) groups. The slope of T1/2 reoxy time against age



FIGURE 2—The relationship between age and T1/2reoxy time in sedentary and active groups. The slope of age-related prolongation in T1/2reoxy time was significantly lower in the active group than in the sedentary group (\* P < 0.05 for both VL and LG).

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FIGURE 3—The relationship between  $\dot{VO}_{2peak}$  and T1/2reoxy time in middle-aged and elderly subjects. The logarithm slope of  $\dot{VO}_{2peak}$  and T1/2reoxy time showed no significant differences between middle-aged and elderly subjects.

was significantly greater in the sedentary group (0.94 s·yr<sup>-1</sup>) than in the active group (0.44 s·yr<sup>-1</sup>) (P < 0.05).

The relationship between  $\dot{VO}_{2peak}$  and T1/2reoxy time. Significant exponential curve correlation was found between  $\dot{VO}_{2peak}$  and T1/2reoxy time. Figure 3 shows the logarithmic graph of relationship between  $\dot{VO}_{2peak}$  and T1/2reoxy time. There was an inverse correlation between  $\dot{VO}_{2peak}$  and T1/2reoxy time in both middle-aged (r = -0.835, P < 0.001) and elderly subjects (r = -0.852, P < 0.001). The logarithm slope of  $\dot{VO}_{2peak}$  and T1/2reoxy time showed no significant differences between middle-aged and elderly subjects (Fig. 3).

## DISCUSSION

Primary findings of this cross-sectional study are as follows. First, T1/2reoxy time was significantly related to age, and T1/ 2reoxy time was greater in the elderly subjects than in the middle-aged subjects, regardless of habitual physical activity levels. Second, T1/2reoxy time was lower in the active group than in the sedentary group in both middle-aged and elderly subjects (Table 1). The slope of T1/2reoxy time against age was significantly lower in the active group than in the sedentary group (Fig. 2). Third, VO<sub>2peak</sub> and T1/2reoxy time was inversely correlated, and the logarithm slope of VO<sub>2peak</sub> and T1/2reoxy time showed no significant differences between middle-aged and elderly subjects (Fig. 3). From these findings, we suggest that habitual physical activity may attenuate age-related prolongation in T1/2reoxy time in working muscles. VO<sub>2peak</sub> appears to be one of the major factors determining T1/2reoxy time, not age.

**Age-related prolongation in T1/2reoxy time.** In this study, there was age-related prolongation in T1/2reoxy time in both the sedentary and active groups, respectively. This result was in agreement with the findings of a previous study (10) in which recovery rate of muscle oxygenation measured in the forearm flexor muscles was not in agreement

TABLE 1. Influences of age	and physical activity	status on oxygen	consumption, heart rate,	, workload, and 1	T1/2reoxy time during peak e	exercise
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	A	ctive	Sedentary		
	Middle-Aged	Elderly	Middle-Aged	Elderly	
VO <sub>2peak</sub> (mL⋅kg <sup>-1</sup> ⋅min <sup>-1</sup> )	$33.1~\pm~3.9$	$24.7 \pm 4.0**$	$21.9 \pm 3.7^{\#\#}$	$19.7 \pm 4.4^{\#\#}$	
HRpeak (bpm)	$163.5 \pm 9.6$	$149.9 \pm 13.5 * *$	157.3 ± 11.5	$145.2 \pm 18.4^{*}$	
Peak load (W)	143.0 ± 21.8	101.1 ± 22.6**	$108.9 \pm 34.8^{\#\#}$	$88.5 \pm 24.9$	
$\dot{V}O_{2} \cdot W^{-1}$ ) (mL·kg <sup>-1</sup> ·min <sup>-1</sup> ·W <sup>-1</sup> )	$0.23 \pm 0.05$	$0.25 \pm 0.05$	$0.21 \pm 0.04$	$0.23 \pm 0.04$	
T1/2reoxy time (s)	$22.5\pm3.3$	$29.6~\pm~8.9^{\star}$	$35.7\pm9.0^{\#\#}$	$45.7\pm13.6^{*,\#\#}$	

Values are mean  $\pm$  SD.

\* P < 0.05 vs middle-aged group of same physical activity status; \*\* P < 0.01 vs middle-aged group of the same physical activity status; ## P < 0.01 vs active group of age-matched group.

with the result for which half-recovery time of muscle oxygenation was measured in vastus lateralis after exhaustive cycling exercise (6). Costes et al. (6) reported that there was no significant difference in recovery time of muscle oxygenation after exhaustive cycling exercise in young (33.5  $\pm$ 17.5 s) and elderly (28.2  $\pm$  10.5 s) subjects. Costes's elderly subjects were engaged in recreational physical activities, and young subjects ranged from sedentary to moderately trained. Thus, physical activity status between young and elderly subjects was inconsistent. In addition, the variation of halfrecovery time of muscle oxygenation appears larger in the young subjects than in the elderly subjects partly because physical activity status among the young subjects was varied. A wide variety of physical activity status may cause a discrepancy of age-related change in T1/2reoxy time between this study and the previous study (6). Therefore, physical activity status would be expected to play an important role in determining the extent to which muscle aerobic function changes with increased age.

It is reported that advanced age impairs vascular function (7,9) and peak blood flow to the working muscles (21). Muscle oxidative capacity declines with increased age (5,13). It is thought that because muscle oxygenation kinetics measured by NIRcws after exercise are controlled by recovery speed of muscle O<sub>2</sub> consumption and O<sub>2</sub> delivery to muscle after exercise, a slower recovery of VO<sub>2</sub>mus and/ or lower O<sub>2</sub> delivery after exercise with increased age results in the prolongation of T1/2reoxy time during recovery following exercise. The results of previous studies having reported age-related decline in blood flow to muscle, peripheral vascular function, and structure, and muscle oxidative capacity may support the age-related prolongation in T1/ 2reoxy time for the sedentary and active groups found in this study. We suggest that age-related prolongation in T1/2reoxy time in working muscles reflects the age-related decline in muscle aerobic function.

Activity-related change in T1/2reoxy time. T1/ 2reoxy time was significantly lower in the active group than in the sedentary group for both middle-aged and elderly subjects. The slope of T1/2reoxy time against age was significantly smaller in the active group than in the sedentary group. In a previous study, T1/2reoxy time in working muscles after sub-maximal cycling exercise was lower in young athletes than in their sedentary peers (8). From these results, we suggest that habitual physical activity can attenuate the age-related prolongation in T1/2reoxy time in working muscles. In cross-sectional studies, peak cardiac output (19), blood flow to working muscles (1,20) during peak exercise, and muscle oxidative capacity (4) were greater in active subjects than in sedentary subjects in both young and elderly subjects. It has been demonstrated that the blood flow to working muscles (1) during peak exercise, vascular functions (7,23), and muscle oxidative capacity (3) improve when physical activity was sufficient. Thus, the results of previous studies, having reported that habitual physical activity can prevent the age-related decline in peak blood flow and muscle oxidative capacity, support the attenuated agerelated prolongation in T1/2reoxy time in working muscles in the active group found in this study.

Relationship VO<sub>2peak</sub> and T1/2reoxy time. In this study, VO<sub>2peak</sub> was significantly related to the T1/2reoxy time in both middle-aged and elderly subjects. Previous studies have reported that high  $\dot{V}O_{2peak}$  was associated with prolongation of the vasodilation duration after arterial occlusion with calf exercise (9), elevated peak vascular conductance after exercise (12), and a faster recovery of PCr measured by <sup>31</sup>P magnetic resonance spectroscopy (<sup>31</sup>P-MRS) after exercise (11). Muscle microvascular oxygen pressure (PO<sub>2</sub>), a reflection of the balance between  $O_2$ delivery to working muscles and muscle O<sub>2</sub> consumption, can be a similar indicator to Hb/MbO<sub>2</sub> measured by NIRcws. It was reported that PO<sub>2</sub> recovery following muscle contractions was faster in high-oxidative-capacity muscle (16-18). Results of these previous studies support that T1/ 2reoxy tine after exercise evaluated by NIRcws can be an index of muscle aerobic function, including muscle O<sub>2</sub> utilization and muscle O<sub>2</sub> delivery.

The slope between  $\dot{V}O_{2peak}$  and T1/2reoxy time and the intercept to the y-axis showed no significant differences between middle-aged and elderly subjects. These results do not distinguish whether an increase in muscle aerobic function improves the systemic aerobic capacity, or whether the improvements of muscle aerobic function and systemic aerobic capacity occur simultaneously. However, the results do indicate that the systemic aerobic capacity is related to muscle aerobic function evaluated by T1/2reoxy time, and the relationship between systemic aerobic capacity and T1/2reoxy time is similar between middle-aged and elderly women. Thus,  $\dot{V}O_{2peak}$ , but not age, may be one of the major factors determining T1/2reoxy time.

**Limitations.** A limitation of this study is its crosssectional design. We did not monitor or control the daily physical activity level and the intensity during water exercise in individuals of the active group, although individuals of the active group had participated in the same water exercise course. In this study,  $\dot{V}O_{2peak}$  and peak load showed significant differences between middle-aged and elderly subjects in the active group but not in the sedentary group. Thus, it is speculated that the difference in physical activity level between the middle-aged subjects and elderly subjects in the active group may be larger than that of the middle-aged subjects and elderly subjects and elderly subjects in the sedentary group. Thus, environmental factors including daily physical activity level as well as genetic or other constitutional factors may have influenced this cross-sectional study. To further understand the effect of habitual physical activity on age-

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in working muscles, it is important to lessen the variation of physical activity level within a similar physical activity status, and longitudinal studies are needed.

In conclusion, habitual physical activity may attenuate age-related prolongation in T1/2reoxy time in working muscles.  $\dot{VO}_{2peak}$  appears to be one of the major factors determining T1/2reoxy time, not age.

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